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PRINCIPLES OF PESTICIDE TOXICOLOGY

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This book is intended for toxicologists, hygienists and all those responsible for evaluation and control of harmful effects of chemicals to human health and the environment. It could also be useful in postgraduate training of specialists in preventive toxicology.

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PREFACE TO THE ENGLISH EDITION

The development of pesticides, chemicals capable of protecting plants from pests, diseases and weeds, is an extraordinary achievement of modern science. Their capacity to increase crop yields and decrease production losses has led to continuous expansion in pesticide production and application all over the world. In 1975, global pesticide production was estimated at 1.6 million metric tons of active ingredient. Since 1975, production of pesticides has grown by 10 to 15 percent per annum. At this growth rate pesticide manufacture world-wide should attain 2.5—2.7 million tons of active ingredient by the year 2000 (Unanyants, 1981).

The tremendous benefits of pesticides for food production must be weighed, on the other hand, against some specific properties making them a potential risk to human health. These include their high biological activity and the ability to persist in the environment and to accumulate in food. The distribution of pesticides over vast areas leads to contamination of soil, water and biota, and consequently to human exposure, not least by the entry of these substances into the human body. The properties of each pest control product demand careful scrutiny of its possible effects so that scientists and legislators can assess it hygienically and set standards and regulations for its use which will assure maximum crop yields and minimum hazard to man and the environment. The toxicological properties of pesticides and the problems they present are, currently, a compelling direct concern for numerous scientists and health officials throughout the world.

This book discusses the goals and methods of modern pesticide toxicology and describes quantitative criteria needed to assess their toxicity and hazard to man. It emphasizes the cumulative effect of long-term exposure to pesticides and the adaptive response of the organism. Furthermore, it considers the biochemical mechanisms responsible for the action of pesticides, and the effects of combined, concomitant and joint pesticide exposure (the latter two terms are adopted in Soviet scientific literature) with other chemicals or with physical environmental factors. Some of the topics covered bear on clinical aspects and diagnosis of pesticide poisonings and current therapeutic techniques. There is extensive coverage of potential ad-

verse long-term effects of pesticide exposure, in particular their carcinogenic, mutagenic, and teratogenic effects. Finally, hygienic standards that apply to pesticide concentrations in ambient air, food, livestock forage, water and soil are presented and discussed.

The necessity to provide a compact text has prevented the author from reviewing with sufficient scope the vast amount of information on all aspects of pesticide toxicology. Rather, the aim here is to highlight the experience of Soviet toxicology in the area of pesticide research. In this vein, the system for hygienic and toxicological pesticide assessment accepted in the USSR is summarized briefly.

The reader will find the book a ready source of data on the specific methodological approaches used by Soviet authors to set hygienic standards for pesticide concentrations in environmental media, biota, food, etc., as well as specific data on current standards for pesticides effective in the USSR.

The author hopes that the contents of this volume will be of use to those with an interest in the problems of pesticide toxicology and welcomes comments and criticisms.

Yu. Kagan.

The nomenclature of pesticides used in this volume conforms to the 1979 Registry of Toxic Effects of Chemical Substances; NIOSH USA, Publication No. 80-111.

References to publications in the text provide information sufficient to locate the full bibliographic citation in the alphabetical listing of "References", that is, author and date.

The terminology follows, as far as possible, that of the English-Russian Glossary of Selected Terms in Preventive Toxicology (Interim Document; Centre of International Projects, Moscow, 1982).

OBJECTIVES OF PESTICIDE TOXICOLOGY. QUANTITATIVE CRITERIA OF THEIR TOXICITY AND HAZARD

The Central Committee of the Communist Party of the Soviet Union and the USSR Council of Ministers recognize the special importance of boosting further the production and application of chemicals used to protect plants against pests. To achieve this, it was found necessary to merge specialized agrochemical services for agriculture into a single system operating on the country-wide scale. Thus the "Selkhozchimiya" Research and Operational Association was created as a new division of the USSR Ministry of Agriculture responsible for agrochemical support of agricultural production. This and other similar measures are designed to expand and rationalize the application of pesticides, the chemicals affording protection to plants.

Pesticide toxicology is the field of hygienic (preventive) toxicology studying the interactions of pesticides with body organs and systems in humans and animals. Its principal purpose is to establish potential and actual risk from the chemicals used for control of crop pests in agriculture and the new agents proposed for transfer into farming practices. The risk, or hazard of substances is understood not merely as their ability to cause acute and chronic poisoning, but also as their potential for selective influence on body organs and physiological systems, macromolecules, and particular segments of metabolism and for modifying the reactivity of the exposed organism. This selective potential resides in blastomogenic, particularly carcinogenic, as well as embryotoxic, teratogenic, and mutagenic properties of these substances.

While formerly toxicology was concerned primarily, if not exclusively, with the toxic properties of chemicals, its scope has considerably broadened today and it has evolved into the science of chemically induced pathology, or the disease state of chemical aetiology. The basic goals of pesticide toxicology are:

1. Toxicity assessment of the chemicals proposed for registration in agricultural use, by their different routes of entry into the body (orally, through skin, or by inhalation); evaluation of their cumulative effects; development of data needed for predicting acute and chronic intoxications.

2. Study of toxicokinetics and toxicodynamics of these substances; detection of the most sensitive tests so as to identify their threshold doses and concentrations; establishment of the dose — response, time — response, and dose — time relationships; estimation of the thresholds of acute and chronic effects.

3. Development of diagnostic techniques for acute and chronic intoxications using data on the mechanism of action of the causatory chemical; formulation of proposals for effective therapeutic remedies (specific, if possible) in treating the poisonings.

4. Recognition of the role of the chemical being tested in the origin and progression of various disease processes; investigation of long-term sequelae of the effects of these substances — blastomogenic, mutagenic, teratogenic, embryotoxic, gonadotoxic, and allergic.

5. Utilization of an integrated experimental data set in addressing the question about permissibility of proposed pesticide uses in agriculture; validation of hygienic regulations and standards for their concentrations in environmental media.

6. Specific appraisal of chemicals from the chemical class under investigation to see whether and how much their effects depend on their chemical structure, physico-chemical properties and time factor; definition by type of the combined effect of the given agent with other chemicals and its joint effect with physical environmental factors.

The term "hygienic toxicology" brings into focus the unity of purpose among the various different fields in toxicology and their subordination to the objectives of hygiene as the science of health. The goals and methods of Soviet industrial toxicology were initially formulated by Lazarev (1938) and Pravdin (1934,1947). Medved (1959), one of the founders of Soviet agricultural toxicology, pointed out that it had borrowed its main methods and means of research from industrial toxicology.

Experimental investigation of new pesticides, as in fact of all other chemicals proposed for agricultural use, has to be completed before their production transfer. At the present time, no new pesticide may be registered for use without appropriate authorization by the USSR Ministry of Health. The lists of agents approved for experimental-commercial applications need yearly coordination with an agreement from the USSR Ministry of Health Sanitary-Epidemiological Department which directs the functioning of the Committee for Pesticide Research and Regulation. Hygienic evaluation of pesticides is performed under a single state (national) plan.

Pesticide toxicology makes a part of agricultural toxicology. In addition to pesticides, the latter studies mineral fertilizers and other substances in agricultural use. Like industrial, food and communal toxicologies, agricultural toxicology is a branch of hygienic (preventive) toxicology.

Agricultural toxicology is presently utilizing on a grand scale current expertise and methods of physiology, biochemistry, analytical chemistry, morphology, histochemistry, molecular biology, gene-

tics, embryology, teratology, immunology, oncology, allergology, and a host of other related disciplines. Toxicologists have to come up incessantly against problems of assessing the hygienic and biological significance of the physiological and biochemical changes identified in experiment. Not every alteration diverging significantly from the control should be considered harmful. The problem then is not one of defining just any threshold of the effect of a substance, but rather the threshold of its harmful effect.

Pesticides affect living organisms in a variety of ways, each manifested by acute or chronic poisonings; by allergenic, mutagenic, carcinogenic, and teratogenic effects; or by other adversities. It is the occurrence probability of these effects that gives a measure of the hazard posed by pesticides. For quantitative evaluation of chemicals in general and pesticides in particular there are three critically important relationships: dose-response, time-response, and dose-time.

Of these, the dose-response relationship is best-understood to-day. The usual method to trace it is "top — down", from the upper limit of toxicity at the level of lethal doses and concentrations down to the level producing no recordable effect. Since the dose-response relationship is probabilistic in nature and bears relation to the species and individual sensitivily of different organisms to the test substance, it is essential that its magnitude be determined with different levels of exposure. Thus one arrives at quantitative probabilistic indices making the results possible to estimate not only for the specific, small animal sample used in the experiments, but also for the whole general population. Probabilistic determination of toxicity parameters for several animal species provides also for the greater ease of animal data extrapolation to man.

The Upper Level of Toxicity. Toxicity patterns are of course easier to investigate and estimate at the lethal and toxic dose levels. The experimenters are therefore fully justified in trying routinely to establish first the upper limit of toxicity. The purpose of this stage in toxicity research is to define LD_{50}, the dose causing the death of 50 percent of experimental animals, and LC_{50}, the corresponding concentration. In contrast with the absolutely lethal (LD_{100} and LC_{100}) and maximum tolerable (LD_0 and LC_0) doses and concentrations, LD_{50} and LC_{50} are probabilistic in character. Theoretically, there is no reason why the LD_{100} and LC_{100} values should be used at all, since they vary with the growing experimental animal population because of an increasing number therein of the animals which will not be killed by the LD_{100}. So the maximum tolerable doses and concentrations must be down as the probability of including "unstable", or non-resistant animals in the experimental sample is up.

Both LD_{50} and LC_{50} are established by probit-analysis (Belenky, 1963) or Prozorovsky's method (1962). With either method it is possible to obtain, along with the median lethal dose levels and concentrations, their standard error and then — with allowance made for the small sample size — their confidence intervals ($LD_{50} \pm S_{LD_{50}} \cdot t$).

The standard error $S_{LD_{50}}$ can be calculated using Miller and Teinter's formula (Belenky, 1963):

$$S_{LD_{50}} = \frac{LD_{84} - LD_{16}}{\sqrt{2N}}, \text{ where}$$

N is the number of animals in the groups used to test the investigated doses whose effect occurs between 84 and 16 percent, and

t is derived from the appropriate table to match the number of animals and the accepted level of probability.

Knowing the confidence limits, it may be stated that, for a specified level of probability and with allowance made for the small experimental animal population, the LD_{50} and LC_{50} values will fall within the calculated confidence interval. This assures adequate probability in comparison of LD_{50} and LC_{50} for different substances or for the same substances but for different animal species. Approximate LD_{50} and LC_{50} values are possible to find using "tentative" methods suggested by Sanotsky (1970) the one-point method, a variant of probit-analysis, being the most simple of them (Van der Waerden, 1940). In the probit grid the line for the dose-response correlation is passed through one data point and parallel to the line for a given substance used on a different species of laboratory animals. To find the experimental point one should know the dose causing the death of any fraction of the test animal population. The latter totals normally 6 to 10 animals. The method is convenient for use on dogs, cats and rabbits. Still another method of Deichmann and Le Blanc (1943) consists essentially in introducing each tested dose to one animal only and then increasing or decreasing it by a factor of 1.5, depending on the results obtained. The minimum dose found lethal is taken to be the LD_{50}.

The values for the upper limit of toxicity would be typically found with respect to the pesticide's routes of entry which may be its actual pathways into the body in occupational or application environments (through the mouth, via the skin or by inhalation). The introduction of chemical substances by any particular route has its distinctive methods, each detailed in appropriate guides and manuals (Pravdin, 1947; Trakhtenberg et al., 1968; Sanotsky, 1970; Elizarova, 1971; Kundiev, 1975; Shitskova et al., 1977). We shall discuss here only some of the quantitative criteria reflecting the toxicity and hazard of substances by entry into the body via the skin or the respiratory system. With respect to highly toxic chemicals that cause animal lethality by dermal application, their upper toxicity parameter is characterized, best, as also in the case of intragastric administration, by the LD_{50} value. Its determination, however, requires the experimental conditions to be strictly standardized. Thus, chemicals are applied to a 2×2 cm^2 bare skin area in the albino rat and a 4×5 cm^2 back area lateral to the vertebral column in the rabbit. The lethal dose levels are inversely related to the application area: the larger areas the LD_{50} values are the lower (Kundiev, 1978). Where a volatile substance

is involved the skin patch to which the substance is applied should be kept under the extract to avoid absorption of the vapours by the inhalation route. Special cylinders with a cover may be also usable (Kundiev, 1975).

LD_{50} values depend also on the mode of application: whether a substance is applied to an open skin patch or wrapped into a water-tight material; on the duration of exposure to the chemical, and other conditions. Kundiev (1975) claims that, in order to ensure complete absorption of the agent and maintain its proper dosage it should neither be washed off the skin nor should a bandage be applied unless so required for some other, special reasons.

Following the determination of the LD_{50} value of a substance by dermal application, it is possible to deduce a dermal-oral coefficient — the ratio of the dermal to oral LD_{50}. Lower coefficients signify the greater hazard of a substance by absorption through the skin. The extent of a chemical's absorption via skin is indicated also by a dermal-venous coefficient — the dermal LD_{50}/intravenous LD_{50} ratio. The latter however has limited applicability because the determination of the intravenous LD_{50} is not routinely a part of the toxicologic evaluation procedure for new pesticides. Whenever no animal lethality is observed as a result of the application of a chemical to limited skin areas, quantitative evaluation of its skin-absorptive toxicity can be efficiently made by dipping the tail of small laboratory animals — e.g. albino mice and rats — into a test tube with the chemical being tested. When animal deaths occur the ET_{50} value, the median lethal time of one half of the experimental animals, may serve as the hazard criterion. The value is defined with confidence limits and therefore has a probabilistic character.

To assess the risk of pesticide poisoning by inhalation, it is important to correlate the LC_{50} values with the volatility rate of the pesticide in question. The effective toxicity of a pesticide is its toxicity times volatility (Pravdin, 1947). But, because toxicity represents the inverse of lethal concentration $(\frac{1}{LC_{50}})$ effective toxicity is $\frac{1}{LC_{50}} \cdot C_{sat} = \frac{C_{sat}}{LC_{50}}$ and the risk of inhalation poisoning is the greater, the higher is the latter value.

Sanotsky (1970) specified further the concept of effective toxicity through its substitution by the expression $\frac{C_{20°}}{CL_{50(120\ min)}}$, where $C_{20°}$ is the maximum concentration at 20° C and $CL_{50(120\ min)}$ is the concentration killing one half of the experimental mice during two-hour exposure. Because the above relation indicates the hazard of inhalation poisoning, the author has introduced what he called a coefficient of potential inhalation poisoning (CPIP).

Using the CPIP value, Ulanova and Pinigin (1974) have divided all substances into four groups: extremely hazardous, with a CPIP above 300; highly hazardous, with a CPIP from 300 to 30; modera-

tely hazardous, with a CPIP from 29 to 3; and slightly hazardous, with a CPIP below 3.

Industrial toxicology employs a coefficient to assess the hazard of sudden acute inhalation poisoning — CSAIP (Lazarev, 1938):

$$\text{CSAIP} = \frac{C_{20^\circ}}{CL_{50} \cdot \lambda}, \text{ where}$$

λ is the coefficient of vapour-gas distribution between the blood and air;

CSAIP less than unity suggests a low hazard of acute poisoning, which keeps increasing as the CSAIP value.

Effective toxicity is also the reciprocal of thermodynamic activity defined as the ratio of the effective vapor pressure of a substance in air (P) to its saturated vapor pressure (P_0) at the same temperature: $A = \frac{P}{P_0}$ (Filov, 1973; Liublina and Mikheev, 1974). Effective vapor pressure, or effective concentration is called the concentration of a chemical giving rise to a certain event (death or intoxication), i.e. $A = \frac{CE_{50}}{C_{sat}}$. The lower the value of the fraction above, the faster will the toxic concentration arise in the air and the more hazardous is the substance involved. Thus thermodynamic activity shows the part of the saturating concentration that a particular toxic (lethal) concentration accounts for. The A values below 0.08 indicate the specific action of substances and those from 0.08 to 1, their non-electrolytic action (Zaugolnikov, 1967; Nemirovsky, 1970).

Since most pesticides are low-volatile compounds, the hazard evaluation of the inhalation poisoning by them should be based, in addition to the indices just described, on consideration of their possible entry into the respiratory system as hydroaerosol or dust. This is indeed the case in the packing and packaging departments of dust-forming pesticides as well as in their dusting and spraying applications for agricultural purposes. The associated hazard of poisoning is dictated by the size of particulate aerosols, the depth of their penetration into the respiratory system, the rate of absorption from the respiratory system into the blood, and the rate of swallowing and further progress from the respiratory system into the alimentary canal.

Lower Level of Toxicity. Determining the lower limit of toxicity, or the threshold of toxic action, is as important in toxicology as the estimation of the LD_{50} and LC_{50} values. One should discuss first in this regard the concept of threshold and the problem of threshold levels. The magnitude of the threshold dose depends on the index selected for its determination and the sensitivity of the method used. The more sensitive the method, the lower the threshold will be. In theory, though, even very minor quantities of biologically active substances will be reacting with biological substrates and thereby have some effect. Doses without effect are non-existent. The question is

how to evaluate the changes they produce as to being harmful or not extending beyond the range of variation common in normal life. The problem therefore is, not to determine just any action threshold, but rather the threshold of harmful effects and thus identify the doses and concentrations of a chemical agent which cause the initial changes poised on the brink of normal and diseased states.

Sanotsky and Ulanova (1975) emphasize the threshold nature of all types of effects, among them mutagenic and blastomogenic, as the guiding principle of Soviet hygiene and preventive toxicology. They conceive of a threshold in general as the threshold of harmful action and examine it at the organismal level. The body harbours constantly occurring adaptations of biological structures and an injury develops solely when the rate of the injurious processes exceeds that of the processes of repair. It remains debatable whether the concept of thresholds is still relevant in the evaluation of mutagenic, blastomogenic and some other effects.

The threshold of harmful effect must be always estimated by resorting to the probabilistic approach. There are two types of indices used to detect the threshold, as indeed any other doses and concentrations. Some of the indices lend themselves to graded changes, as for example an enzyme may increase or decrease its activity. Others are essentially alternative, or quantal, e.g. the "yes-no" response: an animal may die or survive, convulsions or side position may or may not occur. Belenky (1963) believes the quantal (all or none) form of measuring the effects of chemical compounds to be the more important one as it is based on rigorous mathematical methods and thereby offers a more accurate estimation of the effects. It is sometimes possible to shift from the graded to quantal form, for example when one knows the criterion of harm to be expected from a graded alteration.

Kagan (1978), Kurliandsky (1978), Mikheev et al. (1979), and Shtabsky et al. (1979) each validate in their respective studies the need for probabilistic detection of threshold levels of chemical exposures and suggest different methodological approaches to their determination. The latter can be separated into two groups. The first group is of methods that help to gauge the feasibility and desirability of converting graded indices of an effect into quantal ones, with consideration given to the biological significance of an alteration (the criterion of harm); the methods of the second group are designed for direct calculation of DE_{50} and its confidence limits from the results of measuring graded events and their confidence limits.

First, we shall discuss the first group of methods. Kagan (1978) has proposed two alternative probabilistic procedures for the calculation of threshold doses and concentrations by conversion of graded into quantal indices on the basis of the criterion of harm. The first of these applies when the tentative alteration is known and may be considered as unfavourable. For example, a reduction in the activity of erythrocyte acetylcholinesterase and blood serum cholinesterase by 25 percent or greater is evaluated in exposure to organophosphate chemicals as a meaningful unfavourable effect. On this the criterion

workers occupationally exposed to organophosphorus pesticides (OPP) are temporarily relieved from their jobs. It is also the reason why, as the thresholds of harmful effects of new OPP are established experimentally in laboratory animals, the above reduction should be regarded as a sign of the injurious influence and the corresponding graded index should be converted into quantal one. The findings of the experiment are taken to compile a table of dose — response relationships indicating how many of the experimental animals showed the up 25 percent reduction in the activity of the enzyme on exposure to a given dose. The data are subjected to probit-analysis to arrive at ED_{50}, the dose causing 50 percent of the animals to develop the alteration considered as unfavourable, as well as the standard error and the ED_{50} confidence limits adjusted for the small sample size. The exercise allows the threshold doses and concentrations to be identified so as to take into account both, the criterion of harm and the specified occurrence probability of the alteration. A similar approach is feasible in the evaluation of other biochemical and functional disturbances as long as their biological significance can be appreciated.

The second alternative is put to use whenever the quantitative criterion of harm cannot be clearly defined. If so, an alteration extending beyond the confidence limits of the control adjusted for the small experimental animal population ($X \pm S_x t$), can be considered as unfavourable. One relies on the experimental evidence to find the doses causally related to these alterations in all experimental animals. The data are processed by probit-analysis and the DE_{50} is determined with confidence limits. Such an approach was described by Mikheev et al. (1979) though the authors suggest taking $X \pm 2\sigma$ as the alteration to be estimated. To us, this seems poorly grounded, as the method fails to account for the small sample size.

Kurliandsky (1978) regards any alteration as meaningful provided it acts in essentially the same direction as does the average alteration in control. Moreover, even a random deviation having no significant distinctions from the average control may be seen as an alteration.

With the method suggested by Shtabsky et al. (1979) no graded-into-quantal conversion of indices is required. It operates on the assumption of one-to-one correspondence between the confidence intervals of doses and effects. The results can be estimated by graphic or calculation methods.

With the evaluation of the probabilistically determined threshold doses and concentrations in mind, we have staged experiments and performed calculations using the techniques proposed by this author and Shtabsky et al. The probabilistic threshold values as defined with the method of Shtabsky et al. (1979) turned out to be somewhat higher than those obtained by our method. This is because the former method considers the average effect in all animals, whereas the graded-to-quantal conversion requires estimating the responses of the most sensitive animals. Although the striving towards the probabili-

stic determination of threshold levels is legitimate, there is every reason to support the view of Pinigin and Krasovsky (1979) that the determination of probabilistic threshold dose never implies some accurately pinpointed dose, but rather the range of doses of which the threshold of harmful action is one.

An essential input to assessing the biological significance of an alteration comes from the study of biochemical, physiological, and morphological parallels. Biochemical disturbances should be considered harmful when accompanied by long-lasting functional changes involving irreversible structural impairments. Following the trend of these changes over time is of large importance in evaluation of their hygienic significance. Persistent and especially progressing alterations should be classed as harmful without qualification. These alterations should best be described with the method of functional loads revealing the stress of the adaptation mechanisms. If exposure to the tested chemical dose puts a sustained stress on the adaptive processes, such a disturbance should be thought of as unfavourable and taken into account in determining the harmful action threshold. In the same way, the LD_{50} load of a poison is a useful way to clarify the state of the adaptive processes. Whenever administration of poison in a dose equal to the LD_{50} causes death of all experimental animals this suggests clearly a reduced adaptation potential of the organism; if, on the contrary, the animals survive one can safely assume enhanced resistance to the poison (naturally, this must be corrected for probable differences between the experiment and control because of the small sample size).

One can agree with Sanotsky (1970) that integral alterations, if evaluated on the organismal level, can contribute more to proper assessment of the criterion of harm. Nevertheless changes in individual organs and physiological systems, along with disturbances on the cellular and molecular levels, need to be also considered in defining the harmful action threshold. Much depends on the biological importance of the affected system and the character of the observed alteration. An in-depth analysis of specific alterations is necessary with special reference to the type of the pathological state resulting from a higher level of exposure. If the trend of the changes induced by the threshold exposure levels is similar to the basic pattern of the pathological process commonly associated with large dose exposures of a given substance these alterations undoubtedly must be factored-in. This is the way to analyze any quantal or graded response to the test substance.

For proper assessment of graded responses they should importantly be compared with the control and background and, should significant deviations (with a specified probability) surface up, with data on physiological, seasonal and other biorhythmic variations. Based on Anokhin's perception of two vitally important categories of body constants — rigid and plastic — Trakhtenberg et al. (1978) recommend for the variability of an attribute to be determined from the value of the variation coefficient. Accordingly, the authors classify all indi-

ces into rigid, with variation coefficients above 10 percent; plastic, with variation coefficients from 10 to 40 percent; and highly plastic, with variation coefficients above 40 percent. Plasticity data regarding biochemical and physiologic indices must be integrated for the evaluation of alterations.

Plokhinsky (1961) makes a point about three probabilities of errorless judgement commonly applied in biology. For the first probability (0.954), the sample average may not differ from its general population by more than $\pm 2m$, for the second (0.987) by $\pm 2.5S_{\bar{x}}$, and for the third (0.997) by $\pm 3S_{\bar{x}}$. With $t=2$ the probability of error is 1:22; with $t=2.5$ it is 1:81; and $t=3$, 1:370. The third probability of errorless judgement is required for critical studies, in particular the experiments investigating the toxic potency of substances, as suggested by the author. Occasions may arise, however, when a still higher probability of errorless judgement will be needed, as for example in the assessment of carcinogenic effect.

To tackle hygienic regulation problems, one will commonly identify not only the minimum dose exerting a particular type of action with a certain probability (the threshold dose), but also a dose which will not exert it with a necessary degree of assurance (the subthreshold dose). It should be noted that the no-effect or subthreshold dose sets the limit towards which the threshold dose tends as it is reduced. The former dose is called on occasion maximum ineffective, though it is free from any probabilistic connotation. Its reliability will be the greater, the smaller its magnitude, i.e. the lower the occurrence probability of the effect and the higher the probability of no effect. The probability of no effect is the greater, the more the ineffective dose departs from the threshold dose. In critical cases this probability must be as high as 0.997 or higher still.

In order to define the no-effect dose by the up 25 percent reduction of cholinesterase activity the probability level of 0.95 is adopted in biological investigations. Then the error-of-judgement probability of 1:20 can be neglected because even if the subthreshold dose does trigger the threshold effect (the up 25 percent reduction of cholinesterase activity) its impact will not endanger life. For carcinogens, the threshold and maximum ineffective doses must be established to a very high probability that they will not be tumorigenic. Consequently, the determination of threshold doses in critical situations, when no more than few animals in a group — a small sample — are available, should take into account even very low probabilities of adverse effects elicited by the factor under study. For example, the experiment-control differences in response to a carcinogenic exposure may be revealed with a probability much lower than 0.95, but even so they should not be ignored. To ascertain the absence of an effect, i.e. to state positively that the experiment and control apply to the same general population, requires high reliability (a probability on the order of 0.997 or higher). Since with few animals in the sample the distribution of the sample values is likely to be far from normal and follows the distribution law of small populations (Student's distribution), it is

necessary to find for each value of small samples the value of t corresponding to the sample size (Plokhinsky, 1961). In work with small animal populations the effect of low doses may be overlooked altogether. As an example, Knizhnikov (1975) notes that, while a tested dose of a carcinogen induces tumors at the rate of 2,000 per one million individuals, for 100 individuals, the most common sample size in such experiments, the rate will sink to a low 0.2 so that the tumors will not be detectable given the sample sizes the experimenter normally has to work with. Therefore an adequate sample size is critical for the experiments such as these. It is necessary, furthermore, to pay attention not only to the results obtained with minimum dose exposures approximating the threshold levels, but also to the pattern of the dose-response relationship. In addition to making evaluation of the threshold dose more reliable and facilitating extrapolation of the established relationship to the threshold-dose region, this will enlarge the sample and increase the probability of definitive estimation of the threshold and no-effect doses. This approach seems infinitely more promising than the no-threshold principle which, even if supported with experimental data, can at best be used to predict the level of hygienic standards only arbitrarily, by adopting an appropriate "risk" margin. The epidemiologic approach to the evaluation of permissible exposure levels, proposed by the supporters of the no-threshold theory, is unacceptable as regards new substances to be yet accepted for practical use. Knizhnikov (1975) is right in his contention that the maximum permissible exposure must not cause a higher incidence, in a given population, of malignant neoplasms beyond the value corresponding to the error of the mean index. However the probability levels of the no-effect doses should be derived in experiments on test animal samples of the adequate size and at the adequate probability level to allow extrapolation of the results to human contingents of the size likely to be exposed to the agent concerned.

Zone of Acute Toxic Effect. Having established the upper and lower levels of toxicity, we thereby develop an idea about the zone of acute toxic effect as the ratio of the lethal doses or concentrations to the threshold of toxic effect (Tolokontsev, 1963, 1973; Kagan, 1965; Sanotsky, Ulanova, 1975).

$$I = \frac{LD_{50}\ (LC_{50})}{ED_{50}} \text{ or } \frac{LD_{50}}{Lim_{ac}}.$$

If the upper and lower toxicity limits have been determined as probabilistic values, the zone of acute toxic effect will also have a probabilistic character. Then the expression $\frac{LD_{50}}{ED_{50}}$ is more accurate than $\frac{LD_{50}}{Lim_{ac}}$.

The size of the acute action zone gives an idea about the incremental rate of the toxic effect with increasing doses or concentrations

of a poison. The more narrow the zone of acute effect, the faster can a mild intoxication grow into an intoxication with lethal outcome as the dose is increased. The concept is related to that of therapeutic range, a concept applied to evaluate the effect of drugs, which properly characterizes the gap between therapeutic and toxic doses.

With the acute action zone estimated as the ratio $\frac{LD_{50}}{Lim_{ac}}$, chemicals can be classified by that criterion into classes of hazard. Such a classification is given in the section on MAC determination in the air of the working zone, included in the USSR State Standard 12.1.007–76 "Harmful Substances: Classification and General Safety Requirements". It separates all substances into four hazard classes: Class 1 (the most hazardous chemicals), the zone of acute effect below 6; Class 2, from 6 to 18; Class 3, from 8 to 54; and Class 4, above 54. These gradations of the acute action zone may well be adopted also in the hazard assessment of pesticides.

The size of the acute action zone can be judged also by the tangent of the slope of the line illustrating the dose — effect relationship (Tolokontsev, 1963; Kagan, 1965). At the lethal dose level, it is obtained by LD_{50} determination with the method of probit-analysis. If the dose — effect relationship is set out in the probit grid i.e. if the doses and effects are plotted in the logarithmic scale, the tangents are independent of the accepted scale and comparable with one another. Another characteristic of the line slope is the value $S = \frac{LD_{84}/LD_{50} + LD_{50}/LD_{16}}{2}$ (Belenky, 1963) used by Sanotsky et al. to estimate the zone of acute effect for a number of industrial toxic chemicals (Sanotsky, Ulanova, 1975).

Threshold of Chronic Effect. Generally speaking, a properly established threshold of toxic exposure is a key to the validity of the hygienic regulations based thereon. Ordinarily, the effect of a substance is tested in a chronic experiment with two or three concentrations of the substance given by inhalation and another two or three doses ingested orally. The experiment goes on for at least 4 to 6 months in the former instance and 6 to 10 months in the latter. It is conducted in two species of test animals fed a standard diet. The state of the animals is monitored using a set of biochemical and physiological tests which indicate systemic and specific responses of the organism to the toxicant exposure. One of the doses is assumed as the threshold dose and its magnitude is brought down to the level of the hygienic standards by determining a safety factor. The threshold doses and concentrations thus obtained are of value only as guides, for there are too many factors that influence them such as the sensitivity and adequacy of the tests used; the test animal species; the accepted interval, a step, between two tested doses; and the duration of the chronic experiment.

However the method's accuracy is enhanced if the testing protocol makes proper allowance for the magnitude of the acute action threshold determined as a probabilistic value. There are at least three

concentrations (doses) of the substance to be tested, one of them equal to and the other two or three lower than the probabilistic threshold. Each concentration is taken to differ from the next by a factor of 3 to 10, depending on the magnitude of the cumulative effect: the greater the effect, the larger the step should be.

Biochemical and physiologic indices of the animals state are estimated and recorded in dynamics. In the early experimental phase (the first month) samples should be taken more frequently so as not to miss the changes associated with the reaction of anxiety (H. Selye, 1977). Known as the phase of primary decompensation it changes to the phase of stability, or physiological adaptation or, with continued exposure to the agent, likely passes into the phase of compensation for the pathological process and eventually the phase of decompensated pathological state (Sanotsky, 1971), or exhaustion (H. Selye, 1977). It is practical, in assessing the disturbances, to proceed from their degree of hazard. Other inputs in determining the threshold of chronic effect include animals' sex- and age-related sensitivities to chemical agents.

Safety Factor for Setting Hygienic Standards. As the shift is made from the data of laboratory animal experiments to maximum allowable concentrations (doses) for man, this involves setting a certain safety factor. The idea is to render the MACs safe despite difficulties of animal data extrapolation to humans, inaccuracies in evaluating the threshold doses and concentrations in the chronic experiment, and inadequate information about possible effects of exposure to substances or suspicions of them having some undetected, harmful properties. The safety factor is calculated with a formula (Sidorov, 1971) which incorporates the species sensitivity coefficient ($K_1 = \frac{LD_{50\ (max)}}{LD_{50\ (min)}}$, where max and min refer to the least and most sensitive animal species), the cumulative properties of substances, and their long-term effects.

We submit that improvements in the proposed approach to setting the safety factor should be sought in the following directions.

1. Accumulation of information about the relation of species sensitivities to structurally dissimilar substances, with the goal of creating a sufficient data base to assure credible extrapolation to man of the data obtained from experiments in animals.

2. Determination of the threshold and safe concentrations (doses) of chemicals in animals. The probabilistic estimation of their values should involve mathematical modeling of the dose (concentration) — response, time — response and dose (concentration) — time relationships in order to be able to calculate the threshold and subthreshold concentrations at the required probability level. This will minimize the role of random factors due to the arbitrary choice of concentration levels in the chronic experiment, the latter's time-frame, limited experimental animal population, and so on.

Because a study into the cumulative properties of substances seeks to reveal the importance of the time factor in the toxic process, it has certainly a major value for the solution of the stated problem. From our point of view, there is large promise in Liublina's approach (1973) based on mathematical modeling of the process leading to accumulation of inhaled substances in the body.

3. Improvement of the current approach to standard-setting for substances capable of producing unfavourable long-term effects — carcinogenic, mutagenic, and teratogenic. To do this will require, in our opinion, a broader scope for mathematical modeling of the dose-response, dose-time, and time-response relationships in the effort to determine probabilistic action thresholds, that is, such calculated doses and concentrations as would not result in a higher incidence of abnormalities (tumors and mutations) from the control, by more than the value found within the confidence interval of the mean with a specified probability.

To seek out the correlations between the calculated MACs that have withstood the test of experience, and toxicity and cumulative parameters and to derive on that basis regression equations is a most objective approach to setting the safety factors and MAC values for new chemicals. So far the progress in this line of research on pesticides has been quite rewarding (Kagan, Sasinovich, Ovseenko, 1971—1978).

Classification of Pesticides. There exist varied and many classifications of chemicals based on chemical structure, application, mode and mechanism of action, and other parameters and suited to different purposes. An especially relevant classification of chemical substances from the hygienic point of view separates them by the degree of toxicity and hazard. Sanotsky (1970) suggested a graph to calculate substances' relative toxicity by comparison with the most toxic, phosphorus compounds.

Zaugolnikov et al. (1967) brought all chemical compounds under six classes, two of them with subclasses, according to their toxicity by inhalation (on the LD_{50} and MAC criterion and the lethal/saturating concentration ratio) and by the oral route (on the LD_{50} value). Zaugolnikov, Kagan, Sanotsky and Ulanova (1975) suggested a more flexible classification relying on different criteria: the MAC and LD_{50} values by administration into the stomach and by skin application; LC_{50} by inhalation; the coefficient of potential inhalation poisoning (CPIP); and the zone of acute and chronic effects. This categorization adapted and utilized for the classification of chemicals in the national Standard 12.1.007—76, the principal regulatory document for safety evaluation of the toxicants contained in raw materials, finished and semifinished products, and production wastes as well as for their standardization in the air of the working zone (see Table 1).

In the case of pesticides, some of their distinctions require specific approaches to their classification. Above all, this is the fact that pesticides can harm not only the occupationally exposed, but also the

Table 1

Degree-of-Hazard Classification of Chemicals (Standard 12.1.007—76)

Index	Normal for hazard class			
	1	2	3	4
MACs of toxic chemicals in air of working zone, mg/m³	Less than 0.1	0.1—1.0	1.1—10.0	More than 10.0
Median lethal dose by intragastric administration, mg/kg	Less than 15	15—150	151—5,000	More than 5,000
Median lethal dose by dermal application, mg/kg	Less than 100	100—500	501—2,500	More than 2,500
Median lethal concentration in air, mg/m³	Less than 500	500—5,000	5,001—50,000	More than 50,000
CPIP	More than 300	300—30	29—3	Less than 3
Zone of acute effect	Less than 6.0	6.0—18.0	18.1—54.0	More than 54.0
Zone of chronic effect	More than 10.0	10.0—5.0	4.9—2.5	Less than 2.5

general public via consumption of food and water containing their residues and inhalation of polluted ambient air. With respect to the exposed workers, key determinants of the pesticide hazard include, besides the toxicity and volatility of the active ingredient, its quantity in the commercial product (emulsiable concentrates, wettable powders, etc.); for the general population, more important determinants are persistence of the active ingredient, cumulative properties and the ability to accumulate in bodily tissues; for the persons in direct contact with pesticides, their skin-resorptive toxicity must be considered.

Under the classification of pesticides by their designated uses there are four basic groups:

1. Chemicals to control plant pests, animal parasites, and rodents: insecticides (killers of insects), acaricides (of ticks), nematocides (of cylindrical worms), molluscicides (of molluscs), rodenticides and zoocides (of rodents), attractants (attracting insects), repellents (scaring insects away), sex sterilizers, substances of the insect juvenile hormone type, and pheromones (odoriferous substances), etc.

2. Chemicals to control plant diseases: fungicides (against fungal diseases), seed protectants, bactericides, and anticeptics.

3. Chemicals to control weeds and, in broad, affecting plants: herbicides (destructors of weeds), arboricides (of shrubs), algicides (of algae), defoliants (accelerators of leaf shedding), desiccants (dewatering agents), and plant growth stimulators and inhibitors.

4. Pesticides of biological origin: microbial and fungal prepara-

tions and antibiotics obtained by biological methods. Insecticides, acaricides, fungicides and herbicides are the most widely used pesticides, though defoliants, desiccants, zoocides, nematocides, and repellents are also in common use.

On the basis of chemical structure pesticides are commonly classified into organochlorine and organophosphorus compounds; derivatives of carbamic, thio-, and dithiocarbamic acids; dinitrophenol compounds; chlorine-containing derivatives of benzoic, phenoacetic, phenobutyric, phenoxypropionic and other carbonic acids; haloidanilides of carbonic acids; derivatives of urea, thiourea and guanidine; derivatives of sym-triazine, dipyridyl and coumarin; alkaloids and other heterocyclic compounds; hydrocarbons and their derivatives; synthetic pyrethroids; cyan-, rhodan-, fluorine-, iron-, copper-, zinc-, and arsenic-containing agents; organomercury compounds; and also pheromones, insect juvenile hormones, fungal preparations, bacterial preparations, and antibiotics.

Organophosphorus compounds are becoming increasingly important, their scope of production and use ever expanding in Soviet agriculture. Applications are multiplying for methylmercaptophos, phosphamide (rogor, Bi-58), metaphos, chlorophos, methylnitrophos, trichlorometaphos, carbophos, benzophosphate (phosalone), phthalophos, DDVP, and butyphos. The pesticides currently used in farming practices involve over 80 different organophosphates, all of them agents varying in toxicity and selectivity of action.

The derivatives of carbamic, and thio- and dithiocarbamic acids have been applied broadly, some of them showing properties of active insecticides and fungicides; these are esters of N-alkylcarbamic acids and specially aryl ethers of N-methylcarbamic acid (sevin, baygon, benomyl, methyl-N-(2-benzimidazolyl) carbamate, etc.). Others demonstrate properties of active herbicides, as do the derivatives of carbamic acid — isopropyl-N-phenylcarbamate and acylate; of thiocarbamic acid — eptam, tillam, diptal, yalan, carbin, betanal, etc. The derivatives of dithiocarbamic acid are remarkably active fungicides; tetramethylthiuramdisulfide, zineb, ferbam, polycarbacin, polymarcin, etc. The pesticides used as nematocides include carbathion, and derivatives of thiodiazinthion — thiazone; those currently identified and tested for nematocidal applications are methylisothiocyanate (trapex) and chlorine-containing fumigants, particularly DD, a mixture of isomers, 1,3-dichloropropene and 1,2-dichloropropane. Many of the substances in this category are highly toxic to man and warm-blooded animals.

Dinitrophenol derivatives are employed for their insecticidal properties — dinitroorthocresol (DNOC) and nitraphen; acaricidal — acrex and acricide; and fungicidal — kotoran. Fungicidal properties are known in the derivatives of phthalimide — captan and phthalan, of guanidine — cyprex, and of dithiocarbonate — morestan.

The most effective seed disinfectants are organomercury compounds used singly or in combination with other substances (granosan, mercuhexane, mercurbenzene, and phenylmercury acetate).

The non-mercury seed protectants now in use are hexachlorobenzene, pentachloronitrobenzene, TMTD, copper trichlorophenolate, rhodan, vitavax, and formaline, along with the combined disinfectants phentiuram, hexatiuram, and ditan-M-45 — mancozeb.

Zinc phosphide, zoocoumarin and gliftor, all of them potent toxic chemicals, are the most common zoocides.

Among the herbicides being applied in agriculture there are some coming from different classes of chemical compounds. Those heading the list are aryloxyalkulcarbonic acids and their derivatives – 2,4-dichlorophenoxyacetic acid, its esters and salts; 4-chlor-2-methylphenoxyacetic acid; 2,4-dichlorophenoxybutyric acid, and 4-chlor-2-methylphenoxybutyric acid); sodium trichloroacetate, fenac, trisben-200 (2,3,6-trichlorobenzoic acid), tordon, ammonium sulfamate, and triazine derivatives — simazine, artazine, propazine, prometrin, chlorazine, agelon, semeron, etc. Though low-toxic substances, they are nevertheless persistent in the environment and likely to produce an altogether unfavourable effect in the case of chronic exposure. The proximity of these substances to pyrimidine bases focuses attention of them as possible antimetabolites whose effect not infrequently becomes obvious in a long time.

The category of useful herbicides spans also the substituted ureas of the aliphatic and acyclic series and guanidine (dichloralurea, herban, fenuron, monuron, diuron, tenoran, kotoran, linuron, arezine, lenacil, meturin and melprex), together with dipyridil herbicides - reglon (diquat) and some other. Reglon, in particular, is recommended for pre-seeding weed destruction in plow-free land tillage.

Many inorganic compounds are being used as pesticides, among them sulfur- and fluorine-containing preparations, salts of iron and copper, and derivatives of hydrocyanic acid.

Haloidanilide derivatives that are either in the research and development or the early application stage include propanide (stam F-34), solan, dicryl, ramrod, lasso, and CP-52223).

Arsenic compounds are banned for use in the USSR due to their high toxicity, blastomogenicity and low efficacy.

The general evaluation of pesticides to permit their categorization with a particular hazard group relies in part on the principle of limiting criterion. For example, an extremely toxic chemical substance without any other adverse properties should be treated as the substance relevant to the first class of hazard; or a slightly toxic substance with expressed carcinogenic or mutagenic properties comes also into the same hazard class.

The hygienic classification of pesticides takes into account their persistence, volatility, toxicity by intragastric administration, skin-absorptive toxicity, cumulative properties, blastomogenicity, teratogenicity, embryotoxicity, and allergic properties (Medved et al., 1968).

The classification just cited has been utilized by the WHO to design the project of an international pesticide classification according to degree of hazard (1975). The classification distinguishes between pesticide toxicity and hazard. As part of the project development

Hygienic Classification of Pesticides on the Basis of the Main Criteria of Hazard (from Medved et al., 1968)

1. *Intragastric administration*		
Extremely toxic substances	Toxicity (LD_{50})	<50 mg/kg
Highly toxic substances	"	50—200 mg/kg
Moderately toxic substances	"	200- 1000 mg/kg
Slightly toxic substances	"	>1000 mg/kg
2. *Skin-absorption toxicity*		
Strongly marked	"	<500 mg/kg, cut. - oral coeff. <3;
Marked	"	500—2,000 mg/kg, cut. - oral coeff. 3- 10
Slightly marked	"	>2,000 mg/kg, cut.— oral coeff.>10.
3. *Hazard of chemicals by degree of volatility* (chronic exposure)		
Strongly marked		Saturating concentration above or equal to toxic concentration
Marked		Saturating concentration above threshold concentr.
Slightly marked		Saturating concentration exerted no threshold effect
4. *Accumulation*		
Super-marked	Cumulation coefficient (K_{cum})	Within <1
Marked	"	Within 1- 3
Moderately marked	"	Within 3—5
Slightly marked	"	Within >5
5. *Persistence*		
Very persistent	Half-life	1- 2 years
Persistent	"	6 months - 1 year
Moderately persistent	"	1 6 months
Slightly persistent	"	up to 1 month
6. *Blastomogenicity* *		
Evidently carcinogenic		Known incidence of cancer in humans; strong carcinogens in animal experiments
Carcinogenic		Carcinogenicity proved in animal experiments but not proved in humans
Slightly carcinogenic		Slight carcinogenicity in animal experiments
Suspected blastomogens		—
7. *Teratogenicity*		
Evident teratogens		Abnormalities known in humans that are reproducible in animals
Suspected teratogens		Experiment Animal data available
8. *Embryotoxicity*		
Selective		Detectable in doses nontoxic for maternal organism

Moderate	Manifested in conjunction with other toxic effects
9. *Allergic properties*	
Strong allergens	Produce allergic states in most affected persons, even by low-dose exposures occur in real life.
Weak allergens	Produce allergic states in some individuals

* The Classification of Shabad (1966).

effort, the WHO performed a comparative analysis of the degree-of-hazard pesticide classifications now effective in different countries. A novel point centers on different evaluation of commercial pesticide products according to the concentration of active ingredient and the physical state of aggregation (solid or liquid). The need is strongly expressed for a special approach to be taken to the evaluation of chemical product forms. Table 2 summarizes the WHO classification of pesticides by degree of hazard. The LD_{50} were determined in orally dosed rats. The classification gives urgency to the need to introduce standard labeling principles for pesticides.

In 1979 the WHO-recommended classification was updated and supplemented, particularly with the List 1a of extremely hazardous technical pesticide products covering, among others, aldicarb (LD_{50} = = 0.93 mg/kg), chlorfenvinphos (LD_{50} = 10 mg/kg), demeton (LD_{50} = 1.7 mg/kg), dieldrin (LD_{50} = 10 mg/kg), dimefox (LD_{50} = = 1 mg/kg), disulfoton (LD_{50} = 2.6 mg/kg), EPN (LD_{50} = 14 mg/ /kg), hexachlorobenzene (LD_{50} = 10 mg/kg), leptophos (LD_{50} = = 50 mg/kg), M-74, mercaptophos, mevinphos (LD_{50} = 4 mg/kg), parathion (LD_{50} = 13 mg/kg), methylparathion (LD_{50} = 14 mg/kg), phehylmercuracetate (LD_{50} = 30 mg/kg), forat (LD_{50} = 2 mg/kg), phosphamidon (LD_{50} = 17 mg/kg), shradan (LD_{50} = 9 mg/kg), sodium fluoroacetate (LD_{50} = 0.2 mg/kg), sulfotepp (LD_{50} = = 5 mg/kg), TEPP (LD_{50} = 1.1 mg/kg), thiophos, timet and some other agents which are banned for agricultural use in the USSR. The notes to the List 1a identify hexachlorobenzene as causing severe disturbances of porphyrine metabolism in humans; dibromchloropropane as responsible for sterility and mutagenic and carcinogenic effects in animals, leptophos as showing neurotoxic (paralytic) activity, and phenylmercuretate in small doses as affecting the kidneys in mammals and being teratogenic in rats.

Some classifications address pesticide toxicity both in terms of active ingredient and product form. One such, the classification project of the WHO Regional European Office, places in the 1st class: a) all products, regardless of concentration, that incorporate ingredients with the LD_{50} per os of 25 mg/kg or less; b) all other products with the LD_{50} of 200 mg/kg or less; and c) any product to be referred by the LD_{50} for rats to classes 2–4, but which may present a seri-

Table 2

Classification of Pesticides on the Hazard's Degree (WHO)

Class	LD_{50} for rats, mg/kg			
	per os		dermal application	
	solids	liquides	solids	liquides
Ia Extremely hazards substances	5 or less	20 or less	10 or less	40 or less
Ib Highly hazardous substances	5 - 50	20 - 200	10 - 100	40 - 400
II Moderately hazardous substances	50 - 500	200 - 2000	100 - 1000	400 - 4000
III Slightly hazardous substances	more than 500	more than 2000	more than 1000	more than 4000

ous hazard to man. While such an approach, overall, seems more practical to us, analysis of the world literature on pesticides reveals that acute intoxications are caused, for the most part, by the pesticides with the LD_{50} below 50 mg/kg.

The top council of the USSR MA State Commission for Plant Protective Chemicals raised the question about the necessity to further improve and update the existing hygienic classification of pesticides. With this in mind, we have suggested * that the WHO recommendation to differentiate between the degree of hazard posed by solid and liquid pesticide forms and the data on their long-term effects be carefully considered for the new draft classification. In particular, it is suggested to list in the first class, of extremely toxic substances: a) the pesticides with an active ingredient whose oral LD_{50} is below 50 mg/kg, or 300 mg/kg by dermal application and those with a coefficient of potential inhalation poisoning (CPIP, the saturating/lethal concentration ratio) of 300 or more; b) the liquid product forms with the oral LD_{50} of 200 mg/kg or less, or 500 mg/kg via the skin; c) the pesticides responsible in humans for dangerous long-term effects -- carcinogenic and teratogenic -- and endowed with selective organotoxicity, selective embryotoxicity or strongly marked mutagenic or allergic properties accertained in animal experiments.

Class 2, highly hazardous toxicants, should comprise the pesticides whose active ingredient has its oral LD_{50} in the 50 to 200 mg/kg range, or the 300 to 1000 mg/kg range by skin application, as well as the pesticides with a CPIP from 300 to 30 and capable of producing in test animals medium-to-low unfavourable long-term effects, unconfirmed by observations on humans.

Class 3, moderately toxic, refers to the pesticides with the toxicity of their active ingredient -- oral LD_{50} -- from 200 to 1000 mg/kg or those with a CPIP from 30 to 3.

Class 4, slightly hazardous, covers the pesticides with the active ingredient toxicity — LD_{50} per os — above 1000 mg/kg and which cause no animal deaths by dermal application and have a CPIP below 3. All the remaining criteria stand as they are in the hygienic classification of pesticides accepted in the USSR.

The Class 1 pesticides may not be applied in agriculture, those of Class 2 may be applied on a limited scale if necessary, and Classes 3 and 4 may be applied on a wide scale if compliance with the pertinent hygienic regulations and standards is assured. The decision to issue registration and use permits must be based on the relevant limiting criterion.

CUMULATIVE AND ADAPTATIVE PROCESSES IN ORGANISM ON EXPOSURE TO PESTICIDES

As such, accumulation and adaptation is rated among most relevant problems in contemporary hygiene and toxicology. The interaction of chemical substances with the body gives rise to two observable if opposite tendencies – the damaging action of the chemical and the adaptive response of the body it has developed in the course of evolution as a defence reaction to the environment constantly in a state of flux. The severity of assault and the amount and frequency of exposure to the injurious agent decide whether the disruptive changes, elements of break-up, will accumulate and trigger the development of lasting pathological states, or the body will adapt itself to exist under such conditions without pronounced abnormalities.

The former instance is of accumulation prevalent over adaptation so that the body falls prey to a disease, and death if the destruction of its adaptation mechanisms is complete. In the latter, it may well maintain its activity within physiological parameters for a considerable length of time. This basic concept, though it applies to the effect of any environmental agents, is particularly relevant to the science of hygiene. One of its principal aims is to identify the potential and real hazards of chemical environmental factors and define the intensity range of their effects compatible with normal life activity. This is likewise the case with chemical substances for which the study of accumulation and adaptation forms a theoretical basis of their hygienic standardization.

Material and Functional Accumulation. Molecular Mechanisms of Accumulation. Kravkov suggested (1927) an extensive definition for the cumulation of poisons drawing distinction between the concepts of material (chemical) and functional accumulation. With repeated doses, he argues, the toxic effect may arise, not by gradual accumulation of minor quantities of the causative poison, but because of its repeated assault on certain body cells. The impact influence of the small toxicant quantities upon the cells is summated and the toxic effect ensures. But is it relevant to talk about functional cumulation when the poison is destroyed only in part so that fragments of the

substance's molecules remain attached to the receptors as before? Meanwhile the poison itself does not, in fact, accumulate in this case. Obviously, the traditional conceptions need to be somewhat rectified for the reasons suggested.

Looking to the pattern of primary interaction between molecules of the toxicant and biological macromolecules as dictated by the fates of the substance and interested receptors, it is practical to distinguish between at least three, rather than two, types of primary cumulative effect (Kagan and Shtabsky, 1974). When the receptors become inactivated by the presence of not whole poison toxicant molecules but their fragments (parts) the observed cumulative effect is of a mixed type unprovided in classical theory. For the cumulative effect to be sustained, depends on the presence at the receptor site of a material particle. Proper timing in getting rid of the particle is essential if the interested biological macromolecules are to regain their functional activity. This type of accumulation should be considered also in designing a series of toxicologic experiments to set hygienic standarcs for chemical substances.

The considerations outlined above suggest clearly that the cumulative effect of substances, both in type and mechanism, is directly related to, and cannot be discussed apart from, the primary interaction of toxicant molecules with the receptors. Follow a few examples to illustrate the point. Accumulation of metals in the body is a typical instance of its material type. Silver, copper, mercury, lead and also arsenites react vigorously with the functional groups of proteins, sulfhydryl above all, to form mercaptides — complexes of varying stability. It is their stability determining how long the appropriate proteins will remain functional, i.e. the extent of cumulative action. But no matter what the extent, it is the combining of the metal with the protein molecule, that is, the presence of functionally active molecules of the toxicant in the body — a hall-mark of material cumulation — that constitutes a pivotal element in the driving mechanism for this type of cumulative effect. The varying stability of these complexes is made clear by example of arsenic compounds (Lugansky et al., 1957; Petrunkin, 1957).

The mercaptide bonds that arise between trivalent arsenic and monothiols, e.g. cysteine are lower on the stability scale, the arcenic bonds with SH-groups of the protein molecule are placed second and the bonds with dithiols are the most stable (Pokrovsky, 1962). Mercaptides result also from exposure of the organism to mercury and its compounds. The effect of p-chlormercurybenzoate, for example, precisely that mode of action (Dixon and Webb, 1966). Mercury enters into stable complexes with proteins, it thus interferes with enzymatic activity and, given the effect has adequate intensity, leads to intoxication. Since the complexes are fairly stable the entry of added quantities of mercurials into the body produces a more severe enzymatic disfunction and hence the cumulative action of the mercury-containing toxic chemicals. The cumulative action may vary widely in scope depending on the stability of the toxicant molecule complexes with

the receptors, on the one hand, and the functional import of the inactivated receptors, on the other.

The effects of carbon monoxide and cyanides would seem to be also of the material cumulation type. Carbon monoxide inhibits the activity of some iron- and copper-containing enzymes but the worst-stricken of course are the enzymes which react with oxygen. Being the stronger competitor, carbon monoxide displaces oxygen from compounds with hemoglobin yielding carboxyhemoglobin and thereby affecting badly the function of the blood as an oxygen carrier.

Cyanides combine with the enzyme metal necessary to sustain its catalytic function, and block out its influence. Besides, they can interact with the carbonyl group in the enzyme itself — in the cofactor, prosthetic group or even the substrate (Dixon and Webb, 1966). In all of these instances the enzyme stops functioning, in part or in whole, as a result of chemical interaction with the toxicant. The associated disfunction may be enhanced by the newly arriving portions of the poison; what happens then is a material cumulation depending for its magnitude on the stability of the cyanide or carbon-monoxide complex with the cofactors of the protein molecules.

There is a basic difference between material and functional accumulation. The latter features interaction of the toxicant with the biopolymer; the toxicant soon leaves the receptor site though the structure and function of the biopolymer persist for some time without change. The reaction can be assessed by the influence of direct methemoglobin formers (sodium nitrite, etc.).

An example of functional cumulation may be the effect of chemical mutagens. Typically, these are not part of the genes with which they interact; rather they are split off either immediately or soon after the reaction. Compromised secondary and tertiary protein structures which persist after the reaction with a chemical agent illustrate apparently this type of compounds. Sometimes the poison, as it combines with the cofactor or substrate, eliminates them from the body, thus rendering the enzyme incapable of normal functioning. As this happens the toxicant may not be actually present in the body but even so the function remains unhinged for some time. If, during that period, the poison re-enters the body intoxication arises, due to the accumulation of functional changes.

A rather common mixed-type accumulation occurs when some part of the poison molecule adds to the receptor, as in the reactions of acylation of the protein molecule, for example, the phosphorylation and carbamoylation of esterases. In that case, the extent of accumulation is influenced by stability of the complexes thus formed. The phosphorylated cholinesterase remains inactive much longer than the carbamoylated one. It is wrong, however, to class organophosphorus compounds with irreversible and carbamates with reversible cholinesterase inhibitors (Kagan, 1977). The cumulative action of toxic chemicals can be predicted from the basic input of the time taken to reactivate the enzymes and other protein structures.

It should be noted that some OPP have been found capable of inducing reversible non-progressing acetyl-cholinesterase (AChE) inhibition, brought about by interaction of the inhibitor with some part of the active surface of the enzyme responsible for the AChE inhibition by the excess substrate (Brestkin et al., 1971).

Methods to Evaluate Cumulative Properties of Substances. Pharmacologists and toxicologists take a long-standing interest in the quantitative evaluation of the cumulative effect. But because either science has its specific objectives each takes a different approach to it. The pharmacologist finds it necessary now and then to identify the cumulative effect of comparatively high therapeutic doses of a substance given over relatively short periods of time. Unlike him, the toxicologist is concerned with the quantitative evaluation of chemical environmental factors and most of the time has to respond to questions of cumulative effect as related to extremely low quantities of an agent but ones which nonetheless remain active during an indefinite time, sometimes as long as the lifespan of one or even several generations. Because of their distinct approaches pharmacologists and toxicologists apply different methodological tools to study the cumulative action of substances. In pharmacology the researcher relies on Druckrey's method (1957) to gauge cumulation, its key feature being the selection of filling (lacking) dose due to be administered one day after maximum tolerable dose (LD_0). The filling dose should be able to cause the death of 50 percent of the animals.

Thereafter the LD_{50} is subtracted from the sum of the LD_0 and the filling dose and the difference characterizes the extent of cumulation.

Larionov and Grishko (1963) measure the LD_{50} of a substance at different intervals upon LD_0 administration. Both these methods are deficient in that, with the administration of two large toxicant doses as their key feature, they disallow approximation of real-life conditions.

A quantitative evaluation criterion for the substances endowed with material cumulation may be their daily elimination rate expressed as percentage of the administered dose (Votchal, 1963). Knowing it, one is able to determine the time taken for a particular dose of the agent — therapeutic, toxic, or lethal — to accumulate in the body. Another, and similar method measures the residence half-time of a chemical in the body (biological half-time). Under the method, the time taken to eliminate the injected chemical is given by the formula:

$$T_{50\%} = \frac{(t_1 - t_2) \cdot \ln 2}{\ln y_1 - \ln y_2}, \text{ where}$$

y_1 and y_2 are the chemical's contents at moments t_1 and t_2 after its administration.

Shtabsky (1971) has shown that determining the residence half-time of a chemical allows its possible accumulation with any mode of administration into the body to be evaluated in quantitative

terms. If the intervals between two successive administrations (Λ) of a certain chemical dose are shorter than its residence half-time (T), it will accumulate the faster, the lower the ratio $\frac{\Delta}{T}$. With $T = \Lambda$ the amount of the substance likewise increasing and its maximum level after a single injection is doubling after some five or six administrations. Should the given dose continue to enter the body, there comes dynamic equilibrium with the ceiling concentration of the chemical. The level of dynamic equilibrium and the speed with which it is achieved can be calculated for each dose. With $\Lambda > T$ the substance will accumulate the slower, the greater the ratio $\frac{\Lambda}{T}$; with Λ significantly larger than T, material cumulation is distinctly impossible.

Criteria of Cumulative Effect. Cumulation Coefficient. Of the two methods most widely used for the evaluation of functional cumulation, one proceeds by regular (daily) administration of the agent to experimental animals in equal doses that represent certain fractions of their LD_{50} (Kagan, Stankevich, 1964). The other one, called the "subchronic toxicity" test, involves gradually increasing the dose of a chemical (Lim et al., 1961). Under both methods, it is possible to calculate cumulation coefficient, which is essentially the ratio $\frac{\Sigma LD_{50(n)}}{LD_{50(1)}}$ or, to generalize, $\frac{\Sigma DE_{50(n)}}{DE_{50(1)}}$. DE_{50} is taken to mean the dose producing a certain effect in 50 percent of the experimental animals (for the quantal evaluation) or a 50 percent modification of the selected index (for the graded evaluation of the effect). The calculation of cumulation coefficient by the formula given above enables its confidence limits to be established and the coefficient itself to be perceived in terms of probability (Kagan, 1964).

Cumulation coefficient is defined usually in two species of test animals fed daily with a dose of 1/10, 1/20 and 1/50 LD_{50}. The idea is to have every animal receive summarily, under the 2–4-month protocol of five doses a week, 10, 5, and 2 LD_{50} of the substance, which is quite enough to estimate the degree of the cumulative effect.

Since cumulation coefficient may depend heavily in magnitude on the animal species and the daily dose involved on two magnitudes can be compared for different substances unless obtained under similar conditions, including at least two animal species and several equal fractional doses. The coefficients approaching unity suggest a remarkably strong cumulative effect and those above 5, a mild effect. Medved (1965) suggested that pesticides should be classified by the magnitude of their cumulative properties into four groups: super-cumulative, with the cumulation coefficient below unity; strongly cumulative, the cumulation coefficient from 1 to 3; moderately cumulative, the cumulation coefficient from 3 to 5; and slightly cumulative, the cumulation coefficient greater than 5.

Shtabsky (1971) appreciates cumulative action by the ratio of deaths at the 30th to the 1st day after a single toxicant dose. The cumulative action is the greater, the lower is

$$K = \frac{LD_{50}(30th\ day)}{LD_{50}(1st\ day)}.$$

A correlation exists between the time animals die after the injection of a pesticide and the magnitude of the cumulative effect, namely, the shorter the time elapsed from the injection of the poison until the animal's death, the smaller the magnitude of the latter's cumulative action. Thus the highly toxic OPP capable of causing animals to die within the hour post-injection are slightly cumulative. Conversely, the organochlorine agents which upon injection kill animals after 2 or 3 days or later are, as a rule strongly cumulative. This is readily explainable because with a slow-developing pathological process all subsequent poison doses are administered before reparation of the previously disturbed functions has taken place. On that basis, a quantitative index of the cumulative effect, or cumulation index, is estimated thus:

$$I_{cum} = 1 - \frac{LD_{50(1)}}{LD_{50(14)}};\ \text{where}$$

$LD_{50(1)}$ is the dose calculated from data on animal deaths on the first day of the experiment; and

$LD_{50(14)}$ is the same, after 14 days. Should they coincide and all animals die on the first day, the cumulation index is zero. When the animals die later into the experiment the cumulation index tends toward unity.

Shtabsky and Kagan (1974) suggested that the procedure for evaluation of cumulation coefficient should be standardized so as to incorporate the cumulation index.

The standardized cumulation coefficient is:

$$K_{cum}^{stand} = \frac{LD_{50(1)}}{LD_{50(14)}} K_{cum},\ \text{where}$$

$$K_{cum} = \frac{\Sigma LD_{50(n)}}{LD_{50(1)}}.$$

Its estimation excludes the risk of underestimating the cumulative properties of strongly cumulative substances.

The formula $\frac{\Sigma DE_{50(n)}}{DE_{50(1)}}$ allows calculation of cumulation coefficients at the threshold level. Gizatulina (1970) reported that animals injected with triphehylphosphate doses equal to 1/5 and 1/2 of the DE_{50} develop habituation to it within two weeks or so. She went on (1971) to identify cumulation coefficients at the threshold level for eight compounds (including three organophosphates and two organochlorines) and found them to be lower than those at the lethal level.

Ulanova et al. (1972) estimated $K = \frac{\Sigma DE_n}{DE_1}$ and again cumulation coefficients of the same substances at the threshold level had a lower value than at the lethal level. Similar data were obtained by Kagan et al. (1971). Thus it appears that the cumulation coefficients at the threshold level cannot be evaluated on the same criteria that apply to lethal outcomes.

The principal purpose of experiment with repeated administration of a substance must be to define the nature of the alterations that arise, detect primary mechanisms of toxic response and establish dynamics of the cumulative process and the development of compensatory defence mechanisms. Quantitative perceptions of the toxic process and the dose-time-response relationship may be evolved by correlation of the results obtained with repeated exposure to several different doses of a substance. Recognition of trends in the initiation and development of toxic process provides basic clues to identifying safe exposure levels.

Dose-Dependent Accumulation Patterns. Having estimated several cumulation coefficients for various doses of a substance, one can proceed to establish specific patterns of dependence of the extent of cumulation upon the daily dose. The exercise has certain predictive value in forecasting cumulative properties of substances in chronic exposure of the organism to their relatively minor quantities.

We have identified four principal patterns in the extent of cumulation as a function of dose (Fig. 1):

1. The extent of cumulative action decreases with a progressively reduced daily dose. The pattern is characteristic of many highly toxic organophosphorus insecticides. They are not high-risk agents capable of causing chronic poisonings by protracted low-dose exposure. In forecasting, however, one must be careful to see if the chronic effects that arise are not due to the action of their metabolites. For example, the organophosphate chemicals with a paranitrophenol group in their molecule, such as thiophos, metaphos, etc., produce in chro-

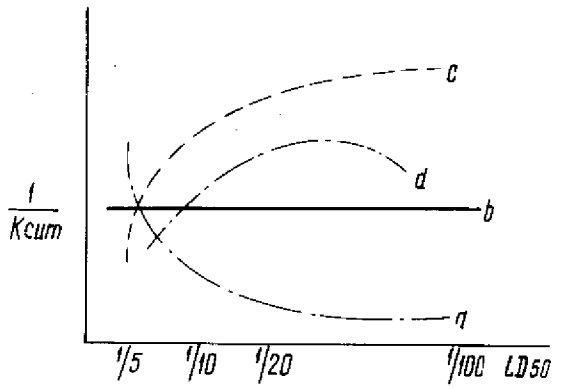


Fig. 1. Patterns of dose-dependent cumulative action of pesticides:
a – organophosphates; *b* – dithiocarbamates; *c* – organochlorines; *d* – diene compounds

nic exposure anemia and other blood changes which apparently have nothing to do with the principal anticholinesterase type of the agent's impact; chlorine-containing organophosphorus agents — chlorophos and DDVP — have a toxic effect on the liver; phthalophos, which contains a phthalimide group, and sayphos having a triazine ring in the molecule affect kidneys, and so forth. These developments may pass unnoticed by a researcher who looks to lethal outcome as the only guide in estimating cumulative action and therefore ignores the necessity of biochemical, physiological, and morphological investigations.

2. The extent of cumulative action is independent of daily dose. In Fig. 1 we see the straight line which is parallel or nearly parallel to the baseline. This is the pattern found typically in some dithiocarbamates (tetramethylthiuramdisulfide). The group is a more dangerous one as chronic intoxication may be manifested upon entry into the body of not only large, but small doses as well.

3. The extent of cumulative action increases with decreasing dose of the agent administered daily into the body. The pattern is common to a number of organochlorine hydrocarbons, likely to produce chronic effects in very low doses and, naturally, being potentially most hazardous.

4. The extent of cumulative action decreases as a two-phase process. First, it climbs as the dose is lowered, and then sets on a downtrend. The pattern applies to the action of some chlorinated hydrocarbons of diene synthesis (aldrin, dieldrin, etc.).

Adaptation, Habituation, Compensation. Adaptation refers to the ability of a living organism to adjust to the everchanging conditions of existence in environment, developed throughout the evolutionary process; without adaptation neither maintenance of normal activity nor adjustment to diverse environmental factors is possible.

Sanotsky (1971) describes MAC, the maximum allowable concentration, as the concentration of a chemical in the environment which acts upon the human organism periodically or through its entire life span directly or indirectly — via ecological systems, but without giving rise to somatic or psychic disorders (including latent and temporarily compensated) or health changes going beyond the limits of the physiological responses detectable with current methods of research, at once or in a longer term of life of the present and future generations. This definition very appropriately links the concept of maximum allowable concentration (MAC) to adaptation processes within the life-span of not one, but many subsequent generations.

As defined by the WHO Working Group (1974), adaptation is the true accommodation of an organism to a changing environment which occurs without any interference with a given biological system and without excess of its normal (homeostatic) capability of response, i.e. when, after a certain period of exposure, its response to a toxic substance (usually at a low concentration) disappears completely and for ever and is no longer detectable by various functional tests or extensive additional investigations.

Habituation is a decrease of body responses to continuous or periodic stimulation (Liublina et al., 1971). While a form of the adaptive response of the body, it may not be an expression of true adaptation, but merely a phase of intoxication. Indeed, the state of habituation is often followed, under continuing exposure, by impairment of compensation and adaptation reactions and the development of intoxication. The WHO definition of habituation emphasizes the disappearance, after a certain time, of the disturbances caused by toxic substances (so that their concentration has to be increased to obtain the effect next time). Such habituation must be seen as a stage of chronic intoxication.

Compensation is the adjustment of an organism to changing environmental conditions (especially chemical), it is a result of the stresses that arise in the biological systems and exceed the limits of their usual (homeostatic) potential. This is a latent pathological state which may show up with time as overt pathological changes (decompensation).

Sanotsky (1971) identifies the following distinct stages of intoxication: primary decompensation, physiological adaptation, compensation of the pathological process, and decompensated pathology.

Starkenstein (1931) maintained that habituation to a certain poison is, in some sense, the reverse of cumulative action. Repeated exposure of the body to chemical substances provokes usually both processes: an increasing injurious action, and cumulation and adaptation. Depending on the severity of assault by the agent and the dose and duration of exposure, either the damaging or the defensive tendency prevails. So the investigator concerned with the study of secondary effects from chemical substance doses always has to confront both, cumulation as well as adaptation.

While studying the effect of OPP on the body we paid attention to the fact that daily administration of the chemical in certain doses to albino rats caused partial death of test animals within the first few weeks, whereas the other part of the animals given the same doses of the agent under identical conditions, stayed alive during several months despite the continued administration of the toxicant. We hypothesized that these animals had developed habituation to the OPP. To evaluate their condition, we applied a test with LD_{50} administration of the same poison. But even so the experimental animals, as contrasted from the control, did not die, i.e. their resistance to the poison kept increasing. This might be attributed to the same mechanism we had earlier observed in experiments with prevention of death in rats from lethal mercaptophos doses by pre-administration to them of subtoxic doses of the same poison or other organophosphorus insecticides (M-81, metaphos) which were less active cholinesesterase binders. Probably, as with the reversible cholinesterase binding by eserine, the case here is one of continuous release of cholinesterate from the compound with an inhibitor in sufficient quantities to support life. Or we may deal here with habituation to

excessive concentrations of acetylcholine competing with the orgarophosphorus compounds for the active centers of cholinesterase.

Tzapko (1966) watched laboratory animals developing habituation to chlorophos. Its daily administration at 1/20 LD_{50} led, after 20 days of the experiment, to a substantial reduction of cholinesterase activity in the blood serum and erythrocytes, liver, kidneys, spleen, and brain. After 40 days, the cholinesterase activity in the serum and erythrocytes was somewhat higher; after 90 days, it surpassed the 20-day level, nearly reaching at this point the initial level in the blood serum and spleen. Some indices of the functional state of the liver changed much like cholinesterase, particularly as regards the release of sodium benzoate in the form of hippuric acid which had attained the initial level by the 90th day. Thus there had been gradual habituation to chlorophos involving the return to normal of the pathological process underlying the toxic response (restoration of cholinesterase activity) and other pathological insults. The development of habituation to OPP was also described by Krasovsky et al. (1970).

Several hypotheses may be put forward to account for the OPP habituation mechanism.

First, repeated OPP doses may trigger the induction of the enzymes responsible for their detoxication. Acceleration of the OPP detoxication processes reduces the number of toxicant molecules which react with the active centers of cholinesterase and thereby contribute to its restoration. This is confirmed also by the data about the faster detoxication of organophosphates by the resistant insects. Consequently, the induction of OPP-detoxifying enzymes may be listed among the major causes of resistance to the organophosphorus compounds.

The second mechanism seems to be the dephosphorylation of cholinesterase. Evidently, the accumulation of excessive acetylcholine quantities via the feedback mechanism increases cholinesterase activity. Lastly, one cannot dismiss either accelerated synthesis of cholinesterase itself by the feedback mechanism through partial inactivation of the active enzyme of OPP. These hypotheses need experimental verification, yet the fact that habituation may be developed and resistance to OPP may be increased, following their repeated entry into the body is not in doubt anymore. As is generally known, destruction of OPP-resistant insects requires toxic concentrations to be raised dozens if not hundreds of times. This says something about the potency of the OPP adaptation mechanisms in the insects: they are consolidated genetically. Habituation in man develops over the life-span of one generation and therefore is not nearly as potent. Nonetheless human habituation, too, has to be reckoned with in evaluating the cumulative effects of OPP.

The literature abounds in evidence of habituation to hydrocarbons (gasoline, benzene, xylol, diethyl ether, etc.; Liublina et al., 1971), organochlorine agents (aldrin and dieldrin, and heptachlor), derivatives of carbamic (carbin) and dithiocarbamic acid (polycarbacin; Martzon, 1968). With repeated exposures to subthreshold doses of

the organochlorine compounds of diene synthesis, Spynu (1965) discovered changes of ascorbic acid contents in the adrenal glands, of the absolute quantity of eosinophiles in the blood, and of 17-ketosteroids in the urea, each of them in a phased pattern. First came increased resistance to additional administration of the threshold or lethal chemical doses and lowered sensitivity to functional stresses. This was followed, at the next stage of intoxication, by heightened sensitivity to poisons and functional loads. The highly toxic and highly cumulative diene insecticides showed the characteristic ability to move quickly from the first to the second stage.

Investigation of the effects produced by different doses (fractions of LD_{50}) in an experimental study of cumulative effect offers a way, in some cases at least, to monitor the adaptation vs accumulation ratio and thereby not merely to define the magnitude of the threshold doses, but also to identify for them relevant criteria of harm. The doses responsible for progressive alterations over time should be assessed as positively harmful; the amounts of poisons which do not harm the adaptation potential when administered into the body are on the threshold of toxic exposure.

Daily administration of polycarbacin doses corresponding to 1/10 and 1/20 LD_{50} was seen to decrease progressively the amount of SH-groups in the blood of the albino rats. Its administration in doses of 1/50 and 1/100 LD_{50} at first caused the SH-group level to drop off and then take a trend towards restoration.

Though the data now available shed light on some adaptation mechanisms to chemical substances, they cannot be considered to be sufficiently well known at present. Perhaps the most detailed findings thus far were obtained by Liublina et al. (1971). Among the mechanisms adjusting the body to the effects of environmental factors within as short a time as possible, the neurohumoral mechanisms are worth mentioning ahead of others by reason of supporting "behavioral" adaptation, one of the most effective of its kind (Lazarev, 1967). In man, adaptation to a change of environmental conditions is made evident by the activity of the far-advanced human brain. Yet humans, like animals, also possess their inherent adaptation mechanisms in the form of "general adaptation syndrome". Adaptation of the nerve tissue comes about through increasing lability and slightly decreasing excitability. Repeated exposures produce moderate excitation in the nervous system which not only boosts the development of habituation, but also magnifies its extent (Liublina et al., 1971). With respect to the nervous system, the processes of habituation to poisons appear to be closely related to the humoral mechanisms. Liublina et al. (1971) believe that the system out of the pituitary body and the cortical matter of the adrenal glands, on exposure to low-intensity chemical factors, has not as large a role as it has in an acute stress.

Adaptation at the cellular level typically resides in microorganisms and, moreover, makes them resistant to drugs. Habituation at the cellular level results presumably from changes in metabolism. Alterna-

tely, it may be associated with the growing activity of the detoxifying enzymes or increasing resistance of macromolecular structures to poisons and expanded synthesis of proteins and nucleic acids. One can cite numerous examples of microsomal liver enzymes increasing their activity on exposure to toxic substances and drugs. DDT and other organochlorine compounds can activate the microsomal liver enzymes involved in the detoxification of organophosphorus compounds, thus producing cross-habituation to them.

Live cells, much like the multicellular organisms in their entirely, present a rather efficient self-regulating system with advanced feedback mechanisms. Allosteric processes are importantly involved in actuating the mechanisms. Final metabolites influence the activity of the enzymes which catalyze the primary reactions leading to their formation, even though metabolites show no structural similarity to the enzyme substrate. It appears that many poisons interact with the enzymes by the allosteric mechanism (Kurganov, 1978) and that the interaction is all too likely to result in adaptive reactions. Though little is known at present about the workings of adaptation mechanisms at the molecular level, their role seems to be far from minor. The mechanisms drive the induction of enzymes and the new enzymes are termed adaptive because their effect is directed most often to removing their causal agent or the consequences of the latter's activity.

The role that the induction of mixed-function oxidases has in the mechanism of adaptation to a number of pesticides was conclusively demonstrated by Popov (1977).

A quantitative evaluation of the state of adaptation employs a variety of loading (effort) tests that provide at least some indication as to the degree of stress imposed on the adaptive mechanisms. Shtabsky (1971) proposed this formula to determine the adaptation coefficient:

$$K_{ad} = \frac{ET_{50} \cdot 100}{T_{50}}, \text{ where}$$

ET_{50} is the mean effective time of lethal outcome following the injection of poison to test animals, and

T_{50} is the average life-span for a given animal species.

ABSORPTION, DISTRIBUTION, ELIMINATION, AND METABOLISM OF PESTICIDES IN ORGANISM

The research on toxicokinetic processes has direct relativity to effective solution of several theoretical and applied problems in pesticide hygiene and clinical treatment of intoxications, such as design of exposure tests and diagnostic methods (detection of toxicants and their metabolites in the biological media); discovery of antidotal and pathogenetic therapies (chemical binding of poisons, potentiation of

detoxifying and elimination processes); preventive programs; study of the adaptation mechanisms and further enquiry into the problem of their control through the induction of detoxifying enzymes, inhibition of the enzymes implicated in lethal synthesis, etc.; prevention of poisons from entering the body; validation of hygienic regulations and standards (selecting more rational operating conditions and documenting the need to alternate pesticides so as to avoid their combined effect).

Toxicokinetics is rested on the theoretical foundation of the doctrine about structural and functional characteristics of cell membranes, for it is on the interaction with the cell membranes and cell organoid enzymes that foreign chemical substances depend most for their transport and transformation.

Pesticides' Entry into the Body. Pesticides can gain access into the body through the respiratory system, skin, and alimentary canal. Though in pesticide handling and application the first two routes have undisputed priority, still one must be aware that a large portion of the chemicals may be swallowed and thus find their way into the alimentary canal as aerosols are inhaled.

By way of the respiratory system, toxic chemicals enter the body in the form of vapors, as do the fumigants methyl bromide, chloropicrin, and others; or vapors and aerosols, as some high-, medium-, and low-volative agents — DDVP, ethylmercurochloride, etc. The specific modes of pesticide application in agriculture (dusting and spraying plants) harbour the possibility of low-volatile substances penetrating into the respiratory tracts as dusts or hydroaerosols. The likelihood of subsequent intoxications is judged by the magnitude of the potential inhalation poisoning coefficient (CPIP) or thermodynamic concentrations. The basic principles for the ingress of toxic substances into the body have been covered extensively and in much detail by Gadaskina and Filov (1971), Golubev et al. (1973), and Tolokontsev (1976). With this in mind, the present discussion will be limited to more general questions inasmuch as they apply to pesticides.

For the speed (rate) of their absorption from the respiratory system into the blood, chemical agents depend on their state of aggregation (vapors, hydroaerosol, dust); the particle-size and charge of the aerosol; the latter's depth of penetration into the respiratory system; Ostwald's partitioning coefficient ($\lambda = \frac{\text{concentration in blood}}{\text{concentration in air}}$, water solubility data may be used here instead of solubility in blood); partial vapor pressure; the extent of pulmonary ventilation; bloodstream intensity in the lungs and the state of the lung tissue (presence of inflammatory foci, exudates and transudates), and, finally, the chemical interaction pattern of pesticides with the biological substrates of the respiratory system. The partitioning coefficient in the $\frac{\text{arterial blood}}{\text{alveolar air}}$ system characterizes the partition of volatile compounds between liquid and gaseos phases at the moment of equilibrium. The lower the coefficient, the faster is equilibrium attained.

The rate of attaining equilibrium and the allied effect increase intensified breathing, i.e. with muscular effort. The partitioning coefficient is calculated from the formula:

$$\lambda = \frac{V \cdot 22.4 \cdot 760 \cdot T}{P \cdot M \cdot 273}, \text{ where}$$

V is solubility in water, g/l;
T is absolute temperature,
P is vapor pressure, and
M is molecular mass.

Coarse dust or liquid aerosol settle for the major part in the nasal cavity, the nasal portion of the throat and the trachea, though a large portion is swallowed (the poison gains access by multiple entry; Katznelson, 1976). The alveoles are penetrated mainly by particles sized 1—2 μm and particulates of larger diameter are retained in the upper compartments of the respiratory system. However water-soluble aerosols may be absorbed also via mucous membranes. In fact many OPP, if inhaled in the aerosol state, make rapid progress into the bloodstream, thus producing a toxic effect in the first few minutes of exposure. The OPP aerosols will normally be more toxic than their vapors (Kagan, 1977). This is perhaps because they are capable of being retained and absorbed all through the respiratory system whereas the vapors are absorbed primarily from the alveoli. Also, percent retention may be higher for the aerosols than vapors.

Reactive and non-reactive compounds have dissimilar absorption patterns. For the non-reactive gases and vapors, their accumulation in the blood is represented by the exponential curve:

$$C = C_0 (1 - e^{-kt}), \text{ where}$$

C is the concentration of a substance in a biological system;
C_0 is the environmental concentration,
t is time, and
k is the coefficient being computed.

The speed with which substances in the process of biotransformation accumulate in the blood and tissue is influenced markedly by their metabolic rate. The faster the substances degrade, the lower their amounts accumulated in the form of primary compounds. For many of them, the metabolites are likely to be found in the biosubstrates. The possibility of accumulation in the rat liver has been demonstrated by Popov et al. (1977) for a toxic metabolite of Dursban, its P = O analog.

One can gain a notion about the pattern of pesticide absorption from the respiratory system by measuring the speed with which signs of intoxication develop and by looking to specific indices of the toxic effect (e. g. cholinesterase activity in OPP exposure). Rapid (in a matter of minutes) absorption of organosphophorus chemicals

was reported by Voitenko et al. (1977) and Kagan (1977) from studies of erythrocyte acetylcholinesterase (AChE) and blood serum cholinesterase (ChE) activity in tissues. While dichloroethane, carbon tetrachloride, hexachlorobutadiene, methyl bromide and hexachlorobenzene pass on from the lungs into the blood in inhaled fumes (Bakhishev, 1970; Gadaskina, Levina, 1976), DDT, kelthane, heptachlor, aldrin, dieldrine, polychloropinene and polychlorocamphene enter it in inhaled aerosols (Spynu, 1965; Gadaskina, Levina, 1976), as do also sevin and other esters of carbamic acid (Yakim, Klisenko, 1967; Abramova, Cherny, 1976), tetramethylthiuram disulfide (TMTD), and ziram (Abramova, Cherny, 1976).

Pesticides move into the body via the skin as well as the respiratory organs. The skin-resorptive ability is known for lipid-soluble substances (even those with medium-to-low water solubility; Lazarev, 1938). Rapid skin absorption is also a feature of organophosphates, organochlorines, organomercurials, derivatives of dinitrophenol, carbamic and thio- and dithiocarbamic acids, the alkaloids nicotine and anabasine (Kundiev, 1975; Kagan, 1977; Lazutka, 1972), and the dipyridyl herbicides paraquat and diquat (Makovsky, 1971). Organophosphate sulfoxides and sulfones become absorbed less readily than nonoxidized compounds (Kundiev, 1975). Ionized compounds and positively or negatively charged molecules poorly penetrate the skin. The passage of polar compounds through the skin is magnified if they are mixed with non-polar solvents, a proven fact as far as some OPP agents are concerned.

The hazard of poisoning via the skin looms large from the pesticides featuring a combination of marked toxicity, easy absorption and medium-to-low volatility. The danger is extraordinary as coming from OPP and some of the more toxic organochlorine pesticides (gamma-isomer of hexachlorocyclohexane, and the compounds of diene synthesis). Kundiev (1975) avers that many organophosphate intoxications that occur to occupationally exposed workers occur by the dermal route of entry. Intoxications result from occupational exposure to thiophos, mercaptophos, TEPP, phosdrin, and carbophos. They happen most frequently where liquid pesticides are in use, though cases have been documented of poisoning by pesticides in the dust form.

Medved (1961) and Trakhtenberg (1969) focused attention on the ability of organomercury compounds to penetrate the skin and pass into the body. The intoxications caused by these pesticides are assumed to be a result of their combined entry through the skin and respiratory system. There are data suggesting absorption through skin as the possible route of entry for the derivatives of carbamic and thio- and dithiocarbamic acids, herbicides from the group of 2,4-dichlorphenoxycarbonic acids, pyridine derivatives, and other pesticide classes.

The mechanism responsible for the skin absorption of pesticides is somewhat complicated, as their direct penetration through the epiderm, hair follicles and sebaceous glands or through the ducts of the sweat glands is a possibility. Different skin areas show a dissi-

milar capacity for the absorption of chemicals, being more permeable to toxic agents on the medial surface of the thighs and hands, in the inguinal region, on the genital organs, breast, and abdomen.

A methodologically meaningful issue deals with comparative skin permeability in humans and experimental animals (Gostinsky, Maeva, 1977). The animal species most typically used to test skin-resorptive action are the albino rats and mice, rabbits, and guinea pigs. Kundiev (1975) notes that human skin is less permeable than the skin of numerous experimental animal species because of the more thick horny layer of the epiderm and the greater length and tortuosity of the glandular ducts. The skin of piglets is almost as permeable as the human skin, yet the small laboratory animals are more suitable for use in skin absorption studies because of the larger body surface area/weight ratio. For proper evaluation of the results of these studies one could consider the influence of temperature, humidity, solvents, emulsifiers and other factors on the absorption rate.

Another route for pesticides to access the body is through the alimentary tract, and some substances begin to be absorbed already in its upper portions. This is true of the compounds that are readily dissolved in lipids and possess a high oil/water coefficient, as in the case of numerous organochlorine and organophosphate chemicals, phenol derivatives, cyanides and some salts. Organochlorine compounds, for example DDT or dieldrin, have been found to be absorbed from the upper portions of the alimentary canal, transported into the liver through the portal vein system and partially the lymph, metabolized in the liver, and either cleared into the intestines or deposited in the fatty tissue. Hexachlorobenzene is absorbed much slower by comparison, but is also deposited in the fatty tissue (Jatropoulos et al., 1975). The stomach, too, may be the source of entry of the same agents into blood. Non-ionized compounds are absorbed by diffusion. The major bulk of the pesticides entering the bloodstream reach it through the small intenstine. The barrier between the intenstinal cavity and blood incorporates the intestinal epithelium, the epithelial membrane on the capillary side, and the capillary basilar membrane.

Compounds featuring very high lipophilicity are fast to penetrate the intenstinal wall but comparatively slow to move on to the blood. Fast absorption requires of the pesticide concerned to be readily dissolved in lipids and water, the altogether necessary property for the entry of substances from the intestines into the blood-stream. The presence, or lack, of lipoid solubility is critical for the diffusion of substances across the erythrocyte membrane. Proteins and especially plasma albumins have a large role in pesticide transport by the blood on account of their large surface area. In erythrocytes, chemicals can combine with hemoglobin (Wilkinson, 1976). The capillary wall is permeable to many pesticides whose amounts conveyed into the tissues are proportional to the capillary bloodstream rate.

Other major influences as regards the pattern and speed of chemical absorption are the extent of accumulation, solubility in the sto-

mach, and pH of the latter's medium. The substances ingested in a fasted state are usually capable of more intensive absorptior than otherwise. Also, the chemical absorption rate is controlled in large measure by the degree of ionization of molecules. Gadaskina and Levina (1976) indicate that a negative logarithm of the ionization constant (pKa) greater than 3 makes absorption possible for the substances of the acidic nature and a logarithm less than 8 for those of the alkaline nature. In other words, poor absorption happens to the substances found in the ionized state in a weakly acidic or weakly alkaline medium. With the increasing pH of the medium the absorption rate of the bases is up and that of the acids is down.

The absorption rate of ions from the small intenstine is very low. Absorption of aminoacids, glucose, uracil and structurally similar foreign substances, e.g. uracil fluoride (Albert, 1971) from the small intenstine occurs with the involvement of specifically active transport systems capable of operating against the concentration gradient.

Distribution of Substances between the blood and tissues obeys the laws of free diffusion and active transport across membranes. Lipoid-dissolved substances access all organs and tissues but tend to form larger accumulations in the lipoid-rich tissues: omentum, bone marrow, and testes. One is well-familiar with the deposition of DDT, aldrin, hexachlorobenzene, and other chlorine derivatives of hydrocarbons. Such depots, however, are not passive and under certain circumstances, e.g. fasting, stress, etc. may release toxic substances into the blood.

Many pesticides tend to distribute themselves comparatively uniformly through the body, and only very early upon absorption can they, and especially those which combine with proteins, be contained in large quantities in the organs that occur some place along their routes of absorption or elimination. But, even with the relatively uniform distribution of most pesticides in the body tissues, one can still note some distribution preferences. The gamma-isomer of hexachloran spreads comparatively uniformly only in the fat, its higher concentrations in the kidneys and adrenals relative to other organs having been reported by Gadaskina and Filov (1971). Dieldrin accumulates higher concentrations in the fat-containing tissues (mesentery and others) and also in the liver and kidneys. More generally, OPP access virtually all organs.

The hematoencephalic barrier appears permeable to many pesticides. Organochlorine and organophosphorus chemicals pass over and through it so that dieldrin, for example, following its intravenous injection to mice, was discovered to have reached maximum concentrations in the brain within a short ten minutes. Nevertheless the hemato-encephalic barrier does retain the organophosphate chemicals whose molecule contains positive nitrogen or sulfur atoms (Kagan, 1977). Similarly, hexachlorocyclohexane (HCCH) isomers as well as DDT and other organochlorine pesticides are detected in the central nervous system (Wilkinson, 1976). Finally, organomercury

compounds reach the brain and affect various regions of the nervous system (Trakhtenberg, 1969).

The placental barrier is crossed by many OCP (Voronina, Pismennaya, 1973; Ackermann, Engst, 1970). Balashova et al. (1973) established in some uncharged organophosphorus inhibitors a potential of selective accumulation in the lung tissues.

The dipyridyl herbicide paraquat possesses the same property, assumed to be underlying its capacity to selectively affect the lungs (pneumosclerosis). The thyroid gland is a deposition site of dithiocarbamates and their metabolites. Wilkinson et al. (1976) reported selective accumulation of ethylenethiourea in the thyroid gland. Alexandrova (1975) identified a thiocarbamate herbicide, ronit, for the ability of fast distribution among all internal organs its higher concentrations detected in the lung, liver, and kidney tissues. Centrifugation of the hepatic homogenate has detected DDT and gamma-hexachlorocyclohexane, largely, in the supernatant fraction containing microsomes, and sevin in the nuclear fraction (Kuzminskaya, 1976). These distinctive features seem to have a bearing on the OCP induction of the microsomal enzymes and the ability of sevin to trigger adverse individual effects (gonadotoxic, mutagenic, etc.).

The kidneys constitute the primary organ for the **elimination** of pesticides from the body. Research on the urinary contents of pesticides and their metabolites can provide important diagnostic clues and in some instances may serve as a control to judge the efficacy of therapeutic measures. Many pesticides are secreted by the bile and excreted from organism in faeces. The blood is partitioned from the bile by a membrane which is well-permeable to many chemical substances whose bilious concentration may therefore match their concentration in the blood. On the other hand, bilious salts and certain of its polar components can conjugate pesticides to promote their excretion in the intestines and elimination in the faeces. If the conjugates are unstable and hydrolyzed in the intestines, the pesticides may be reabsorbed into the bloodstream (Wilkinson, 1976).

Some volatile compounds (halogen-containing hydrocarbons, e.g. methyl bromide and also hydrocyanic acid, etc.) are cleared by the lungs in expired air. Also detected in expired air are carbon disulfide, a metabolite of dithiocarbamates (Korablev, 1971; Svetly, 1969), and carbon tetrachloride which is there to recognize long after intoxication.

The factors to consider for the excretion of toxic substances by the kidneys include glomerular filtration and passive and active transport in the tubules. Filtration is accomplished in the glomeruli, the non-electrolytes' concentration in the filtrate being the same as in the plasma (Gadaskina, Levina, 1975). Although in the renal tubules a significant proportion of the non-electrolytes is reabsorbed into the blood, this is concurrent with the process of their passive and active transport by the carrier into the tubular urine. As a result, the concentration of the substances in the urine is controlled by the interrelation of these phenomena.

With respect to electrolytes and ionized compounds including the majority of toxic metabolites, their filtration into the primary urine is influenced in many ways by the strength of the bond with plasma protein. The filtration is more difficult in the case of substances tightly bound with albumins. Ionized compounds are immune from reabsorption in the tubules and fast-excreted from the body. Some of the pesticides in the ionized state, e.g. paraquat, are secreted by the transport system of organic cations. It is reckoned that the toxic effect of paraquat on the kidneys arises from a breach in the active transport processes.

There is diagnostic value in the urinary excretion of not only original pesticides, but their metabolites as well. The detection of paranitrophenol in the urine provides a sensitive diagnostic and exposure test for intoxications by the nitrophenyl esters of phosphorus acids thiophos, metaphos, and methylnitrophos. Leptophos metabolites — phenylphosphonic acid, O-methylphenylphosphonic acid, and O-methylphenylphosphonthionic acid — have been recognized in the urine and faeces. When carbamate pesticides are administered to the rat, their phenol metabolites are detected in the urine as well as the unaltered chemicals. Mercury detection in the urine, either in the cases of intoxication by organomercurials or upon injection of unithiol, serves as a diagnostic tool and a control for the efficacy of therapeutic intervention.

Persons exposed to hexachlorocyclohexane (HCCH) were identified as having alfa- and gamma-HCCH, traces of beta- and sigma-HCCH, hexachlorobenzene, pentachlorocyclohexane, pentachlorobenzene, pentachlorophenol, tetrachlorophenols, and trichlorophenols in their urine (Engst et al., 1976). Following rat poisoning with hexachlorobenzene, there was pentachlorophenol, tetrachlorohydroquinone, and pentachlorothiophenol detected in their urine and pentachlorophenol and pentachlorothiophenol in the faeces (Koss et al., 1976). When the animals were injected ^{14}C-labelled 2-methyl-4-chlorophenoxyacetic acid their urine was found to hold 92.26 percent and the faeces 6.76 percent of the incorporated radioactivity.

Excreted in breast-milk in animals as in humans are hexachloran, 2.4-D and its derivatives, chlorophos, valexon and also mercurial and arsenic compounds.

Pesticide Metabolism in the Body. Foreign substances (xenobiotics) are metabolized in the body in essentially the same ways as the substances natural to it. Xenobiotics undergo oxidative transformations, reduction, hydrolysis, and synthetic reactions to form twin compounds. The polar watersoluble substances that result from these conversions are commonly less toxic and removed from the body by urinary excretion (since they will not be reabsorbed to the blood in the renal tubules). This way detoxication occurs — the over-riding trend in the metabolism of xenobiotics, growing from evolution and having a protective and adaptive nature. Instances are documented, however, of the chemical transformations in the body acting to synthesize compounds more toxic than the original ones (the so-called

lethal synthesis). In this manner, for example, thionic esters of phosphoric acid turn as they oxidize into thiolic esters which are active anticholinesterase agents and highly toxic substances. Epoxides of the organochlorine pesticides of diene synthesis, aldrin and heptachlor, are more toxic and more carcinogenic, compared with the initial products. The processes need to be considered for effective management of preventive and therapeutic measures.

The effect of pesticides is caused as much by the parent substances that enter the body as by their metabolites formed therein (some effects may be due to their combined action).

In addition to pesticides, the body may be exposed to their toxic metabolites from the environment, notably through their uptake in ingested plant and animal foods. Accordingly, adequate provisions must be made in the design of diagnostic procedures and exposure tests in order to determine in the biological media (blood, urine) the parent pesticides as well as their metabolites. In the same vein, preventive and therapeutic programs must be aimed at increasing detoxication of the pesticides which enter the body and, if they metabolize on the pattern of lethal synthesis, at preventing the formation of highly toxic metabolites there.

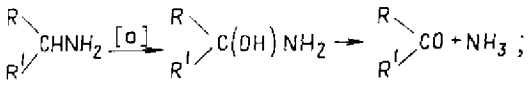
Though the liver is the principal body organ to metabolize xenobiotics the detoxifying capacity is inherent in other tissues as well (the kidneys, lungs, gastric and intenstinal walls, etc.). The organelles located in the liver cells contain a large amount of highly active enzymes, more or less importantly involved in the metabolism of chemical substances. The endoplasmic network (endoplasmic reticulum) is thought to be of special importance because it incorporates microsomal enzymes. In cell centrifugation, the latter persist in the microsomal fraction. Initially, at 10 thousand g and 10 min, the heavier cell parts, that is, the nuclei, cell fragments, and mitochondria are settled, and so are later, at 100 thousand g and one hour, the rest of the organelles, among them the rough (containing the ribosomes) and smooth reticulum. This cell fraction came to be known as microsomal and it is widely held at present that the microsomal enzymes play a large role in the metabolism of most pesticides (Doterman, 1971; Archakov, 1975; Kuzminskaya, 1976; Popov, 1977; Kagan, 1977; O'Brien, 1967; Park, 1973; Wilkinson, 1976).

Oxidative pesticide transformations catalyzed by mixed function oxidases (MFO). Mixed-function, or multi-purpose, oxidases are located in the microsomal fraction of cells. They are involved in the metabolism of steroids, lipids and polysaccharides in the synthesis of proteins, and the metabolism of numerous extraneous compounds. Mixed-function oxidases constitute an essential element in the biotransformation of most lipoid-dissolved compounds, pesticides included.

The microsomal cell fraction spans a broad selection of enzymes supporting many varied reactions. The smooth and rough endoplasmic reticulum is immediately linked with the activity of NADPN-cytochrome-C-reductase, NADN-cytochrome-b_5-reductase, glucoso-6-

phosphatase, cytochromes b_5 and P_{450}, esterase, and nucleoside phosphatase (Archakov, 1975). The microsomal fraction may additionally involve the monoaminoxidase and cytochromoxidase from mitochondrial membranes and the 5-nucleosidase from the plasmatic membrane, along with some soluble enzymes (Morelli, Nakatsugawa, 1975). Cytochrome P_{450}, the end-oxidase that perfoms hydroxylation, is an oxygen-activating enzyme for most oxidases. In the warm-blooded animals the enzyme is detectable in cell microsomes of the liver, kidneys, lungs, alimentary canal, skin, and placenta. Localization of mixed-function oxidases in the organs that occur along chemicals' pathways into the body affords protection against their toxic action, a feature developed in the process of evolution. The types of the MFO-catalyzed oxidative processes are as follows (adapted from Park, 1973).

Aromatic hydroxylation: $C_6H_5X \xrightarrow{[o]} HOC_6H_4X$; acyclic oxidation: $RCH_3 \xrightarrow{[o]} RCH_2OH$; O-desalkylation: $ROCH_3 \xrightarrow{[o]} ROCH_2OH \rightarrow$ $\rightarrow ROH + HCHO$; N-desalkylation: $RNHCH_3 \xrightarrow{[o]} RNHCH_2OH \rightarrow$ $\rightarrow RNH_2 + HCHO$; desamination:



sulfooxidation:

$RSR' \xrightarrow{[o]} [RS^+(OH)R'] \rightarrow RSOR' + H^-$.

These reactions take place in the presence of the reduced $NADPN_2$ coenzyme and oxygen. As a process, the microsomal oxidation of xenobiotics is competitively inhibited by substances themselves oxidized by the enzymes. This can explain the cases of potentiation as several toxics act jointly upon the body and one interferes with another's detoxification.

Aromatic hydroxylation presents an MFO-catalyzed reaction responsible for the metabolism of some insecticides. When acted-upon by MFO; naphthalene and other structurally similar insecticides are converted to epoxide, thereafter regrouped to 1-naphthol. The same metabolization pattern applies to sevin (4-hydroxy-1-naphthyl methyl carbamate, 5-hydroxy-1-naphthyl methyl carbamate; 5,6-dihydroxy-1-naphthyl methyl carbamate) and propoxur (Morelli, Nagatsugawa, 1975).

Aliphatic hydroxylation and *epoxidization* are essential to the metabolism of many pesticides containing alkyl groups. The latter's microsomal oxidation constitutes the prime mover for the metabolism of several carbamates, OCP, diene-synthesis insecticides (heptachlor, aldrin, isodrin, chlordane) and some pyrethroids. As shown by Krieger et al. (1976), the epoxidization of aldrin by monkey liver homogenates attains maximum velocity in $NADPN_2$ and oxygen are pre-

sent. The reactions of epoxidization and hydroxylation in rats and monkeys go much faster after an injection of phenobarbital. The rat liver microsomes, in the presence of $NADPN_2$, metabolize dieldrin into four metabolites. Phenobarbital and DDT boost the metabolic rate while the MFO inhibitor SKF525A slows it down. Phenobarbital in combination with fasting in rats activates MFO induction and contributes to the elimination of DDT and its metabolic products from the body (Lambert, Brodeur, 1976).

O-, S-, and N-dealkylation involves the stage of hydroxylation of the alkyl group conjugated with a heteroatom (oxygen, sulfur, or nitrogen). This pattern of metabolism holds for many of the pesticides structurally involving alkyls conjugated with the heteroatom. Hydroxylation of N-alkyl groups occurs in the metabolism of several carbamates (zectran and others) and also of nicotine. The carbamate propoxur is metabolized by O-alkylhydroxylation of the isopropyl group. Rotenon is likewise subject to O-alkylhydroxylation (Nakatsugawa, Morelli, 1975).

In the transformation of esters of thiophosphoric acids the lead role is played by *oxidative desulfuration*, the reaction in which mixed-function oxidases are involved and which leads to lethal synthesis because of the much higher anticholinesterase activity and toxicity of the oxidized organophosphate derivatives. Phosphamide and dursban provided Popov et al. (1977) with suitable examples to demonstrate their oxidative desulfuration during the in situ perfusion of an isolated rat liver. Parallel to incremental growth of the perfusate content of P=O analogs there was a marked inhibition of acetylcholinesterase. A mathematical model has been proposed to simulate dynamics in the disappearance of phosphamide and dursban over time.

The influence of MFO induction and inhibition on phthalophos metabolism in the in situ perfusion of an isolated rat liver was ascertained in the laboratory by workers from the National Research Institute of Hygiene and Toxicology (Popov, Kagan, Voronina, Pismennaya, 1980). The MFO induction has been found to speed up and its inhibition to slow down the disappearance of phthalophos from the perfusate (Fig. 2). Accordingly, the MFO induction accelerates and its inhibition decelerates the accumulation rate of phthalophos metabolites. The anticholinesterase activity of phthalophos diminishes with both, the induction (by faster detoxification) and inhibition of mixed-function oxidases (by slower oxidative activation). Occasions arise, however, when the enhanced MFO activity fails to increase the toxicity of the OPP compounds subjected to lethal synthesis, and vice versa. The disparity of the effects produced by the inductors and inhibitors of mixed-function oxidases seems to be varying with their respective roles in the activation and detoxification of OPP.

In most cases a rise in the MFO activity promotes the detoxication of even those OPP compounds which are susceptible to lethal synthesis. Zlatev (1976) discovered in experiments with the induction of mixed-function oxidases by the organochlorine acaricide milbex that it lowered the toxicity of several thiophosphates. Another factor mak-

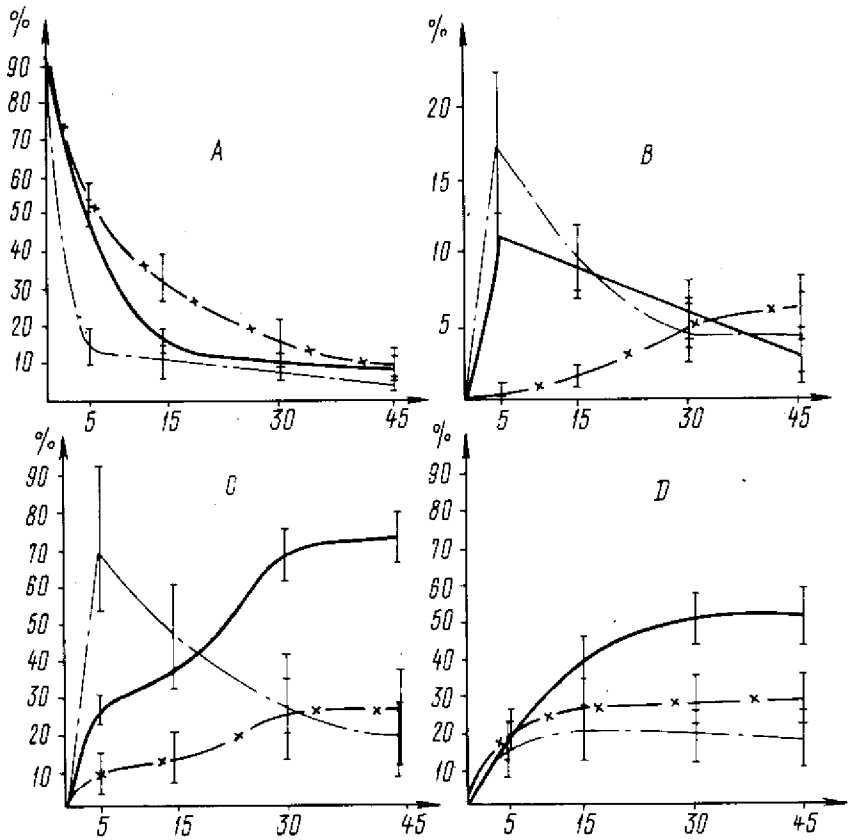


Fig. 2. Influence of MFO induction and inhibition on phthalophos metabolism:
A — phthalophos concentration in perfusate; *B* — oxymethylphthalimide concentration; *C* — phthalimide concentration; *D* — degree of inhibition of ChE activity; - - - MFO induction, -X-X- MFO inhibition, ——— control

ing the detoxification possible is the microsomal oxidases, way of functioning in conjugation with the enzymes of secondary metabolism which ensures rapid conjugation and elimination of the oxidized metabolites from the body.

The system of mixed-function oxidases constitutes an organism's principal mechanism of biochemical adaptation to the action of toxic substances. Numerous investigations have proved that substances which provide substrates for the system stimulate, at the same time, its activity mainly by intensifying the synthesis of cytochrome P_{450}. The classic inductors for mixed-function oxidases include phenobarbital, polycyclic aromatic hydrocarbons, organochlorines, and steroid hormones. It has been established that DDT, lindane, mirex, chlordane and hexachlorobenzene can double or treble the level of cytochrome P_{450} in rats. Kuzminskaya et al. (1976) reported that administration

of three organochlorine pesticide doses to rats (DDT, gamma-isomer of HCCH, and polychlorocamphene) at the rate of 1/5 LD_{50} induced a much higher activity in the liver of enzymes involved in the demethylation of aminopyrine. Conversely, their action was inhibited by tetramethyl thiuramdisulfide (TMTD).

Kagan, Ovsyannikova, and Khomenko (1979), with the aid of EPR-spectrometry, discovered that, following the induction of cytochrome P_{450} and accelerated detoxification, polychloropinene (PCP) and polychlorocamphene (PCC) induce neither paralysis nor the development of pareses in rats, but do cause paralyses when administered to hens and guinea pigs. At the adequate level for the development of paralyses the activity of cytochrome P_{450} is reduced. This is of interest in explaining the causation of different animal species sensitivities to the neurotoxic effect of chemicals.

Feeding rats a diet containing 0.2 percent of hexachlorobenzene has given rise to hepatic porphyria, attended with increased anilinhydroxylase activity and a heightened concentration of cytochrome P_{450} in the microsomal fraction of liver homogenates.

Dietary intake of comparatively minor OCP quantities leads to the induction of mixed-function oxidases. In some instances, the OCP prove stronger MFO inductors than either methylcholanthrene or benzpyrene (Mullen et al., 1966), though less active than phenobarbital. Anticholinesterase insecticides (OPP and carbamates) are poor inductors of mixed-function oxidases though sevin was seen to stimulate their induction when administered in three 1/5 LD_{50} doses (Stevens et al., 1973). It is known that the activity of cytochrome P_{450} is pushed up by pyrethrum, methylendioxyphenyl and other pesticides. Quite likely some pesticides, while not direct MFO inductors, can potentiate their activity via the hormonal system.

MFO-independent transformation of pesticides. Enzymatic hydrolysis is a fairly common type of transformation for many of the pesticides in the ester category. It represents also one of the major OPP detoxification pathways. Depending on the latter's structure, hydrolysis may be predominantly phosphatase, amidase or carboxyesterase by type (Kagan, 1977).

Carboxyesterase hydrolysis is important to metabolism of pyrethroids and some carbamates (Casida, 1973). In addition to oxidative processes, reducing reactions are also involved in pesticide metabolism. The nitrogroup, known to reside in the benzene ring of some organophosphorus pesticides like thiophos, metaphos, and methylnitrophos, is reduced to an aminogroup with the aid of nitroreductase. The latter has been identified in the liver of mammals, birds and fish. The process has major importance in ruminant mammals whose intestinal bacteria reduce nitrocompounds with remarkable vigour.

One of the way in which pesticides get detoxified is conjugation with sulfur, glucuronic acid, glucose, and amino-acids, as well as some other synthetic processes giving rise to polar metabolites readily disposable by urinary excretion. In fact, many chlorinated hydrocarbons yield when metabolized hydroxy- and other functional groups

capable of conjugation. The gamma-isomer of hexachlorocyclohexane is metabolized in the alkaline medium through conjugation with SH-glutathion, leading to the synthesis of thiophenols. Dehydrochlorination of hexachlorocyclohexane is also feasible with the involvement of dehydrochlorinase, to form a variety of isomers (tetra-, tri-, and dichlorobenzenes and their appropriate chlorophenols) eliminable from the body as conjugates with glucoronic and sulfuric acids and glutathion. Though the exact sequence of these developments is not clearly defined it is hypothesized that the enzymes involved include mixed-function oxidases, uridine diphosphate (UDP), glucuronyltransferase, sulfotransferase, L-SH-S-aryltransferase and other enzymes. In DDT metabolism synthesis of the conjugates is likewise probable.

That the reactions of conjugation make part of the mechanism for the metabolism of carbamates and phosphorus compounds is beyond question. Products of the conjugation of hydroxylated sevin metabolites with sulfuric and glucuronic acids were detected in mammals. OPP also produce twin compounds with sulfuric and glucuronic acids in the presence of uridine-phosphatglucuronic acid with the help of glucuronyltransferase, an enzyme found in the microsomal fraction of the liver. Glucuronic acid is capable of association with phenol and hydroxylamine groups, hydroxylic groups of alcohols, and carboxyl-, sulfhydryl-, amino-, and iminogroups of various compounds. Sulfates add to aromatic aminogroups, phenol hydroxyl and the alcohols of the aromatic series. These reactions are catalyzed by arylsulfotransferase which is responsible for the transfer of sulfate groups (Doterman, 1972). Enhanced formation of glucuronides has been proved for the warm-blooded organisms exposed to chlorofenvinphos, gardona, DDVP, and parathion. Sulfuric acid conjugates with colep and famphur were defined and the synthesis of glucosides in plants was reported from metabolic studies of abate and phosalone. Excreted as glucuronides in the rat urine are hydroxylation products of pyrethrines (Casida, 1975) and other insecticides of plant origin (rotenoids) as well as some hormonal insecticides. G-SH-S-transferase is involved in the metabolism of thiocyanites. Aminoacid conjugation results in glucuronides, glucosides, and sulfates metabolizes a number of insecticide synergists. The conjugation is commonly anticipated by the oxidation of methylendioxyphenyl groups or a side-chain. The primary physiological role attributed to the conjugation of chemicals is detoxifying them and exploiting the polarity and water-solubility of their metabolites to efficiently clear them from the body. Detoxifying the substances brought in from outside is not the only activity aided by these mechanisms, as they apparently regulate in the elimination of an axcess of biologically active compounds, particularly steroids.

Isomerization, another type of pesticide transformation in the body, is common to several OPP. Both in living organisms and in the environment isomerization may be initiated and maintained by the influence of physical and chemical factors. Isomerization may be looked upon

as a type of lethal synthesis (Kagan, 1977). Reactions of isomerization are known for many thiophosphates.

Light influences metabolism of some pesticides. The effect of ultra-violet rays on thiophos results in several active cholinesterase inhibitors. Ultra-violet radiation gives rise to sulfoxides and sulfones (timet, intrathion, etc.). OPP transformation into more toxic compounds may be a result of transalkylation. Some workers (Kundiev, et al., 1975; Altareva, 1977; Dunaisky, 1978) hypothesize that polychloropinene, as it metabolizes in soil under the action of ultraviolet rays, temperature and other factors (perhaps as a result of their interaction with mineral fertilizer), forms more toxic substances capable of triggering intoxications. Since polychloropinene represents a complex mixture of numerous ingredients, the identification of its toxic metabolites presents a very difficult problem.

MECHANISM OF ACTION AND TOXICODYNAMICS OF PESTICIDES

As the substances endowed with chemical reactivity enter the body they begin to react with biochemical components of the cells and fluids there. The result is a complex set of reactions working to modify the functions and structure of internal organs and the organism as a whole. In studying the mode and mechanism of action of the chemicals just described one should necessary address their toxicodynamics. In so doing, one will most probably observe not just the primary toxicological response but also its sequelae, as well as the developments associated with the action of metabolites of these primary substances which may and sometimes do set off a pathological process. This action is essentially determined by characteristics of the toxicant, notably its structure and physical properties and its amount, on the one hand, and the state of the organism's defence and adaptation reactions and the functioning level of its biochemical, physiological, and immune systems, on the other. The mechanism of action and the toxicodynamics of chemical substances need to be investigated if maximum effective techniques are to be devised for prevention and control of the pathological processes likely to be caused by them.

Primary Toxicological Response (by analogy with primary pharmacological response) results as the toxic substances that entered the body react with biological macromolecules. The reaction involves, on the one hand, the molecules of the toxic substances and, on the other, enzymes, structural and functional proteins, lipoproteids, nucleic acids and other biomacromolecules.

In order to get to the heart of this reaction, one must have a detailed knowledge of the structure and physico-chemical properties of the reacting molecules; the pattern and properties of the bonds that may link the atoms and molecules of substances both foreign and native to the organism; the kinetics of the substances' interaction

with enzymes and receptors; and the latter's functioning mechanism. Of key importance to the primary toxicological reaction is the interaction of toxic substances with enzymes and other protein receptors. According to Fischer's theory, the enzyme and substrates possess a rigid spatial structure and must fit each other. The last few years have seen growing acceptance of the notion about a flexible structure of active enzyme centers; in the course of conjugation with the substrate the structure modifies the spatial disposition of atom groups, or conformation (Koshland, 1964). Specifically regarding the active enzyme centers, their conformation can be changed by the effect of coenzymes, substrates, and inhibitors. Similar conformational shifts occur in other protein receptors. Pauling (1961) theorized that narcotic drugs displace from cerebral cell proteins the molecule of water which forms microcrystals. This is said to interfere with the conformation of protein molecules, the patent cause of narcosis.

Inhibitors may be different in their mechanism of action. Webb portrays it as follows: the inhibitor may join the apoenzyme thus precluding its enteraction with one of the substrates, with the coenzyme, and with the second substrate; the inhibitor may attach to the site of the apoenzyme molecule located near the active center and thereby prevent its combining with the substrate; the inhibitor may prevent the enzyme's reaction with the substrate sterically or electrostatically, thus altering the conformation of the protein bound also with the coenzyme, substrate, and apoenzyme; with the first or second substrate or the cofactor.

The enzyme-inhibitor interaction results first in a reversible enzyme-inhibitor complex:

$$E + I \underset{K_{-1}}{\overset{K_{+1}}{\rightleftharpoons}} EI, \text{ where}$$

K_{+1} is the constant for the complex-forming rate, and
K_{-1} is the constant for the rate of the inverse reaction.

The ratio K_{+1}/K_{-1} influences the constant for reversible inhibition. In irreversible inhibition, the inverse reaction rate is equal to zero, that is, the reaction is going in one direction only. In practice however K_{-1} is not equal to zero, even for irreversible inhibitors like e. g. OPP cholinesterase inhibitors; in other words, the enzyme-inhibitor complex is slowly dissociating. The enzyme inhibition processes may progress in several stages, as follows:

$$E + I \underset{K_{-1}}{\overset{K_{+1}}{\rightleftharpoons}} EI \xrightarrow{K_{+2}} EI' + P_1 \xrightarrow{K_{+3}} E + P_2.$$

First, the reversible enzyme-inhibitor complex (EI) takes form; then the complex's structure changes (EI') and the reaction product is released (P_1); or else the reaction may involve segregation of the enzyme (E) and release of a second reaction product (P_2). Kinetic characteristics underlie the classification of chemical reactions into

zero-order, when the reaction rate is constant and independent of the substance concentrations involved; first-order, when the reaction rate is proportional to the concentration of one of the substances; and second-order, when the reactions rate is proportional to the product of the concentrations of two reacting components. The number of the interacting molecules idensifies the reactions as monomolecular, bimolecular, and trimolecular. The enzyme conjugation with the inhibitor constitutes a bimolecular reaction. If, however, it occurs with a significant excess of the inhibitor it may be kinetically described as pseudomonomolecular. An illustrative example of the reaction is cholinesterase inhibition (phosphorylation) by OPP inhibitors. The constant for the phosphorylation rate was arrived-at from the following equation (Yakovlev, 1965):

$$K_2 = \frac{1}{t[I]} \ln \frac{V_0}{V_t}, \text{ where}$$

V_0 is the rate of enzymatic hydrolysis of the substrate with no inhibitor; and

V_t is the rate of hydrolysis t minutes after the enzyme incubation with the inhibitor. The inhibition rate constants present quantitative characteristics of OPP anticholinesterase activity. To start any chemical reaction requires energy expenditure to trigger molecules into an activated state (activation energy). Enzymes as well as other catalysts reduce free activation energy and make the chemical reaction easier to carry on. The major feature of the enzymatic reactions is their fine susceptibility to the concentration of the substrate. At a low substrate concentration, the reaction rate increases as the substrate concentration (the first-order reaction) but as it becomes greater the reaction rate grows progressively slower and the reaction already becomes one of a mixed order. Further on, the reaction rate becomes constant and no longer dependent on the substrate concentration (the zero order). Michaelis and Menten derived an equation to relate the enzymatic reaction rate (V) to the substrate concentration [S]:

$$V = \frac{V_{max}[S]}{K_m + [S]}, \text{ where}$$

V_{max} is the maximum reaction rate, and

K_m is the constant numerically equal to the substrate concentration at which the reaction rate is a half of its maximum velocity.

The Michaelis-Menten equation can be converted to a form convenient for graphic representation of empirical data. The inverse values of the left- and right-hand parts are equated to each other: $\frac{1}{V} = \frac{K_m + [S]}{V_{max}[S]}$. The resultant expression $\frac{1}{V} = \frac{K_m}{V_{max}[S]} + \frac{1}{V_{max}}$, known as Lineweaver-Burk's equation, is used to construct a graph by a method of double inverse values and thus determine V_{max} (Fig. 3). The graph is a ready source of information about the pattern and mechanism of enzyme inhibition.

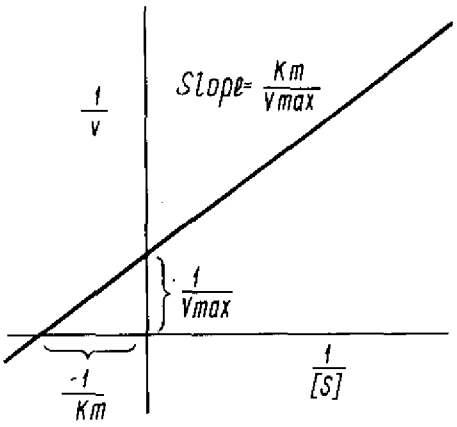


Fig. 3. Graphic representation of Lineweaver-Burk's equation

Enzyme inhibition may be reversible or irreversible. Under irreversible inhibition a change occurs in the structure of the enzyme functional groups that makes it lose its catalytic activity (e. g. phosphorylation of the esterase center of cholinesterases in intoxication by organophosphorus inhibitors). Reversible inhibition is either competitive or non-competitive. An inhibition is called competitive when an increase in the substrate concentration decreases the extent of inhibition. When its alteration has no effect on the extent of inhibition, it is non-competitive. Quantitative evaluation of a particular inhibition type makes use of the Lineweaver-Burk graph. In competitive inhibition, all lines cut the coordinate axis at one point, that is, the value of V_{max} does not change with or without competitive inhibitor; in non-competitive inhibition the lines intersect it at different points. The former can be exemplified by the inhibition of succinate dehydrogenase by malonic and other dicarbonic acids. The inhibitory effect of malonate is reduced by succinate, the substrate of the enzyme. The extent of the inhibition depends on the succinate/malonate concentration ratio. An example of non-competitive inhibition of enzymes is the action of the substances reversibly binding SH-groups (ions of mercury, silver and copper). If metal ions (iron, magnesium, etc.) are needed for the enzymes to become active the substances making complexes with them, as for example cyanides with iron or ethylendiaminotetraacetate (EDTA) with magnesium and other bivalent cations, are non-competitive inhibitors for the enzymes.

Along with reversible and irreversible inhibition, researchers have identified also a combined type. Brestkin et al. (1971) showed that some OPP possess the property of AChE inhibition both reversible and irreversible. The latter is associated with phosphorylation of its esterase center and the former arises by the interaction of the inhibitor with the site on the enzyme active surface responsible for AChE inhibition by the excess substrate. The reversible inhibition of AChE and BuChE (butyrylcholinesterase) by some organophos-

phorus compounds is explained by the authors in terms of a steric obstacle to emergence of the Michaelis enzyme-inhibitor complex. The enzyme-inhibitor complex thus formed in lieu of the Michaelis complex is similar to that which originates from the action on cholinesterase of reversible inhibitors, for example tetraalkylammonium ions.

Classification of Pesticides by Their Mechanism of Action. The action of a toxic substance on the body begins with its primary interaction with appropriate biochemical structures, first of all enzymes. Several attempts at classifying toxic substances according to their principal mechanism of action have been undertaken (Pokrovsky, 1962; Tiunov, 1963) and it is from this perspective that the major groups of pesticides in current agricultural use will be discussed in the present section.

Structural Analogs of the Substrate. Substances structurally similar to enzyme substrates can compete with them for the interaction with the enzymes and competitively inhibit their activity. A clear example of that comes from anticholinesterase compounds, notably the organophosphate and carbamate pesticides. The latter may be viewed as complete structural analogs of acetylcholine if the interaction occurs via the esterase and anion center or, as is the case more often, incomplete analogs, if it goes via the esterase center alone. As well as acetylcholine, organophosphate and carbamate compounds can acylate serine hydroxyl in the esterase center of cholinesterase. The reaction with acetylcholine results in an enzyme acetylated at the esterase site and choline sorbed on the anion center. The latter is soon desorbed and the deacetylation of the enzyme esterase site fosters rapid recovery of its original structure. Cholinesterase has a high number of revolutions (molecular activity), that is, the number of the substrate molecules capable of splitting one enzyme molecule per minute. The number of cholinesterase revolutions can be estimated for the enzyme active center, rather than the protein molecule, to express the activity of the catalytic center.

The conjugation of phosphate and carbamate pesticides with cholinesterase is a result of their structural similarity to acetylcholine. The effect of organophosphates and carbamates devoids cholinesterase of its inherent catalytic activity. But unlike the organophosphates, carbamates react with cholinesterase to form less stable, carbamyolated enzymes. Monomethyl- and dimethyl-carbamyolated AChE at pH 8 and the temperature of 25°C has a half-life of 30 and 56 minutes (Aldridge, 1972). Other types of carbamyolated AChE are more stable. The deacetylation rate in the AChE reaction with OPP and carbamates is on average 10^5 to 10^6 times less rapid than in its reaction with acetylcholine.

Precursors of structural substrate analogs. This group encompasses the substances which emerge as structural analogs of substrates in the course of metabolism. These are, for example, the esters of thio- and dithiophosphoric acids which undergo oxidative desulfuration in the body to yield P=O analogs — active anticholi-

nesterase agents. Much more biologically active than the initial P=S compounds, the P=O analogs can produce lethal synthesis. Compounds of the octamethyl type are transformed in the organism into more active phosphoramidoxides.

Another, classic example of precursors of structural substrate analogs derives from fluoracetates, currently applied in agriculture as rodenticides. Fluoracetic acid, with the involvement of the coenzyme A, enters into a condensation reaction with oxaloacetic acid to form fluorocitric acid, a structural analog of citric acid and an inhibitor of aconitase, one of the essential enzymes in the Krebs cycle. By inhibiting aconitase, fluorocitric acid makes the enzyme-substrate complex strong enough to be immune to assault by the enzyme. The intoxication by fluoracetates has a characteristic latent period associated with the processes of lethal synthesis.

Toxics interacting with coenzymes. These substances are collectively termed antivitamins. Pyridoxine (Vitamin B_6) derivatives are the more important among the coenzymes for their catalytic role in the various conversions of aminoacids; they act, in particular, as the coenzymes of aminotransferases to catalyze the transport of aminogroups from substrate to substrate. A number of pyridoxal kinase inhibitors are known that modify the process of pyridoxine phosphorylation. They include hydrazine, semicarbaside, and isonicotinyl hydraside. Faulty synthesis of pyridoxalphosphate is responsible for the inhibition of enzymes: aminotransferases, decarboxylases, kynureninases, and others (Tiunov, 1963). Some pesticides are also known to hold down the activity of aminotransferases.

Derivatives of folic acid can also act as coenzymes, one example being tetrahydrofolic acid involved in the synthesis and transport of hydroxymethyl groups. Dihydrofolic acid complexes with methyl groups and transmits them to the acceptor.

One consequence of this is that the processes of methylation and hydroxymethylation proceed with the involvement of enzymes for which folic acid derivatives act as their cofactor. Substitution of an amine group for the oxygroup in tetrahydrofolic acid gives rise to diaminomethylpterine (aminopterine), a potent inhibitor for the enzymes containing tetrahydrofolic acid. Antifolic activity has been found to play a lead role in the mechanism of action of herbicides (derivatives of symmetric triazines) (Okunev, 1968). Nucleotides are also involved in metabolism, as consisting of a nitrogenous base, pentose and a phosphoric acid residue. The nitrogenous bases may be either purine (adenine, guanine) or pyrimidine (cytosine, uracil, and thymine); the pentose represents, in effect, either ribose or desoxyribose. Nucleotides contribute to the synthesis of adenosinetriphosphate and other macroergistic compounds that provide a source of energy for cellular metabolism. There are some structural analogs of nitrogenous bases (e. g. benzimidazole and its derivatives), pyrimidine (e. g. triazole-pyrimidines) and of adenine as well (e.g. diaminopurine). Of these, benzimidazole and pyrimidine have been the basic ingredients to develop several agents with pesticidal properties (Mel-

nikov, 1974). Those presently applied as fungicides are: benomyl (benlate, fundazole), BMC — methyl-N- (2-benzimidazole) carbamate, and others. They have been discovered to be teratogenic.

The coenzyme A aids in the transport of acetyl as well as other acid radicals. It is additionally and strongly involved in the metabolism of fatty and aminoacids. Its active group, the SH-group of mercaptoethylamine, adds the corresponding acyl. With the addition to it of an acetic acid radical, the acetylcoenzyme A will result. The compounds blocking the SH-groups (mercury, arsenic, and other chemicals) inhibit the enzymes containing the coenzyme A.

Nicotinamide (pyridine) coenzymes — nicotinamidadenindinucleotide (NAD^+) and nicotinamidadeninedinucleotide-phosphate ($NADP^+$) — are present in the composition of the dehydrogenases catalyzing electron transport. Kuzminskaya (1976) found them to play a major role in the toxicity mechanism of quite a number of pesticides. A typical feature of DDT exposure, for example, is inhibition at the transport site of NAD^+ dependent electrons.

The plant insecticide rotenon and the insecticidal antibiotic pyricidine A inhibit the respiration of nerve and muscular cells by inhibiting the enzymes involved in electron transport. Rotenon is a specific inhibitor for pyridine-nucleotide-containing oxidase and pyricidine an inhibitor for NAD-N-dehydrogenase (Fukami, 1976). Some insect species owe their resistance to rotenon and pyridicine to their having quinone, the agent enabling electron transport to bypass some enzymes of the respiratory chain. The insecticidal effect of these agents can be abated by vitamin K_3, a quinone derivative, found to have restored breathing in rotenon-inhibited mitochondria of the carp liver (Fukami, 1976).

Catalase and peroxidase are ferroporphyrine coenzymes that catalyze the oxidation of several compounds by hydrogen peroxide. The coenzymes form part of the cytochrome system, otherwise composed of several cytochromes and cytochrome-C-oxidase which activates molecular oxygen. Cytochrome-C-oxidase constitutes the end-enzyme of the respiratory chain, its inhibition brings on impairment of electron transport. Most of cytochrome-C-oxidase inhibitors conjugate with the enzyme's hemic part containing diatomic iron. Among the classic inhibitors of cytochrome-C-oxidase are carbon monoxide, cyanides, nitriles, hydrogen sulfide, and azide. Some pesticides, e.g. carbophos, also demonstrate the capacity to inhibit citochrome-C-oxidase (Fukami, 1976). Yet the mechanism for the interaction of various substances with metal-containing porphyrine enzymes is different. While some of them bind metals in the enzymes, as do cyanides, carbon monoxide, nitriles, and dithiocarbamates, others interfere with the synthesis of porphyrines (aminotriazole). Diethyldithiocarbamate, tetramethylthiuramdisulfide and other dithiocarbamates identified for use as fungicides are remarkably active in binding copper-containing enzymes. Dithiocarbamates were shown to have an inhibitory effect on the activity of ceruloplasmine (Korablev, 1971; Svetly, 1971).

Toxics interfering with protein synthesis. This group includes structural analogs of aminoacids, several antibiotics and a number of other complex organic compounds. Of these, chloramphenicol, tetracyclines, phosphonomycine, D-cycloserine, penicillins, etc. are specific inhibitors for protein synthesis (Pokrovsky, 1962; Gaia et al., 1972). This mechanism underlies also the effects of the antibiotics employed in agricultural uses.

Toxics reacting with functional groups of proteins. These substances interact with thiol, amino-, and carboxylic groups of proteins. Conjugation of SH-groups is the key factor in the mechanism responsible for the action of arsenic- and mercury-containing compounds, not least of organomercurials (Medved, 1961). Arsenic and mercury complexes with dithiols are the strongest of their kind and this is the basic principle for current applications of the BAL (dimercaptopropanol) and unithiol antidotes containing two adjoining SH-groups in their molecule.

The thiol protein groups bind also a variety of alkylating agents such as halidealkylamines, some carcinogens and mutagens.

Uncouplers of oxidative phosphorylation. Oxidative phosphorylation, the process which ensures the synthesis of macroergistic phosphate compounds, relies on mitochondria-localized enzymes to carry it through. Substances structurally impairing mitochondria or causing them to swell, uncouple the enzymes implicated in certain stages of oxidative phosphorylation. Dinitrophenol is a classical uncoupler of oxidative phosphorylation and the similar toxicity mechanism resides in some herbicides such as the dinitrophenol derivatives dinitroorthocresol (DNOC), dinitrobutylphenol (dinoseb) and pentachlorphenol (Tiunov, 1963). Salicylanilides are in this category, too. Disruption of oxidative phosphorylation also has a certain role to play also in the toxicity mechanism of DDT and sevin (Kuzminskaya, 1976). It has been known for some time that the OPP having a phenol radical in the molecule — thiophos, metaphos, and methylnitrophos — can inhibit oxidative phosphorylation if present in comparatively high concentrations (Fukami, 1976). Uncoupling agents are capable of speeding the ATP degradation rate by stepping up the activity of ATP-ase. Their structural characteristic is the presence of an anion and lipophilic group. Aside from nitrophenol compounds and halogen-containing hydrocarbons, the similar effect has a role in the mechanism of action known for the synthetic pyrethroids of some antibiotics and unsaturated fatty acids (Fukami, 1976).

Toxics denaturing protein. Enzymatic activity and functional protein properties may become irregular as a consequence of changes in the secondary, tertiary, and quaternary protein structures. Among the substances one can cite as examples of the mild denaturing agents which impair the tertiary protein structure there are urea derivatives, some of them active herbicides: dichloralurea, diuron, arezine, herban, dicuran, dosanex, and kotoran. An altered enzymatic activity during denaturation comes about because of the spatial uncoupling of functional protein groups.

Toxics interacting with hormones or affecting hormone formation. Thiourea, uracil and its derivatives, and dithiocarbamates inhibit the synthesis of the thyroid hormone (Tiunov, 1963; Korablev, 1971; Svetly, 1971). DDD, and ortho-, para- isomer of dichlorphenyldichloroethane, is a specific inhibitor for the function of the cortical substance of the adrenal glands (Komissarenko, 1977). The mechanism of action for this DDT metabolite hinges decisively upon acutely reduced secretion of corticosteroids. DDD's high selectivity of adrenocorticolytic action has been key to its effective use as a chemotherapeutic agent to treat cancer of the cortical substance of the adrenal glands and Itsenko-Cushing's disease.

Toxics of biological origin. The substances referred-to here are snake venoms and insect poisons as well as several other toxicants. Their mode of action emphasizes the role of the enzymes that are parts thereof.

SELECTIVE TOXICITY IN PESTICIDE TOXICOLOGY

The principal aim of the pesticides is to destroy harmful insects, causative agents of plant diseases, weeds, and so on. It is only natural that such an effect can be inferent only in substances that are highly biologically active. Yet their practical use demands of the substances to possess a certain measure of selectivity in their action. Their toxicity to man, useful animals, insects, and plants must therefore be many times less than to the organisms they are intended to control.

The principles adopted in the development of agents with selective toxicity are being applied on a wide scale in contemporary medicine and agriculture. A difference between pharmacology and chemotherapy is that the drugs employed should be able to exert selective action on different target cells of the same organism in the former case and on different organisms in the latter case. If seen in this light, pesticide applications may be placed into the realm of chemotherapy (Albert, 1971). The development of selective insecticides is more of a problem than identifying selective chemotherapeutic agents to control bacterial infections. This is because the insects are phylogenically closer to man than the microorganisms and so divergencies in their biochemical processes are more difficult to find out. But, while a chemotherapeutic agent is intended for the destruction of microorganisms inside human body, a pesticide operates in the environment and must be prevented from gaining access into the body through mechanization of jobs, application of personal protective equipment, and by various other means. It is a more challenging task to block the way to pesticide penetration into food, water, and ambient air and thereby to avoid their adverse health effects.

Selective toxicity is either ecological or physiological (O'Brien, 1964). Selectivity is defined as ecological if a species is not susceptible to a chemical agent though not because it has no intrinsic sensitivity to it but because it avoids the exposure. Physiological selectivity featu-

res reliance on physiological (biochemical) mechanisms; it is further divided into the selectivity associated with different absorption patterns of substances in the body and true, or internal, selectivity. In the latter case, a poison moves into the body but is quickly metabolized and eliminated or not allowed to assault the sites responsible for the toxicological response.

The hygienic measures accepted in the regulation of chemicals' application rates and application schedules and the use personal preventive equipment effectively support ecological selectivity. Similarly, control of crop pests through the use of their natural predators or other means of biological protection is also managed under the principle of ecological selectivity.

The present use of chemicals producing a pesticidal effect but fast-destroyed in the warm-blooded organism and totally ineffective; even if they manage to get inside the body grew out of the clear current perspective on the mechanisms of physiological selectivity.

Pesticide toxicology today encompasses a wide array of selective insecticides, acaricides, fungicides, herbicides, growth regulators, and other plant protection (pest control) agents. Of the organophosphate and organochlorine insecticides, acaricides, and synthetic pyrethroids that have been synthesized to date, some are extremely important toxics for animals and human beings. The former, highly toxic fungicides (organomercurials) have been replaced by moderately and slightly toxic dithiocarbamates, derivatives of urea, guanidine and phthalimide, and heterocyclic compounds.

Distinction is made between selective sensitivities that are specific to the species, sex, age, individual and other features of the organism concerned.

Species-specific Selectivity. On a quantitative basis, the degree of selective toxicity of insecticides to various animal species can be measured using the so-called selectivity coefficient the ratio of the lethal doses (LD_{50}) for the respective organisms. Pesticide toxicology examines it in two aspects. The first is the ratio of the insecticide LD_{50} for the warm-blooded animals to the LD_{50} for the insects it is meant to destroy. The higher the coefficient, that is to say, the larger the difference between the chemical's toxicities for these animals, the greater its selectivity and the more safe its use. The second aspect is the relationship between the sensitivities of different laboratory animal species to the pesticide of interest. Its investigation s all-important for the extrapolation of animal data to humans. Relative to various chemical compounds, the issue was researched by Ulanova (1971), Krasovsky (1973), and Roll (1973). Its discussion here will be circumscribed to pesticides alone.

Table 3 provides selective toxicity data on some insecticides for the housefly relative to warm-blooded animals. Octamethyl, nicotine, aldicarb, carbaryl and carbofuran are more toxic to rats than houseflies and therefore present selective mammalicides. Sulfotepp is not selectively toxic while endrin, mevinphos, demeton, propoxur, azinphos-

Table 3

Selectivity Coefficients of Some Insecticides

Insecticide	LD_{50}, mg/kg		$K = \frac{LD_{50}\text{ for w.-b.}}{LD_{50}\text{ for h. f.}}$	Source
	for mice or rats	for house-flies		
Octamethyl	42	1932	0.022	Hollingworth, 1976
Nicotine	20	500	0.04	»
Aldicarb	0.6	5.5	0.11	»
Carbaryl	500	>900	<0.56	»
Carbofuran	4	4.6	0.87	»
Sulfotepp	5.0	5.0	1	»
Endrin	7.5	3.15	2.4	»
Mevinphos	3.7	1.5	2.5	»
Demeton	2.5	0.75	3.3	»
Propoxur	86	25.5	3.4	»
Azinphosmethyl	11	2.7	4.1	»
Toxaphene	80	11	7.3	»
Chlorfenvinphos	13	1.4	9.3	»
Thiophos	3.6	0.9	4.0	»
	14.5	1.25	11.6	Kagan, 1974
Carbophos	450 *	28	16	»
	1000	26.15	38	Hollingworth, 1976
Metaphos	35 *	1.25	28	Kagan, 1974
	24	1.2	20	Hollingworth, 1976
Aldrin	38	1.5	25	Brooks, 1976
Acetophos	250 *	6.5	38	Kagan, 1974
Acethion	820 *	21	39	»
Dieldrin	46	1	46	Brooks, 1976
Chlorophos	730 *	9.4	78	Kagan, 1977
Methylacetophos	1020 *	12	85	»
Methylnitrophos	470 *	5	94	»
DDT	118	2	59	Hollingworth, 1976
Allethrin	920	15	61	»
Abate	13,000	205	63	Brooks, 1976
Chlordane	457	7	65	Hollingworth, 1976
Heptachlor	162	2.25	72	Brooks, 1976
	100	1	100	»
Diazinon	285	2.95	97	»
Fenthion	245	2.3	107	Hollingworth, 1976
Lindane	91	0.85	107	»
Phosphamide	135 *	1	135	Kagan, 1977
Fenitrothion	570	2.3	248	Hollingworth, 1976

Continued

Insecticide	LD_{50}, mg/kg		$K = \frac{LD_{50}\text{ for w.-b.}}{LD_{50}\text{ for h.-f.}}$	Source
	for mice or rats	for house-flies		
Bromophos	1370	3.2	541	»
Alodane	>15,000	15.5	>968	»
Dihydroheptachlor	5,000	3.75	1333	»
Metoxychlor	6,000	3.4	1765	»
Permethrin	1500	0.7	2143	»
Phoxim	8,500	2.3	3696	»
Bioresmethrin	>8,000	0.4	>20,000	»

* LD_{50} for mice.

methyl, toxaphene, chlorfenvinphos, and thiophos are selective insecticides with a moderate-to-slight degree of selectivity (K_{sel} from 1 tc 10).

Carbophos, metaphos, diazinon, etc. are somewhat more selective (K_{sel} from 10 to 100); fenthion, lindane and alodane are very selective (K_{sel} from 100 to 1000); and metoxychlor, permethrin, phoxim (valexon) and bioresmethrin (K_{sel} over 1000) are extremely selective insecticides.

Pesticide assessment for practical utility should integrate, in addition to the selectivity coefficient, also their absolute lethal dose levels. The chemicals with low LD_{50} for insects are more effective insect killers but, for the same selectivity coefficients, may be more hazardous to warm-blooded animals. As an example, the insecticide with a selectivity coefficient of 100 but having the LD_{50} for insects and warm-blooded animals equal to 0.2 and 20 mg/kg respectively is in the class of extremely toxic chemicals for the warm-blooded, with a considerable probability of being toxic to humans. In contrast, the agent having the same selectivity coefficient but with the insect and warm-blooded LD_{50} of 20 and 2000 mg/kg is practically safe.

Although the housefly provides a convenient test object for insecticide toxicity evaluation, it is likely to differ widely from other insects in sensitivity to various compounds. In this regard, the selectivity coefficients listed in Table 3 should be perceived to have only relative value. Hygienical regulation of chemicals, nevertheless, takes into account their values obtained for the most sensitive animal species. If the LD_{50} of a pesticide as defined from data on some animal species is below 50 mg/kg the pesticide should be categorized among the Extremely Toxic Chemicals and banned for transfer into practice. With LD_{50} data available from three species of laboratory animals, one is able to define a species sensitivity coefficient (SSC) as the LD_{50} ratio of the most sensitive to the least sensitive species. Sanotsky and Ulanova (1975) separate three classes of substances according to the magnitude of this coefficient: differences in species sensitivity are not marked if it is less than or equal to 3; marked if 3 to 9;

Table 4

Differences in Species Sensitivity to Chlorfenvinphos (from Data Summarized in Hollingworth, 1976)

Species	LD$_{50}$, mg/kg	Selectivity coefficient (vertebrates/flies)
Housefly	1.36	—
Rat	10	7.4
Mouse	100	74
Rabbit	500	368
Dog	>12,000	>8,824
Pigeon	16.4	12
Pheasant	107	79
Quail	148	109

and strongly marked if over 9. Of the 52 chemical substances analyzed for their species sensitivity coefficients, 27 were found to belong to the first, 23 to the second, and only two to the third class. With respect to the class 3 chemicals, special care is needed in data extrapolation to humans. For example, among the warm-blooded animals rats are the most and dogs the least sensitive to chlorfenvinphos (Table 4), their species sensitivity coefficient more than 12000. Naturally, in the rare cases such as these the causes for such sensitivity differences need to be examined in order to validate a possible toxicity forecast for man.

Roll (1973) considers it worthwhile to analyze species-specific differences in sensitivity at each stage through the toxic process: absorption, distribution, delivery to the active site of the toxic effect, and reaction with the receptor. Absorption patterns, for example, appear to have much in common, anatomically and physiologically, in mice, rats, other mammals, and man. The circulation rate depends upon body size. In mice, the ratio of cardiac output to blood mass equals 20, as against one in man, thus suggesting a much higher circulation rate for the mice relative to man and, consequently, a shorter elimination half-time of a substance from the tissues in mice than in man. The body surface area versus weight in man and mice has the ratio of 12:1. Sometimes a better correlation of chemical doses for humans and mice results if the dose is computed, not on the basis of body weight, but per unit body surface area. Small mammals will ordinarily metabolize chemical compounds much faster than large animals and the rate of metabolism in herbivors is higher than in carnivors. In small laboratory animals the renal elimination of substances by glomerular filtration and tubular excretion proceeds with a greater speed than in humans (for some substances the elimination rates in mice, rats, and humans differ by a factor of two or three).

Krasovsky (1973) estimates that human sensitivity to acetophos is 5 to 8 times the sensitivity in mice and 2 to 4 times that in rats. Normally these differences would be within the same order of magni-

tude. Based on an analysis of 1119 variation coefficients characterizing comparative sensitivity of humans and several animal species to pesticides, drugs, and other substances, divergencies in the sensitivity to toxics between man and albino rats were found to be in the order of 1—10 for 80 percent of the cases. To some of the agents tested, human sensitivity was greater. Compared to rats, man has been 10 to 25 times more sensitive in 9 percent of the cases, 25 to 450 times in 3 percent of the cases, and less sensitive in 8 percent of the cases. The largest variations were recorded in estimating sensitivity to some pharmaceutical substances, of which alkaloids — atropine, morphine, nicotine, physostigmine, and muscarine — are an illustrative example. The pattern as identified by the a u thor is that the larger the animals, the more likely will they possess high sensitivity to a majority of echemical substances.

Krasovsky (1973) holds the view that the problem of determining extrapolation coefficients to man needs to be solved on a case-by-case basis and its solution to be tied to the chemical being tested and the test animal species involved. Ulanova (1971) insists that special care must be exercised whenever the sensitivity coefficient exceeds 9.

Hollingworth (1976) performed a correlation analysis of the link between the LD_{50} values for two species of laboratory animals, drawing upon data on several dozens of pesticides but with emphasis on the organophosphates. From the data cited in Table 5 it is evident that a satisfactory correlation of the LD_{50} values is the case in almost all animal pairs. The correlation is somewhat higher in the organophosphate group than it is for all insecticides excepting the low correlation of toxicities in the rat-chicken pair for both, all insecticides and specifically for the organophosphates. This is presumably the result of chickens' extraordinary sensitivity to the neurotoxic action of some pesticides, particularly OPP. These results agree with the data of Ulanova (1971) and Krasovsky (1973).

Table 5

Correlation Coefficients of LD_{50} Values for Warm-Blooded Animals (Hollingworth, 1976)

Animal species	For entire group of insecticides	For OPPs
Rat — mouse	0.869	0.889
Rat — guinea pig	0.777	0.881
Rat — rabbit	0.840	0.819
Rat — dog	0.751	0.847
Rat — chicken	0.547	0.161
Mouse — guinea pig	0.698	0.929
Mouse — rabbit	0.762	0.843
Mouse — dog	0.560	0.785
Rabbit — guinea pig	0.930	0.918
Rabbit — dog	0.800	0.855

Mechanisms of Selective Toxicity. According to O'Brien (1964, 1967), physiological selectivity is subdivided into internal and related to absorption. The former may well depend on differences between the substances' pattern of penetration to the site of assault (target), the nature of this target, the effects of the assult, differences in the substances pattern of metabolism (activation and breakdown), their time of circulation and the elimination rate. Although any one of these variables may be responsible for the initiation of selectivity, the important ones are the differences in the pesticide penetration pathways across barriers, their metabolic patterns, and the mechanisms of action.

Differences in the penetration pathways across barriers may dictate dissimilar patterns of absorption into the body and subsequent distribution in and elimination from it. These factors may each influence selective toxicity. Transport of the substances is simultaneous with their metabolism.

There are major structural differences between insect external coverings and the barriers pesticides have to overcome to gain access into human body. Although in both instances the barriers will be more efficiently penetrated by lipoid-dissolved compounds, the passage of substances will vary in speed and extent because of their structural dissimilarities. Insect shells cover the range from a hard and thick armor, as in some beetles, to a thin membrane, as in young mosquito larvae. There are broad variations also in the amount of lipids present in the shell structure. All this constitutes one of the causes for the selectivity of the insecticides effective by contact exposure.

The rate of absorption through insect coverings and the skin of warm-blooded animals is influenced by the nature of the solvent, concentration of the chemical, temperature, humidity, and the metabolic rate of substances down their absorption pathways. In relation to insects, these factors were closely examined by O'Brien (1964, 1967) and Hollingworth (1976) and in relation to the warm-blooded animals and humans by Kundiev (1975). It would be of interest to compare the $\frac{LD_{50}\text{ externally}}{LD_{50}\text{ by injection}}$ coefficients for insects with the cutaneous-oral and cutaneous-venous coefficients for warm-blooded animals.

Of some importance are the different pathways of pesticide passage across the internal barriers of the body particularly the hematoencephalic barrier. The compounds which, at nearly neutral pH, become highly ionized as well as the molecules conveying charges display low penetrability into the nervous system of insects and warm-blooded animals. Yet the toxic effect of some compounds on warm-blooded animals does not spring from their central effect alone. The agents such as these have no future as insecticides because they are more toxic to the warm-blooded animals than to insects. While this so, some polar neurotoxic chemicals like nicotine, tetram, octamethyl and mipafox are potent toxics for plant bugs and ticks. In them, the barrier between the blood and nervous system is permeable if only in part to the polar compounds.

The lipoid solubility of chemicals appears in some instances as the factor responsible for their concentration in the body of some animal species. According to Hollingworth (1976), lipophilicity operates as a sponge of sorts, to adsorb and concentrate pesticides from the environment. It is the very property which renders some pesticides (e.g. carbamates and abate) selective toward mosquito larvae as contrasted from the insects that habitate on the ground. The larvae concentrate 98 percent of the abate content in water and toxicity to them is proportional to lipophilicity. To many of the lipophilic pesticides such as chlorinated hydrocarbons and pyrethroids, the sensitivity in fish is remarkable — the latter concentrate them from water solution by driving it through the gills with their large surface of lipoprotein membranes and ramified vascular network.

Still another significant factor of pesticide toxicity is their conjugation by various components of body cells and fluids as a result of which a large portion of the chemicals never reach the point of attack responsible for the reaction. Of particular importance in this context is pesticide interaction with proteins.

Kabachnik et al. (1965) have discovered that an extension of the alkyl radical in O-ethyl-S-alkylmethylthiophosphinates enhances dramatically their anticholinesterase properties. The enhancement is due to the changing sorption and orientation of the compounds on the enzyme surface, a steric factor, and the interaction of hydrocarbon radicals with the hydrophobic cholinesterase sites located near its esterase center. But besides that, a study on the toxicity of the compounds in this series has revealed it to be by no means augmented with the elongation of the hydrocarbon radical. Rather, it showed a regular pattern of decline (Kagan, 1967). This may be attributed to an increase in the nonspecific sorption of the organophosphates having long hydrocarbon radicals on the proteins, and their binding. As a result, the major part of the compounds fail to attain the enzymes responsible for the toxic process. Phosphorylation of the various esterases having no major functional role in the body can be also assumed to afford protection, as the esterases bind irreversibly the organophosphate inhibitor. There are cases, however, when the reversible conjugation of a toxic substance by proteins can be considered an additional source of its entry into the body as it is increasingly released from the depot.

The pesticide interaction with proteins in insects and warm-blooded animals can modify their selective toxicity. One of these selective toxicity mechanisms *exploits* varying elimination rates of substances from the body. It seems, though, that these distinctions in the elimination patterns will have little obvious effect since urinary excretion helps the body to dispose of the polar metabolites that already possess no appreciable toxicity.

We have sought to identify *distinct metabolic patterns* for a number of structurally similar organophosphates with different toxicity for warm-blooded animals and different selectivity toward insects (Kagan et al., 1965).

Hollingworth (1976) provides data concerning different activities of enzymes in detoxifying xenobiotics in different tissues (in vitro experiments). Thus, A-esterase, which metabolizes paraoxon-methyl and paraoxon-ethyl, exibits, in the liver microsomes of warm-blooded animals (rat, rabbit, and horse), many times its activity in the tissues of the housefly. Nonetheless the mixed-function oxidase responsible for the epoxidization of aldrin has been found more active in the housefly than in the rat. It is not uncommon to see structurally similar compounds display observable, marked differences of metabolic patterns. E.g. the A-esterase activity of the liver microsomes in relation to paraoxon-methyl is 58 times the activity toward paraoxon-ethyl. Since this enzymatic system is totally lacking in houseflies and some other insects, both paraoxon-methyl and -ethyl are of course highly toxic to them.

There is no doubt about the important role, in selective toxicity mechanisms, of mixed-function oxidases (MFO), the enzymatic system involved in the metabolism of different pesticide classes. It is a fact that the MFO induction by the organochlorine acaricide milbex diminishes the toxicity of the organophosphate insectoacaricide phosalone, the magnitude of the MFO induction (5.3 times) consistent with the degree of antagonism between them (5 times; Zlatev, Yakushko, 1975). On the other hand, the MFO inhibitors sesamex, piperonylbutoxide, and SKF-525A act as synergists for a number of insecticides, by inhibition of their oxidative detoxification. The synergism of these compounds has been ascertained for organophosphates, carbamates and a host of other pesticides whose detoxification comes about through their oxidation involving mixed-function oxidases (Hodgson a, Fate, 1976). Thus, the induction of mixed-function oxidases in the warm-blooded animals and their inhibition in the insects is one of the ways to enhance selective toxicity.

The possibility to guide MFO activity in the desired direction has been demonstrated by Popov et al. (1979) in an exemplary study of phthalophos metabolism by rat liver perfusion. There the MFO induction was induced by daily oral administration of milbex to the test animals in a dose of 240 mg/kg, or 1/5 LD_{50} and inhibited by injection of tetramethylthiuramdisulfide (TMTD) at 110 mg/kg, or 1/5 LD_{50}. The MFO induction caused a swift rise in the biotransformation rate of phthalophos, manifested by an increased rate of decline of the phthalophos concentration in the perfusate with a corresponding, faster increase in the concentrations of metabolites, oxymethylphthalimide and phthalimide. Conversely, with the rats pre-treated with TMTD, there followed a deceleration of the curve for the disappearance of phthalophos from the perfusate and a reduction in the concentration of its metabolites. However both milbex and TMTD are capable of scaling down the anticholinesterase activity of phthalophos, though the mechanisms to produce this phenomenon were different in either case. While milbex diminishes the anticholinesterase activity of phthalophos by boosting its detoxification, TMTD achieves the same by inhibiting oxidative desulfuration as a result of MFO inhibition.

Differences in the mechanism of action. Distinctive modes of action and structures of the enzymes and receptors which are targets (points of assault) for the action of pesticides have been suggested as the cause of their selective toxicity. A clear example may be the ability of some herbicides to interfere with plant photosynthesis. And, because photosynthesis is specific to plants alone, many herbicides are distinguished by low toxicity for the warm-blooded animals and human beings.

The low toxicity of pyrethroids for the warm-blooded animals arises, it seems, from the varying sensitivity of peripheral nerves to them. In the insects and crawfish, sensitivity to pyrethroids is relatively high, whereas frog and rat nerves are pyrethroid-resistant so that no blockade comes up even with high-level exposure (Holingworth, 1976). Cholinergic synapses in insects and warm-blooded animals are known to vary in sensitivity to OPP. Presumably, this is because the nerve ganglia of some insects are glutaminergic, whereas nervous excitation in warm-blooded animals is mediated by acetylcholin. So far it has not been possible to obtain effective antagonists for the glutaminergic receptors for the reason of their high specificity, but the search for selective insecticides on that basis has a good chance of success.

The mechanism of action of the growth regulators for insects works basically by their activation at critical periods of growth and development. Analogs of juvenile hormones are highly selective pesticides because no points of assault for their effect exist in warm-blooded organisms.

Thus, to summarize, selectivity has by and large a polyfactor nature so the elucidation of the contribution therein by each of the factors contributes to effective discovery of new pesticides with an optimal degree of selectivity.

New Trends in the Quest for Selective Pesticides. The most fundamental approach to the discovery of selective pesticides has been to trace differences of biochemical and physiological processes between the organisms concerned; to identify the process segments vitally important for some species and absent from or unimportant to some others; and to mount a quest for the chemicals that assault the parts of metabolism in harmful organisms that are non-existent in useful ones.

High selectivity of action is a feature of several herbicides. For example, triazine derivatives interfere selectively with the process of photosynthesis at the stage of Hill's reaction. They are only slightly toxic to warm-blooded animals as photosynthesis is unique for plants alone. Triazines have no effect on useful plants, particularly cereals because the latter hydrolize them. The herbicides that act basically by suppression of Hill's reaction include the derivatives of phenylurea, acylanilides, and phenylcarbamates. Phenoxyacetic acids affect plants in much the same way as auxine, their natural hormone (indolylacetic acid). Yet many plants are unable to degrade excessive amounts of

phenoxyacetic acid and hence its herbicidal properties. A pronounced auxine-like effect is exerted by tordon (trichloramine pyridincarboxylic acid), a chemical more stable than 2,4-D.

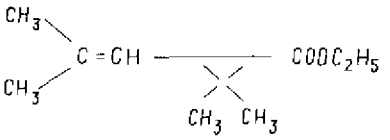
2,4 — dichlorophenoxyderivatives of aliphatic acids exhibit selective herbicidal effects. When applied on sensitive weeds, they are broken down in their cells to form toxic 2,4-dichlorpenoxyacetic acid. Numerous cereal crops lack betaoxidase of aliphatic acids responsible for lethal synthesis, the fact underlying the selectivity of 2,4-D derivatives to weeds (Albert, 1971). Dithiocarbamates exert fungicidal action, due probably to their metabolisation into isothiocyanates which block the thiol groups in the causative agents of plant diseases; the similar effect of captan (trichloromethyl-thio-tetrahydrophthalimide) comes about through disruption of the thiol groups. In fungus cells captan reacts with thiols to release carbonylsulfide producing a potent fungicidal effect.

Growth regulators of plants and insects offer a good example to the high selectivity of pesticides. They affect strongly the critical phases in the growth and development of exposed pests but leave them unaffected in warm-blooded animals (Baskakov, 1978; Hollingworth, 1976). The substances influence selectively morphogenesis in weeds or harmful insects only at their certain development periods and do not influence other organisms. The principal guidelines for the use of plant growth regulators include speeding their maturation, increasing resistance to low temperatures and drought, avoiding crop lodging, and performing defoliation to facilitate mechanized harvesting. Ethephon, a retardant from this very group of chemicals, owes its defoliating capacity to the release of ethylene by plants, as a result of the degradation of beta-chloroethylphosphonic acid. Ethylene is endowed with hormonal activity. The addition to ethephon of modest quantities of endotal precludes its displacement from the leaves and potentiates the effect. Benzylaminopurine disturbs the state of dormancy in weed seeds, provokes their ahead of time sprouting and thereby exposes the shoots to easy destruction by low herbicide doses.

A new promising trend in obtaining highly sensitive insecticides came with the discovery of growth regulators for insects. The particular stage of insect growth at which they are applied makes all the difference between their effects. Of great interest are the analogs of juvenile hormones, by virtue of their extremely low toxicity for warm-blooded animals and some useful insects. Hollingworth (1976) reported the discovery of highly selective juvenile hormone analogs, their action based on the use of specific targets efficiently involved in metabolism in harmful insects and non-existent is useful organisms (warm-blooded animals). The author points out that there have to be other alternatives in this respect, e.g. the glutaminergic nerve ganglia in the insects as contrasted from the cholinergic ones in the warm-blooded animals. This makes it potentially rewarding to look for glutamine agonists and antagonists which may prove insecticidal.

The recent years have seen a surge of researcher interest in pyrethroids, the analogs of natural pyrethrines, some of which — allethrin, bioalethrin, resmethrin, bioresmethrin, dimethrin, and tetramethrin are very active insecticides with low toxicity for the mammals. Moreover, their use rates are so low that they never accout for more than one tenth of the total usage of common organophosphate and carbamate pesticides. Lastly, pyrethroids have the advantage of rapid detoxication in the warm-blooded organism.

Natural pyrethrines represent esters of d-transchrysanthemum monocarboxylic (CMC) and d-transpyrethric acids (CDC) and cyclic ketone alcohols (pyrethrol, cinerolone, and jasmolone). Synthetic pyrethroids are esters of d-transchrysanthemum monocarboxylic acid esters:



Ethyl ester of d-transchrysanthemic acid.

Among the synthetic pyrethroids, compounds have been isolated which by far exceed natural pyrethroids in insecticidal activity and, moreover, are slightly toxic to warm-blooded animals. Another recent break-through came with the discovery of a new class of esters of alfa-substituted phenylacetic acids ($C_6H_5CHRCOOR'$ — R = = CH_3,C_2H_5, iso-C_3H_7, iso-C_4H_9) which surpass even pyrethroids in the insecticidal action they exert (Promonenkov, Korotkova, 1978).

Within the last few years there have been numerous attempts to control insect behavior and reduce harmful insect populations using pheromones. Today pheromones have been isolated and identified and their structure determined for 42 insect species (Kondratiev et al., 1978). They are unsaturated aliphatic compounds with 10 to 20 carbon atoms and a different locus and configuration of the double bond. Their varied interrelations in different insect species make pheromones strain-specific. Beyond that, their specificity has to do also with the seasonal patterns of attraction and habitation areas of insects, availability of synergists, and several other conditions. Optical isometry has a role of its own as far as pheromones are concerned. The advantages of pheromones include low toxicity for warm-blooded animals, high biological activity, and no environmental harm. LD_{50} for them lies within 2 to 34 g/kg. The use of pheromones significantly reduces if not totally rules out the need of insecticide application. Their very low quantities are required for insect traps: e.g. codlemone needs to be applied at a dose of one mg per trap and phunemone at 5 mg. Still another use of pheromones comes in raising shelter belts and bands of insect traps, to lure males away from the fields and thereby bring down dramatically the pest numbers present.

SOME SALIENT QUESTIONS IN CLINICAL MANIFESTATIONS, DIAGNOSIS, AND TREATMENT OF PESTICIDE POISONING

Acute poisoning is the most numerous case on records, while chronic intoxications are common to persons permanently exposed to pesticides. In the United States the statistics for 1966 stood at 72,560 intoxications overall including 4,434 intoxications by pesticides; the figures for 1967 were 83,704 and 5,002 respectively and for 1968, 105,178 and 2,919 (Kaloyanova-Simeonova et F. Fournier, 1971).

As suggested by data from Barbaly (1968), acute poisonings are most typically caused by drugs, less frequently by household chemicals including pesticides, then by foodstuffs, and lastly by plant and animal poisons. Among the toxic household chemicals, insecticides head the list, followed by cosmetics, detergents, and other agents. Kuznetsova (1968) names carbon monoxide as the agent responsible for 30.5 percent of all intoxications, acids for 21.7 percent, medicaments for 12.5 percent, insecticides 4.8 percent, alkalies 4.8 percent, and solvents 2.6 percent.

The findings of a WHO analytical survey (1979) of 10,000 incidents of poisoning in Belgium revealed that among the substances causing intoxications in children medicaments come first accounting for 52 percent of the total, household chemicals are second — 22.4 percent; industrial chemicals third — 6.5%, and pesticides fourth, 4.4 percent.

The number of poisonings is the largest in the United States, second-largest in Japan and third-largest in Turkey. Within the two decades from 1945 to 1966 foreign literature reported 15.032 episodes of pesticide poisoning in Asia (44.3 percent of the total), 14, 452 in America (42.6); 3,410 in Europe (10), and 955 in Africa (2.8 percent). In Europe, despite its highest level of pesticide use, intoxications are reportedly less numerous than in America and Africa, seemingly because of the more advanced and efficient preventive programs.

Of the pesticides implicated in these intoxications, OPP hold the lead, representing 73.4 percent of their total number; the second-placed OCP represent 12.6 percent, arsenic-containing chemicals 6.1 percent, organomercurials 2.4 percent, methyl bromide 1.1 percent, zinc phosphide 0.9 percent, and cyanic compounds 0.6 percent (Poichenko, 1968).

Two thirds of the intoxications listed in foreign countries are occupational and the balance one third domestic. The former emanate from pesticide manufacture (23 percent) but more so from their marketing and application (77 percent). Numerous intoxications result from direct work exposure to pesticides during spraying, dusting, seed desinfecting and other operations. In the USSR the incidence of occupational poisonings is much lower than abroad.

The key causes of pesticide intoxications include careless storage and handling, and underestimation of their hazard potential. Many intoxications are traceable to misuse of pesticide containers.

Polchenko (1973) found out that 86 percent of all intoxications are linked to extremely toxic chemicals (LD_{50} below 50 mg/kg), about 6 percent to highly toxic (LD_{50} between 50 and 200 mg/kg), 2 percent to moderately toxic (LD_{50} from 200 to 1000 mg/kg) and 6 percent to slightly toxic (LD_{50} above 1000 mg/kg). Thus, the use of the extremely toxic chemicals presents the major hazard for the origin of acute intoxications, this being the reason why this group of pesticides is banned for agricultural use in the USSR.

Once an intoxication starts, its rate of progress will depend on the chemical structure, physicochemical properties, quantity, and routes of entry of the causative agent. The progression rate is especially high with the entry via the respiratory system. Although most of the pesticides currently in use have low volatility they too may find their way into the respiratory system in the aerosol form (dust or hydroaerosol) during crop dusting or spraying operations. On the other hand, methyl bromide, chloropicrine, hydrocyanides, and some organophosphates (DDVP) are highly volatile so that severe poisonings may result from the inhalation of their vapors. Whereas fine-dispersed particles from 5 to 10μ in size reach the alveoli with inhaled aerosols, larger-size particles settle in the respiratory passages, to be swallowed and carried on into the alimentary canal.

The progress of intoxications can be influenced in large measure by the age of exposed persons. Children are more sensitive to toxic substances in general and pesticides in particular. But because of their greater body surface area/weight ratio and the specific skin structure they are capable of much faster pesticide absorption.

Some physiological characteristics of the female organism such as menstrual cycle, gestation, lactation period, and menopause make it more susceptible to poisons. The resistance of the female organism to toxic exposures is decreased by the lability of the hemogenetic system, heightened capillary permeability in the menstrual period, and endocrinal and neural effects. This is not to exclude the possibility that some female individuals can be and are in fact more resistant to poisons than males. The development pattern of intoxication also responds to variations in individual sensitivity to poisons. In a sensitized organism even minor toxic quantities can give rise to violent responses.

High temperatures of ambient air make the course of intoxication more severe and even small quantities of pesticides are sufficient to provoke it (Yakubov, 1965). High-altitude Hypoxia and enhanced ultraviolet radiation at high altitudes also influence the organism's sensitivity to poisons (Yusupov, 1977).

Among the clinical features of pesticide intoxications there are some distinct common syndromes generally acknowledged for the effect of many toxic substances. They are hypoxia; various respiratory, circulatory and metabolic disorders; diseases of the central and peripheral nervous systems; lesions in the liver, kidneys, gastrointestinal tract, and other internal organs; and allergic symptoms. Though the direct determination of pesticides and their metabolites assists immen-

sely the credible diagnosis of pesticide poisoning a detailed analysis of clinical and laboratory changes is also important. Combined together, they can identify the affection as having been caused by a particular pesticide. Burkatskaya et al. (1978) recommend, for periodic or routine medical examinations of persons occupationaly exposed to toxic chemicals or suspected intoxications cases to apply as, part of the toxic chemicals or suspected intoxication cases to apply, as part of the protocol, specific clinical and laboratory tests appropriate to the particular pesticide class of which the pesticide concerned is one. Thus, common work exposure to organochlorines necessitates, quite apart from the routine general hematologic examination and urinalysis required for cases of contact exposure to any pesticides, pesticide determination in the biosubstrates and of protein and urobilin in the urea. For suspect intoxication cases this set includes an additional estimation of sugar and lactic and pyroracemic acids in the blood; tests to assess the functional state of the liver, the state of the coagulative blood system, and the activity of alkaline phosphatase, alcolase, arginase, LDG isoenzymes; tests on the functional state of the kidneys (Zimnitsky's test); measurement of Ambar's constant and clearance coefficients for urea and endogenous creatining, and residual nitrogen; and examination of the cardio-vascular system. In sum, these indices permit evaluating the state of those functional systems worst-affected in OCP intoxication.

Suspects of OPP poisoning are examined for cholinesterase activity, the state of the coagulative blood system, the protein spectrum of the serum, the glycemic curve and the activity of aminotransferases and LDG isoenzymes, and the functional tests of the liver and kidneys.

Following OMP exposure (to organomercurials), one will ordinarily test for mercury in the urine and sulfhydryl groups in the blood, define the blood protein spectrum, measure the contents of sugar, sodium and potassium in the blood and of 17-ketosteroids in the urine, and examine the functional state of the central and autonomous nervous systems, liver, and kidneys.

Poisoning by some carbamine pesticides such as sevin, pirimor, and dicresyl produces a side-effect by inhibiting blood cholinesterase and affecting the functional state of the liver and kidneys. Besides, 1-naphthol is detectable in the blood in the intoxication by sevin and paraaminophenol in the intoxication by carbin and chlorisopropyl phenylcarbamate.

Dithiocarbamates diminish the quantity of the blood sulfhydryl groups and modify the incorporation rate of radioactive iodine into the thyroid. They are metabolized to carbon disulfide, thereafter detectable in exhaled air. In the case of carbathion poisoning, thiocyanates are found in the blood.

With the intoxication by nitro- and chlorine derivatives of phenol (dinitro-o-cresol/DNOC/, nitraphene, acrex, carathane, pentachlorophenol, etc.), the pesticides and their metabolites are detectable in

the biosubstrates and elevation of the body temperature is rather likely, along with the formation of methemoglobin and Heinz bodies in the blood and altered activity of oxidative enzymes.

Methyl bromide increases the quantity of bromides in the blood and decreases the activity of SH-groups (alkylation).

When copper-containing pesticides gain access into the body the copper can be recognized in the blood serum and urine, and elevation of the body temperature may follow. Cyanide intoxications proceed with urinary excretion of thiocyanic compounds.

Like most other toxic compounds, pesticides are substances of the non-protein nature but ones that can combine with proteins to form complexes with antigenic properties. Thus, allergic dermatites and eczemas develop after contact exposure to DDT, hexachloran, carbin, thiuram, zineb, maneb, carbophos, and thiophos (Despotov, 1967; Karimov, 1969) as well as other OPP, OCP, OMP, carbamates, cyanides and fluorides. In addition to retarded allergic responses, the body comes up also with the more immediate type of responses in the form of bronchial asthma or Quincke's edema (Leliukh, Maliuk, 1966).

A fundamental if not ultimately resolved question concerns the threshold levels in the allergizing action of chemical substances. Some authors (Alexeeva, Shumskaya, 1970; Shumskaya et al., 1967) favour the idea of setting the threshold dose and concentration levels for the substances on the basis of their allergenicity. Others (Kryzhanovskaya, 1968) object on the grounds that chemical allergenes have a virtually no-threshold action potential. But, since one can ordinarily correlate the doses of substances with their allergic effects there seems to be no reason why the establishment of the threshold doses and concentrations should be impossible in principle. In fact, this is the basic assumption in setting hygienic standards and specifications for the substances with allergic potential. On the other hand, one must consider disparity of the threshold dose levels in terms of toxic and allergic effects. Routinely, the sensitizing doses of substances will be smaller than their toxic doses. Sensitization may or may not come from exposure to the toxic doses by reason of the immunologic paralysis that develops. Kryzhanovskaya (1968) argues that with current methods of hygienic research the principle of setting standards for chemical allergenes according to their potency is warranted and recommends to define the potency from the incidence of anaphylactic responses to a given substance, the latter's sensitizing dose, level the development rate of the allergic state, and the magnitude of the sensitization titre. The author recognizes chemical allergenes as potent (80 percent of the cases exhibit observable allergic responses and their obvious manifestations), moderate (50 to 80 percent of the animals develop positive responses) and weak (positive responses recorded in less than 50 percent of the animals).

The rise and development of an allergic state is merely one of the ways in which a chemical agent can conspicuously interfere with immune processes. Semencheva (1967, 1970) indicates that toxic sub-

stances can give rise to autoimmune processes. Their onset may be signalled by auto-antigene formation as chemical compounds damage the tissue proteins. In laboratory animals, pathological liver antigenes and autoantibodies result from exposure to several carcinogens, DDT, and propazine (Korosteleva, 1966; Semencheva, 1970). Antihepatic antibodies were likewise identified in persons previously exposed to organochlorines and organophosphates (Kachalai, 1968; Sosnovik, 1959).

Ongoing studies look at the role of autoimmune processes in the origin of the diseases induced by different pesticide exposures, with special attention to the neurotoxic complications observed after previous intoxication experience with organophosphates or organochlorines. An excess of polyneurites progressing into serious pareses and paralyses was noted after intoxications by chlorophos. The data become particularly meaningful if seen in the light of Tarashchuk's results (1970) with the induction of de-myelinization in sciatic nerves by chlorophos and other OPP, and the research findings of Zhabotinsky (1970), Rodshtein and Konovalov (1971) regarding the indisputably immuno-pathological character of this phenomenon.

Therapeutic remedies in acute intoxications may be general or specific. The general measures form part of the emergency care, their purposes including removal of the poison from the stomach or body surface, avoidance of its absorption into the blood, intensified excretion from the body of the fraction of toxicant already absorbed, maintenance of normal functioning of vital bodily systems, and elimination of symptoms of the adverse effect of the pesticide. The specific measures center primarily on the antidotal therapy.

Antidotes may be physico-chemical, chemical, and physical. Of these, the physico-chemical antidotes, their action based on the physical processes of adsorption, encapsulation, and altered solubility, are effective against immediate contact exposure, for example, in the gastric cavity. Very recently activated coal began to be used to sorb the toxic substances that have found way into the bloodstream. Hemosorption is remedial for various intoxications (Luzhnikov, 1977).

The molecules of a chemical antidote react with the poison to neutralize or bind it, thus promoting its alimination from the body. Examples of chemical antidotes are unithiol, succimer, penicillamine, mecaptide and other thiol drugs with the capacity to bind arsenic and mercury. Some complexones, e.g. EDTA-ethylendiamine tetraacetic acid, also act on the principle of chemical antidotes, but only those which combine with metals. Physiological antidotes of OPP include cholinolytic drugs which effectively control the toxicity of excessive acetylcholine which accumulates as cholinesterase is progressively inhibited by the poison. They include additionally cholinesterase reactivators capable of boosting its dephosphorylation and restoring the function of synaptic formations. Another possible factor to be considered for the mechanism of action of the cholinesterase reactivators is their direct chemical interaction with organophosphate molecules (Golikov, Zaugolnikov, 1970), the capacity leading to their

classification as mixed-type antidotes with dominant physiological antagonism.

The best therapeutic effect is achieved through the integrated use of antidotes with different types of effects. Thus, cholinolytic drugs are prescribed in combination with cholinesterase reactivators to treat organophosphate poisonings (Luzhnikov, 1977). The 2PAM reactivators dipyroxim, toxogonin, and other agents contain in the molecule a quaternary, positive nitrogen atom, making them ill-equipped for effective penetration into the central nervous system. They can therefore control only peripheral OPP effects. By way of partial contrast, isonitrosine, the agent proposed by Zaugolnikov and Golikov (1970), has remarkably low toxicity and penetrates well into the central nervous system but is a comparatively slow cholinesterase reactivator. Krivenchuk et al. (1969) synthesized diethyxim, a derivative of thiohydroximic acid somewhat similar in chemical structure to acetylcholine. Our investigations (Kagan et al., 1971, 1975; Kokshareva, 1975) of diethyxim have demonstrated its well-defined antidotal effect in organophosphate intoxications (DDVP, chlorophos, phosphamide, metaphos, phosalone, etc.), remarkably low toxicity (5 to 7 times less toxic than dipyroxim), efficient cholinesterase reactivation in the tissues of laboratory animals, and a far greater ability, compared with dipyroxim, to remove a DDVP-caused nervous-muscular block. The deblocking effect of diethyxim is associated with cholinesterase reactivation, release of cholin-receptive proteins from their conjugation with organophosphorus inhibitors, and resumption of normal acetylcholine discharge by the presynaptic formations (Kovtun, Kokshareva, 1974). Diethyxim is also effective in contributing to functional recovery of the peripheral nerve, previously damaged by DDVP (Kovtun et al., 1978). Data on the reactivating effect of diethyxim relative to cerebral AChE are summarized in Table 6 (Kokshareva et al., 1977).

Table 6

Percent AChE Reactivation in Different Brain Regions of DDVP-Poisoned (10 mg/kg) Rabbits after Treatment with Cholinesterase Reactivators

Initial Substrate	Inhibition of AChE Activity by DDVP, %	90 min after administration	
		diethyxim (20 mg/kg)	dipyroxim (3 mg/kg)
Medula oblongata	73	96	3.3
Superior and inferior colliculi of mesencephalon	65	86	28
Hippocampus, hypothalamic region	68	88	25
Caudate nucleus	73	25	5

With the use of the therapeutic diethyxim dose, AChE reactivation in the medula oblongata, superior and inferior colliculi of the midbrain roof, hippocampus, and hypothalamus is almost complete, with dipyroxim, virtually po AChE reactivation occurs in the medula oblongata and caudate nucleus and little reactivation in the other regions. Whereas diethyxim brings EEG back to normal in DDVP-poisoned test animals, dipyroxim failed to restore brain biopotentials of the rat for as long as 90 minutes after injecting the reactivator, its EEGs little or no different from the brain biocurrent records of poisoned animals, though none of the rats was killed. Beznosko et al. (1977) showed from work in cats with blocked peripheral M-cholin-receptors that diethyxim eliminates promptly the central hypotensive effect of armin – the ability present neither in dipyroxim nor in the quaternary analog of diethyxim.

In DDVP poisonings, results looked encouraging from paired diethyxim/dipyroxim application (Kokshareva, 1976). The associated index of therapeutic effectiveness (incremental growth of the poison's LD_{50}) reached eight. Their combined use enables the dose of either reactivator to be reduced and their toxicity to be lowered. Diethyxim stems the development of the neural-muscular block that normally occurs in DDVP intoxications, the fact suggesting a fairly rapid cholinesterase reactivation in the myoneural sunapse. As the diethyxim-based treatment was carried on signs of DDVP intoxication were progressively disappearing in cats and their arterial pressure and breathing were coming back to normal (Fig. 4). In rabbits, as far as can be judged from their ECGs, the drug has a favourable effect on cardiac activity (Lukaneva, Kokshareva, 1975). As reported by workers from the Sklifosovsky Institute for Emergency Medical Care, diethyxim exhibits a strongly marked therapeutic effect in the treatment of different OPP intoxications.

A new trend of the ongoing enquiry for cholinesterase reactivators focuses on the synthesis and efficacy assessment of complex metal compounds with aminoalcohols and other biological ligands (Evreev et al., 1968; Sasinovich et al., 1976; Kagan et al., 1977, 1978). The inhibited OPP enzyme needs for its reactivation the nu-

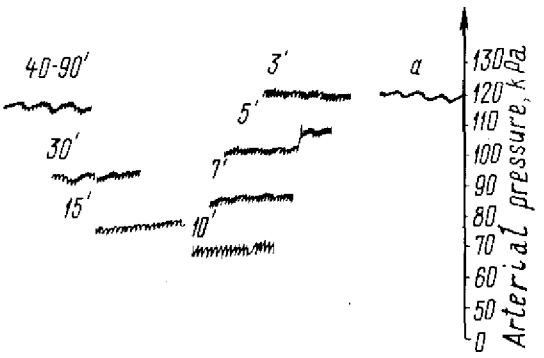


Fig. 4. Changes of arteria pressure in DDVP-poisoned cats (10 mg/kg) treated by diethyxime (20 mg/kg; Kokshareva's experiment):

cleophilic reagents capable of weakening the serine oxygen bond with the phosphoryl radical. The role of specific electrophilic catalysts can be delegated to the ions of transition metals. Since the salts of heavy metals are highly toxic it seems more advantageous, potentially, to test the less noxious metal complexes with the ligands containing free nucleophilic groups. While reacting with phosphorylated cholinesterase, they may be expected to combine nucleophilic with electrophilic effects and display the properties of reactivators. Dialcob, a complex of bivalent cobalt with N-allyldiethanolamine synthesized by Evreev and experimentally studied by Kagan, had been shown to provide a most effective cholinesterase reactivator of the type just discussed (Sasinovich et al., 1977).

Dialcob is slightly toxic, its LD_{50} for intramuscular injection to rats just upwards of 1 g/kg. The optimal therapeutic dose of dialcob represents 1/200 LD_{50}, which compares favourably with 1/15 for dipyroxim. Toxicity studies of the drug found its multiple doses to be moderately stimulating erythrocytopoiesis so that neither the functional state of the liver nor the cardiovascular and endocrinal systems are affected. It can cause primary irritation of the injection site if used at concentrations below 10 percent. The morphological changes and histochemical alterations (RNA and glycogen contents) observed in some of the experimental animals are moderate, reversible and therefore not dangerous (Rodionov, 1976).

In the event of intoxication by the organophosphates DDVP, chlorophos, phosphamide, phthalophos, valexon, bromophos, and phosalone dialcob is therapeutic within a broad range of doses, from 5 to 25 mg/kg. Survival in albino rats is thus doubled or tripled. It has the ability to potentiate the therapeutic effect of atropine sulfate. Combined injection of atropine sulfate (20 mg/kg) and dialcob (25 mg/kg) magnifies the LD_{50} of DDVP by a factor of 8.2. Dialcob reactivates the OPP-inhibited cholinesterase, and the normalization of tissue cholinesterase activity, the brain tissues included, has been confirmed histochemically (Fig. 5). Its application results in restored activity, chiefly, of extracellular cholinesterase, functionally the more essential one.

Dialcob has a normalizing effect on the metabolism of proteins, activity of reamination enzymes and other liver functions; it promotes the deblocking of myoneural transmission and removal of the respiratory changes resulting from the inhibition of the respiratory center as well as to the recovery of the EEG, pneumogram, and ECG in the DDVP-poisoned rats (Fig. 6).

In poisoning by the dithiophosphoric acid derivatives phosphamide and phthalophos, dialcob is a more potent drug than 2PAM, apparently because it can interact directly with the atoms of sulfur. It ends up or sharply relieves the convulsions in rabbits caused by DDVP intoxication and restores the normal breathing and cardiac output (Sasinovich et al., 1979).

The positive therapeutic effect of 2PAM, dipyroxim (TMB-4) efoxim (toxogonin), diethyxim, dialcob, and other cholinesterase reac

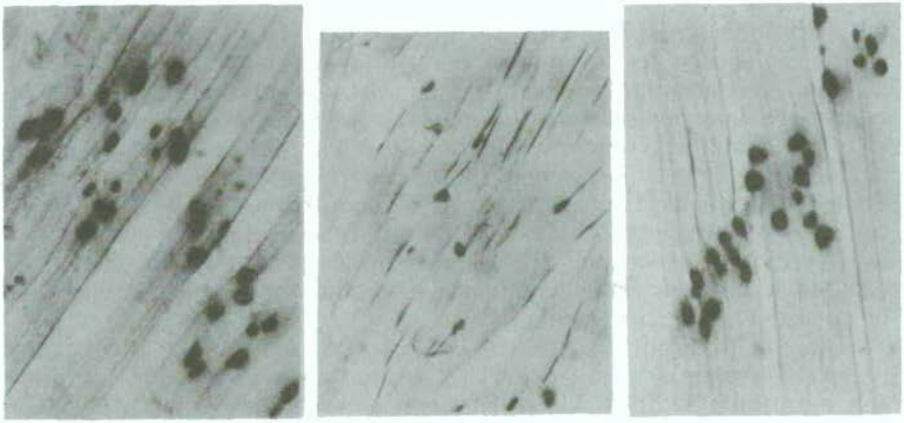


Fig. 5. Cholinesterase reactivation by dialcob in rat cross-striated muscle (Rodionov's preparation):
a — control; *b* — thirty minutes after DDVP injection at 20 mg/kg; *c* — twenty minutes after dialcob injection

tivators is attributed primarily to its resumed activity. However, some other mechanisms are secondarily involved. The reactivators have been found to weaken the OCP complex with the cholinreceptor and bring to normal the release of acetylcholine by the nerve ending (Rybolovlev, 1965; Dishovsky et al., 1973; Kovtun and Kokshareva, 1974; Golikov and Sanotsky, 1976). Direct interaction of the reactivators with organophosphates is also entirely possible (Golikov, Zaugolnikov, 1970). Some of them produce ganglioblocking and spasmolytic effects (Fig. 7) and diethyxim has the additional property of blocking M-cholinreceptors, evidenced by the reduction of the acetylcholine-induced intestinal contracture (Kokshareva, 1975). Although the mechanism behind the antidotal action of the cholinesterase reactivators in OPP intoxications is not fully understood at present their practical value is unquestioned. For the intoxications by organophosphates, a good therapeutic effect is achieved by paired injection of cholinolytic drugs with central and peripheral reactivators; for the intoxications by chlorophos, thiophos and carbophos, the use of atropine sulfate in conjunction with dipyroxim, toxogonin, and isonitrosine is remedial (Luzhnikov, 1974; Kosarev, 1975). The cholinesterase reactivators alleviate the severity of intoxications, decrease the incidence of serious complications and reduce lethality.

The antidotal performance of the cholinolytic drugs and cholinesterase reactivators is potentiated in the induction of cytochrome P-450, an agent in detoxifying OPP, aided with phenobarbital. Experiments in rats made it clear that phenobarbital causes the induction of cytochrome P-450 (by 46 to 153 percent) over a wide range of doses, from 70 to 1.5 mg/kg; the yardstick to gauge the induction is the signal intensity of electron paramagnetic resonance (Fig. 8). The maximum degree of the enzyme's induction after a single phenobar-

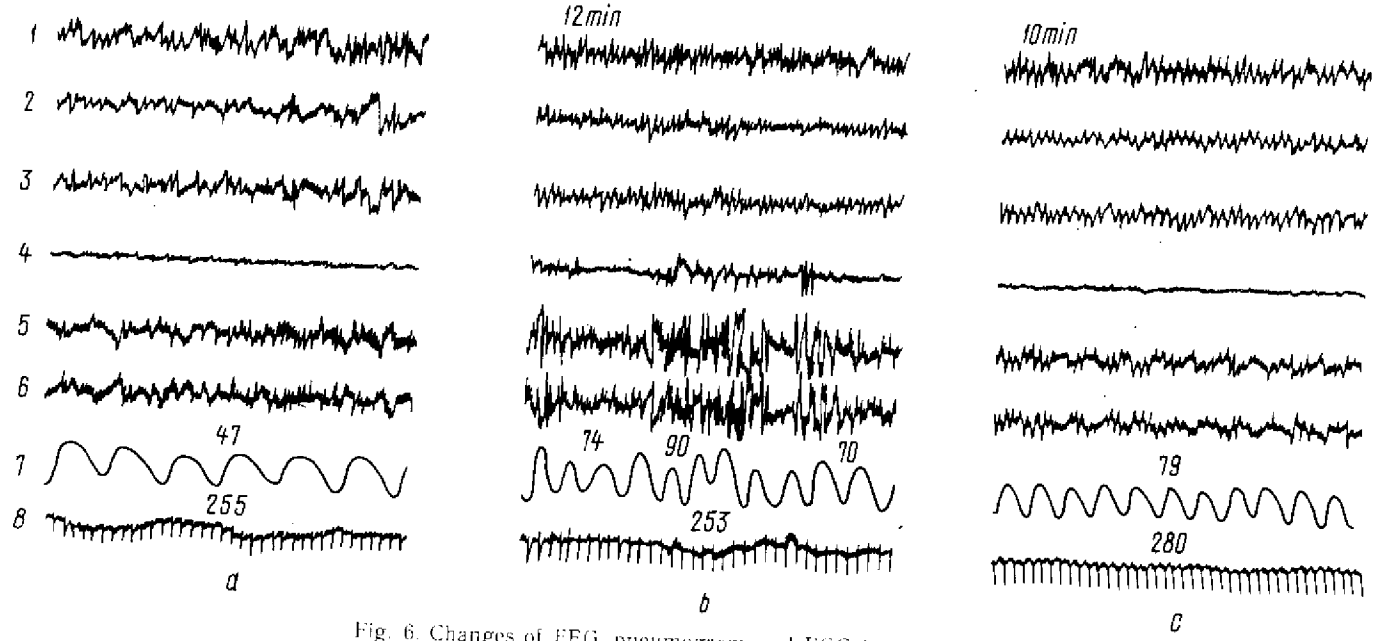


Fig. 6. Changes of EEG, pneumogram and ECG in a DDVP-poisoned (b) intact rabbit (a) treated by dialcob (c):
1, 2, 3 — frontal, parietal and occipital cortex regions; *4, 5, 6* — supraoptical; lateral and ventromedial hypothalamic nuclei; *7* — pneumogram; *8* — ECG

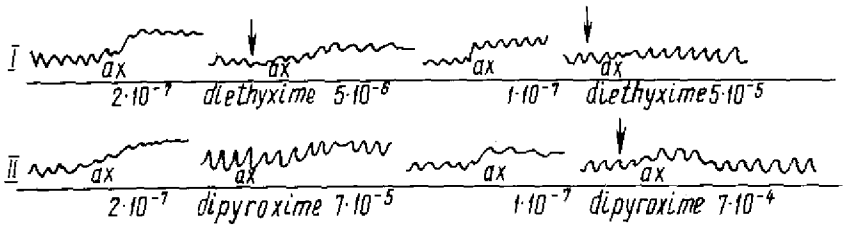


Fig. 7. Removal by diethyxime of rat duodenal contracture caused by acetylcholine (Kokshareva's experiment)

bital dose is in no way inferior to the degree of induction after three doses, and develops between the first and third days. Given the induction of cytochrome P-450, the toxicity of several organophosphates (DDVP, valaton, etaphos, actellic, and chlorophos) declines markedly -- the effect that can be applied as an innovative principle to combined treatment of OPP poisonings (Kagan et al., 1980).

Phenobarbital prevents the neuromuscular block developing in DDVP-exposed rats (Fig. 9) and is beneficial for cases of subacute intoxications by organophosphates (Fig. 10) and leptophos.

Perhaps the most difficult challenge is finding specific antidotes in poisonings by organochlorines as well as by other pesticides based on halogen-containing hydrocarbons because of their polytropicity. Miziukova et al. (1978) documented from experiments in laboratory animals the therapeutic effect of free cystein in intoxications by methyl chloride, chloroethanol, metallylchloride, methyl bromide, allyl bromide, methyl iodide, and ethyl iodide. The theoretical pre-requisite for its use in the intoxications by halogen-containing hydrocarbons stemmed from their ability to act as effective alkylating agents,

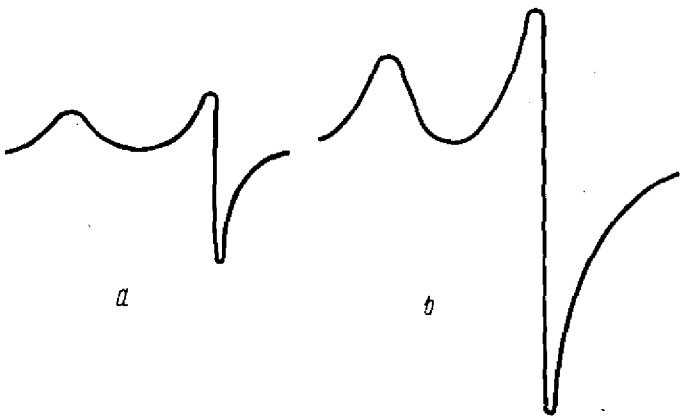


Fig. 8. Cytochrome R-450 induction following phenobarbital injection: EPR-spectrum (Ovsyannikova's experiment)

a - control; *b* - following injection of three phenobarbital doses (20 mg/kg)

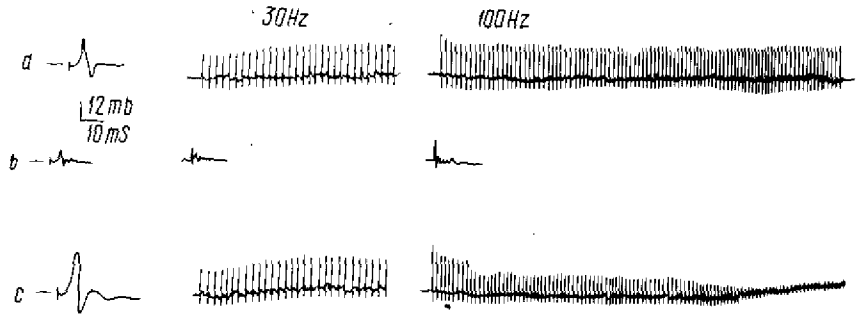


Fig. 9. Effect of phenobarbital in the origin of DDVP-induced neuro-muscular block (Kokshareva's experiment):

a – action potentials of rat musculus gastrocnemius stimulating its sciatic nerve by single and frequent stimuli (control); *b* – same, twenty minutes after oral DDVP injection (40 mg/kg); *c* – same, after preventive injection of three phenobarbital doses (20 mg/kg)

particularly in regard to the substances containing sulfhydryl groups. The action of unithiol was less obvious and monothiols (cysteine and acetylcysteine) were more active.

Their use increased survival of the test animals and restored sulfhydryl groups in the blood and liver. In dichloroethanepoisoned animals a positive effect arose with the acetylcysteine dose equal to LD_{99}. It also persisted at a high level when the treatment was commenced one or two hours after dosing. The therapeutic effect of acetylcysteine is explained by some authors in terms of its capacity for vigorous detoxification of chloroethanol, one of the principal metabolites of dichloroethane. The mechanism for the therapeutic effect of thiol compounds in intoxications by halogen-containing hydrocarbons of the aliphatic series must be inherent in their chemical interaction and production of slightly toxic precursors of mercapturic acids capable of being rapidly excreted from the body.

The antidotes of arsenic and metals in common use are unithiol, succimer (Arkhipova et al., 1975) and D-penicillamine (Ashbel et al.,

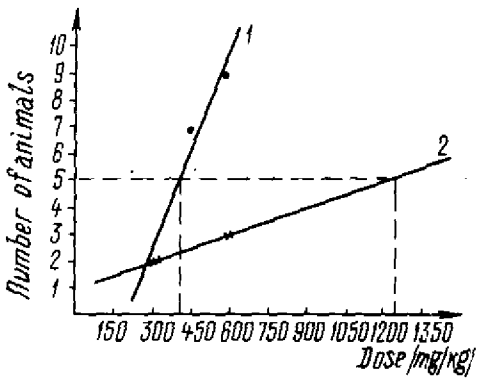


Fig. 10. Effect of phenobarbital on afugan toxicity in subacute poisoning (Kokshareva's experiment):

1 – total dose/lethal effect relationship in daily afugan administration (150 mg/kg); *2* – same, after four phenobarbital doses (20 mg/kg)

1974; Lubianova, 1979). In intoxications by hydrogen arsenide, the administration of mecaptide (2,3 dimercaptopropyltolylsulfide) has been the best remedy (Lugansky et al., 1966).

Intoxications by heavy metals are treated with a number of antidotes (EDTA/ethylenediamine/tetraacetate/,tetacin-calcium, pentacin, penicillamine) categorized as the complexones which combine with the metal ions to produce stable claw-like complexes (chelates).

Another effective way to treat acute pesticide poisoning, besides the specific antidotal treatment is through their faster elimination from the body. The means commonly applied to this end in the recent years have included forced diuresis, substitution transfusion, peritoneal dialysis, hemodialysis, hemosorption, and other resuscitative measures (Luzhnikov et al., 1977; Golikov, 1977).

LONG-TERM EFFECTS OF PESTICIDES ON ORGANISM

The problem bearing on long-term effects of pesticides is perhaps second to none in complexity. The effects are as hard to identify as it is difficult to model in experiment all the variables, extrapolate the resultant animal data to humans and commit the resources necessary for lengthy experimentation.

The long-term effects from pesticide exposures can be grouped into blastomogenic and mutagenic including effects of pesticides on progeny; effects on body organs and systems and their influence on the development of several pathological processes.

Blastomogenic Effect of Pesticides. The property of blastomogenicity is inherent in pesticides from different chemical classes. Low-level blastomogenicity was identified in the organochlorines DDT, aldrin, heptochlor, and metoxichlor (Chepinoga, 1970). Turusov (1975) confirmed carcinogenicity of DDT and its metabolites (DDE and DDD) in experiments on successive generations of mice (CF = 1). The males which received in the diet DDT doses from 2 to 250 mg/kg exhibited later an increased incidence of hepatic tumors, as did the females but only after the 250 mg/kg dose. DDE has been found more carcinogenic than DDT. Tomatis et al. (1972) reported varying incidence rates of hepatomas from DDT exposure, depending on the sensitivity of the strain of mice involved. It is important to note that examination of 1300 workers exposed to DDT during 10 to 20 years failed to indentify cancer in any one of them (Lawset et al., 1967). Thus it is difficult to estimate the real carcinogenic hazard of DDT for humans. But in view of the pesticide's cumulativity, i.e. the ability to accumulate in living systems, even its low-level carcinogenicity must be considered potentially hazardous.

Likewise, one finds conflicting data as regards carcinogenicity of the organochlorine insecticides of diene synthesis. Davis and Fitzugh (1962) concluded from experiments with feeding aldrin and dieldrin to mice that these substances are blastomogenic but nonetheless increase the incidence of benign tumors without transfer of metastases.

In the experiments carried out by Carbal (1972) heptachlor by intragastric injection caused tumors of different sites in female Wister rats. The author, though, would not consider these findings as proof of the agent's carcinogenicity by this route. Rather the carcinogenic activity of heptachlor must be due to its conversion into an epoxy form.

Walker et al. (1972) was feeding mice with various doses of aldrin and DDT and observed pretumor changes in the liver cells: nodular parenchymal growth and development of papillomas. After ceasing pesticide administration the morphological disturbances showed very slow recovery.

According to Didenko (1975), neither HCCH gamma-isomer, nor polychlorocamphene and polychloropinene are blastomogenic. Yet Tharpe and Walker (1973) cited their two-year experience of feeding mice (CF-1) with HCCH beta- and gamma- isomers to conclude that hepatic lesions could conceivably represent hyperplasia with the nodular tumor growth of parenchymatous cells on the pattern of papillomatous or adenomatous expansion. In individual cases the authors detected metastases in the lungs. Nonetheless Fishbein (1975) thought their evidence insufficient to classify HCCH as a carcinogenic substance.

Gibel et al. (1971, 1973) injected chlorophos intragastrically to mice at 30 mg/kg or apllied it to the skin in the same dose three times a week during five months. In the animals surviving more than six months after the protocol was commenced pathological changes of varying severity were detected in the liver. They included central necroses in the lobules, evidence of postnecrotic cirrhosis and hemorrhages, and tumors in some individuals. Gibel et al. (1973) noticed, on exposure to chlorophos and phosphamide, the rise of myeloid leukosis and malignant and benign tumors in some of the experimental animals. Lohs et al. (1974) attribute the carcinogenic action of dimethyl ethers of phosphorus acids to their alkylating properties.

Petrovskaya (1975) confirmed the data of the German authors, having found malignant and benign tumors of various sites and diseases of the hemopoietic organs in a large proportion of the C_{57} black strain of mice with low incidence of cancer and in Wistar rats after a dose of 30 mg/kg of chlorophos, orally or by epi- and subcutaneous application.

It remains to be known with certainty what lowest doses of chlorophos and phosphamide can increase the incidence of tumors and whether other organochlorines, specifically dimethyl ethers of phosphorus acids are also tumorigenic. It seems, no final conclusion about the potential carcinogenic hazard presented by these substances can be reached until tumor incidence data have been secured with human persons exposed to chlorophos and phosphamide over a number of years.

Experiments on laboratory animals discovered potential blastomogenic risks from several derivatives of dithiocarbamic acid and also TMTD, zineb and ziram (Chepinoga, 1970). Ethylene urea and thiuram caused tumors of the thyroid in rats.

Didenko (1975) found a correlation between the biochemical changes evolved in the pretumor period and the blastomogenic potential of urethane, DAB, ziram, zineb, and maneb. She reported increasing activity of deoxyribonuclease in the blood serum of noninbred albino rats in the pre-cancer stage of chemical carcinogenesis, concurrent with histologic changes developing in the absence of visible tumors. The lesions persisted as adenomas were appearing in the lungs. An increased deoxyribonuclease activity in response to low-level blastomogenic exposures (to derivatives of dithiocarbamic acid) showed later than with strong blastomogens (urethane).

The low-level blastomogenicity of sevin is still an issue. Chepinoga (1970) reports no evidence of tumors in X and C_3HX mice exposed to sevin and its tumorigenicity in noninbred rats. These results match other data on the low-level mutagenic potential of sevin.

The carcinogenic effect of triamino-1,2,4-triazole is proved beyond doubt (Chepinoga et al., 1968), as it is also in the case of some derivatives of symmetric triazines. In experiments on noninbred rats and C_{57} albino mice Pliss and Zabezhinsky (1970) recognized slight blastomogenicity of 2,4,6— trioxotrihydrosymtriazine and simazine. The substances were subcutaneously injected in a suspension or fed to the test animals (30 and 5 mg five times a week to rats and mice respectively) or applied by pouring on the skin a 20% suspension of the chemicals in benzene. The maximum summary dose totaled 20.58 g for the rats and 3 g for the mice. After 1.5 years less than 30 percent of the animals were identified as having tumors of various sites (fibroadenomas of the mammary glands, tumors of the stomach, hemangiomas and adenomas of the liver, myeloid leukosis, subcutaneous sarcoma, and lymphosarcoma of the lung). Though the tumour incidence was not high, some of them — subcutaneous sarcoma and hepatic adenoma — were not in evidence among the intact animals. The authors submit that the slight carcinogenicity they observed was associated with the structural features of symmetric triazines.

Low blastomogenicity of some urea derivative herbicides, particularly monuron, was reported by Chepinoga (1970) and Rubenchik (1970).

The classification used to evaluate pesticides for carcinogenic properties was offered by Shabad (1966) who separated all blastomogens into four groups: 1) overt carcinogens, known to cause human cancer; 2) strong carcinogens, known to cause tumors in various animal species by different routes of administration, but not carcinogenic in human beings; 3) mild blastomogens, known to cause tumors in a relatively minor percentage of the total population e.g. 20 percent, and at later dates — towards the end of their life-span; 4) suspected carcinogens, stated to be blastomogenic on the evidence that is either unconvincing or conflicting.

The carcinogenic substances of the first group include 2-naphthylamine, benzidine, some types of asbestos, chromates, arsenic and nickel compounds, and diethylstilbestrol, but no pesticides. While earlier on there used to be agricultural applications for some arsenic

compounds, e.g. sodium and calcium arsenites, calcium arsenate, and Paris green, they have since been removed from the list of registered chemical uses as being prohibitively toxic or carcinogenic. The second group comprises polycyclic aromatic hydrocarbons, nitroso-compounds, etc. In this group, consideration should be given to the possible synthesis of nitroso-compounds, particularly of nitrosoamines from nitrates and amines, both in the environment and in the alimentary canal of humans (Rubenchik, 1977). This circumstance is increasingly emphasized now that N-containing fertilizers are being used on a grand scale.

The third group is of the organochlorine pesticides DDT, aldrin, dieldrin, heptachlor, and metoxychlor; derivatives of dithiocarbamic acid — TMTD, zineb, ziram, and maneb; derivatives of symmetric triazines — symazine; the urea derivative monuron, and a host of other pesticides. These chemicals are potentially hazardous and some of them banned for use in agriculture (DDT, aldrin, dieldrin, ziram, and maneb) while the use of some others is strictly limited (heptachlor, TMTD, zineb, chlorophos, and phosphamide).

Hygienic regulation (limitation) of carcinogenic chemicals, specifically pesticides in the environmental media is an extremely complex area. The subject stirred wide-ranging discussions at several conferences and in the media. In one such discussion, following L. M. Shabad's report at the 4th National Scientific Conference on Pesticide Hygiene and Toxicology we (Kagan, 1968) stressed the importance of the dose — effect and time — effect relationship for the pesticides with blastomogenic properties. The Conference recognized the need for and feasibility of identifying pesticide doses from which the probable risk of tumor causation would be negligible. The pesticides of Groups 1 and 2 must be banned for use in agriculture and those of Groups 3 and 4 may be registered for use under clearly defined conditions (compliance with stringent hygienic standards). Albert (1975), Knizhnikov (1975), Shabad and Ilnitsky (1978), Kurliandsky and Nevzorova (1978) speak strongly for the possibility, indeed necessity to develop and enforce standards for environmental carcinogens. To accommodate this concern Maximum Allowable Concentrations (MAC) for benz(a) pyrene in the air of the working zone, ambient air and water (Ilnitsky, 1965; Shabad et al., 1973; Kurliandsky and Nevzorova, 1978) have been developed and approved in the USSR.

Sanotsky (1972) conceives the carcinogenic and blastomogenic effects as having a clear-cut threshold; he assumes that by defining the thresholds of blastomogenic action a theoretical base can be built for the hygienic standardization of carcinogenic substances. Pliss (1972) notes however that no dose of carcinogen, however small, leaves the organism unaffected and rejects on that basis the possibility of setting MAC for carcinogens because every concentration "contributes" to tumour development. The author proposes a series of chemical engineering measures to lower the carcinogenic risk of industrial toxics. With respect to the carcinogens found in normal plant and animal tissues, such as arsenic, selenium and so on, their na-

tural background can be taken as the maximum allowable concentration for them.

In theory, carcinogenicity is without threshold, because the interaction between several molecules of a substance and the cell DNA suffices to produce a mutation that can start tumor growth. In practice, however, it is always possible to decide on safe quantities of carcinogens from data of animal experiments (Sanotsky, 1972). One should keep in mind for such an experiment that it will commonly make use of limited experimental animal samples and therefore leave the possibility that tumors will appear as the sample size is extended. But given a high probability level of errorless judgement (for example, 0.999), the no-effect doses of carcinogens can be detected whose probability of tumors occurrence within the life-span is negligibly low (Kagan, 1978).

A method to determine the lowest effective amount of a carcinogen was proposed by Kurliandsky et al. (1978). It is based on locating the intersection point of the dose-effect curve with the baseline variation of the spontaneous background level according to the incidence of blastomogenicity in a given population. Different authors suggest reducing 50- to 100-fold the minimum effective dose of a blastomogen and stress the necessity to have a reliable safety factor.

Knizhnikov (1975) considers the intergration of safety factors in relation to threshold concentrations to be without foundation and insists on the design of standards for chemical carcinogens to be approached in the same manner as the radiation factor.

Pesticides may be suspected of carcinogenicity if they are mutagenic. The interrelation of carcinogenic, mutagenic, and teratogenic effects was perseived by Sanotsky (1972), Varshavskaya (1975), Turusov (1978), and Sanotsky and Fomenko (1979). Indeed, chromosomal aberrations are recognizable in tumor cells and carcinogenic compounds are all too often also mutagenic. However this parallelism between mutagenic and carcinogenic effects is often but not always the case. For one thing, some non-carcinogenic compounds have cytogenetic potential; for another, carcinogens do not trigger chromosomal rearrangements on all occasions. The presence of a mutagenic effect is therefore merely an impetus to proceed with carcinogenicity studies of chemical compounds. Also, the covalent conjugation of a substance with proteins and DNA may be an indirect due to its possible carcinogenicity (Rubenchik, 1977).

The hamster fibroblast culture has been widely accepted recently as the test system in primary screening of carcinogens (Turusov, 1978). In the opinion of some workers, it provides a 90 percent discrimination capability of carcinogens from non-carcinogens.

Because many carcinogens are simultaneously also teratogens the evidence of teratogenicity in a chemical prompts to start its investigation for probable carcinogenicity. Whenever a substance is teratogenic this attests to its ability to cross the placenta and may invite a study of transplacental blastomogenesis.

Khodosova (1976), Didenko (1977) and Rubenchik (1977) point to the predictive value of some biochemical alterations when they are detected.

Chepinoga et al. (1969, 1972) sought methodological approaches to the discovery of early biological tests in predicting tumor development and forecasting on a short-time basis the presence, or lack, of blastomogenic properties in a substance. The deoxyribonuclease activity in the blood serum of noninbred rats was found to go up in the precancer stage of chemical carcinogenesis, when histological changes develop in the absence of apparent tumors. The chemical blastomogenesis caused by urethane involved observable changes in the major chromosomal component, the nucleoproteid of cell nuclei, in the lungs and bone marrow of albino mice. These alterations, including an excess content of histones and an altered relationship between the histone fractions rich in arginine and lysine, emerge in the precancer stage and persist through the development of adenomas in the lungs. The deoxyribonuclease activity in the blood serum of noninbred rats is magnified not only by urethane exposure but also by exposure to the dithiocarbamic acid derivatives zineb, ziram, and maneb used as effective fungicides. The activity is at its highest by the time proliferative changes appear in the lung tissues. On the other hand, the increase of deoxyribonuclease activity fostered by low-level exposures to the blastomogenic pesticides that are dithiocarbamic acid derivatives comes somewhat later than with urethane injection. Based on these data, the authors state positively that the determination of deoxyribonuclease activity can provide a means for early detection of blastomogenic properties in chemical compounds.

Didenko and Gupalovich (1975) found that urethane, zineb, and ziram push up DNAse activity and cause hexokinase activity to appear in the blood serum of mice. None of these changes is present with lindane exposure. The former three substances are blastomogenic.

Rubenchik (1972) suggests that the similarity of the biological changes in the liver (rate of glycolysis, activity of gluco- and hexokinase, swellability of mitochondria) provoked by administration of monuron and known hepatocarcinogens is important for predicting the former's blastomogenicity. These results were borne out by data of morphological investigations. Monuron induced malignant tumors of the stomach, liver and lungs in the rat.

Rapid methods for detecting carcinogens are a strictly tentative guide and the detected positive effect merely warrants further chronic experiments. Conversely, the negative result of such tests does not necessarily rule out a possible expression of carcinogenicity.

Mutagenic Effect of Pesticides and Their Influence on Progeny. The influence of pesticides on progeny and future generations can be a variety of mechanisms. An important factor to note in this regard is mutagenicity of quite a few toxic chemicals enabling them to cause changes in the genetic apparatus of somatic and sex cells. Unlike the somatic cell mutations that are potentially hazardous to the host individual, the changes of sex-cell chromosomes and DNA can lead to

pathological alterations in the progeny and thereby endanger future generations and indeed the entire general population if mutagens become widespread. In addition to mutagenicity some pesticides can produce embryotoxic and teratogenic effects. The latter arise by placental transfer of the chemicals into the embryo organism. A particular hazard looms large from the substances with a capacity for selective action upon the fetus in doses non-toxic to the maternal organism.

Another adverse effect of pesticides is that which affects the reproductive function by impairing the gonads, interfering with normal maturation of sex cells, crippling the ability for fertilization and conception, and so on.

Contramination of the environment with mutagenic factors in the absence of appropriate regulations and controls threatens humanity with a genetic disaster (Dubinin, 1977).

The effect of mutagens may trigger gene or point mutations (breaches of DNA structures modifying the synthesis of various proteins and inducing enzymopathies), along with changes in the number of chromosomes and chromosomal abnormalities visible under microscope.

Weak mutagens, if widely spread in the environment, can pose a greater hazard than limited applications of strong mutagens (Rapoport, 1970).

Dubinin (1977) categorizes environmental chemical mutagens into three principal groups:

— natural inorganic (nitrogen oxides, nitrites, nitrates, lead, radioactive materials, etc.) and organic substances (alkaloids, hormones, etc.); processed natural substances (petro- and coal-burning products, etc.); and chemical products that do not occur in nature (pesticides, food additives, medicinal drugs, etc.).

He distinguishes among nine classes of chemical mutagens by the mode of action: alkylating compounds, peroxides, aldehydes, hydroxylamines, nitrites, antimetabolites, salts of heavy metals, dyes prossesing essential properties, some derivatives of the aromatic series including carcinogens, pesticides, drugs, and other chemicals. The agents in the pesticide category come under different categories of mutagens.

An important step in evaluating the genetic hazard of substances identifies correlation between abnormalities in the chromosomal apparatus of cells and subsequent development of disease states. This refers to both somatic and sex cells. Of additional importance with respect to sex cells is examining correlations between chromosome alterations in particular cells — because of the methodological difficulties associated with the study of chromosomal aberrations in sex cells. A substance is as mutationally active as the total number of the gene, chromosome, and genome mutations (changes in the number of chromosomes), induced by it.

The apparent numeric proliferation of all types of mutations with the increasing mutagen dose may occur unevenly in the sense that gene mutations have a greater proportion with low doses and chromo-

some mutations with larger ones. This is a factor to consider for the evaluation of chromosomal aberrations. With the aberrations present one is faced obiously with a symptom suggesting possibile mutagenic, blastomogenic, and teratogenic risks from substances, because there is a certain relationship traceable between specific chromosome alterations and these pathological effects.

Tumor growth sets off changes in chromosome cells, both qualitative and quantitative. It appears that the accumulation in the body of cells with chromosome lesions may well lead to their malignizations. Chromosomal alterations may also cause fetal death in uterus as well as a number of birth defects and hereditary diseases. Chromosomal anomalies in the embryos are frequent in spontaneous abortions and stillbirths. A fundamental requirement for weighing the significance of changes in the chromosomal apparatus dictates their comparison with the data directly obtained from several generations of laboratory animals.

Consequently, an excess of chromosome abnormalities due to pesticide exposure must be perceived as a sign of their potential hazard and consideration must be given to the levels at which chromosomal anomalies become detectable and the latter's magnitude.

Mutagenicity is not an all-or-none (quantal) property, for it has many quantitative gradations. In the assessment of a pesticide for real mutagenic hazard, its quantities that may enter human body should be compared with the doses capable of inducing chromosomal changes in the cells of exposed warm-blooded animals. Given the mutagenic effect — dose relationship, it is distinctly possible to establish the adequate pesticide doses to keep the incidence rate of mutations in the experiment at or below its level in the control. These doses can then be thought of as subthreshold and used as a guide in setting hygienic standards whenever a particular mutagen cannot be fully banished from man's living environment. To expect all mutagens to be banned right away is unrealistic, whereas the lack of hygienic standards for their environmental concentrations can merely disarm public health authorities.

In trying to characterize the hygienic significance of mutations, one must be aware that they can originate not only from direct interaction of a chemical agent with nucleic acids, but also as a consequence of the action that toxic substances exert on the cell's regulation mechanisms. Hence follows the impossibility for all substances to be divided on the all-or-none (quantal) principle into mutagens and non-mutagens. The extent of mutagenic activity and the specific conditions needed for its realization should be described on a case-by-case basis, with special attention to the likelihood of reparative processes, yet another factor to consider in the evaluation of risks from chromosomal alterations. Dubinin (1970, 1976) holds that primary molecular damages in the chromosome, which cause "perturbation" in the DNA structure (excitation, ionization, and breakaway of atoms or the radical) can be repaired there and then. Even if disturbances of the DNA structure should persist long enough to produce a pre-mutation

state the latter can still be repaired. In fact, the pre-mutation state and mutation may be a long way to go from one to the other, because mutations are characterized by already stable molecular rearrangements of DNA. There are a number of reparative systems acting to eliminate pre-mutations. Yet reparative processes, too, can seem to have a role in the origin of a mutation, for reparative enzymes may, by mistake, excise not the lesion itself, but rather the length of a normal strand lying opposite it.

Kurinny and Pilinskaya (1976) reveal that of the 239 substances they had investigated for the ability to cause a mutagenic effect, it was actually detected in 119, or 49.8 percent. The evidence that a given agent was mutagenic for some object is in itself not sufficient to state it to be a real hazard. Other relevant factors include the pesticide doses in which it was studied, the adequacy of a particular test system, the relevance of data extrapolation to humans, the magnitude of detected changes relative to the control, and several other variables.

For the majority of pesticides as well as other chemical substances the mutagenic effect is most commonly studied cytogenetically, by a metaphase analysis of the chromosomes. Failure to detect microscopically visible chromosome impairments is still no reason to rule out the possibility of gene point mutations, i.e. cell DNA changes not visible in the microscope. One does well, therefore, to supplement the cytogenetic methods with the studies to detect gene mutations. The assertion of a chemical having no mutagenic properties cannot be fully credible unless confirmed by a series of negative results from several test objects.

All we are having now is a set of guidance data on the potential mutagenic hazard of several pesticides. Those most thoroughly studied to date at the National Research Institute of Hygiene and Toxicology are: fungicides (derivatives of dithiocarbamic acid), herbicides (derivatives of 2,4-dichlorphenoxyacetic acid), organophosphate insecticides, acaricides, and some organochlorines (polychloropinene, polychlorocamphene, and dilor). A research into the genetic activity of dithiocarbamates on the Drosophila has identified the effect for TMTD alone, while zineb, ziram, and maneb were proved non-mutagenic (Kurinny, Pilinskaya, 1976). Oddly enough, the same authors have detected, in a study of bone-marrow cells in mice, an increased incidence of chromosomal aberrations in response to both TMTD exposure and the injection, subcutaneously and intragastrically, or ziram, zineb and maneb. Whilst in the control the metaphases with aberrations accounted for 0.46 percent, in intragastric exposure to the chemicals in a single dose of 100 mg/kg the incidence of the metaphases with aberrations rose to 3.13 percent for ziram, 1.4 percent for zineb, and 1.8 percent for maneb. In a study on the cytogenetic activity of zineb and ziram in the lymphocite culture of peripheral human blood in vitro (maximum zineb concentration 0.5 μg/ml and ziram 0.06 μg/ml) the incidences of the metaphases with aberrations for zineb and ziram were, respectively, 19.75 and 13.33 percent (up from 6 percent in the control).

In a cohort of workers occupationally exposed to TMTD, with the duration of employment from one to 10 years, a cytogenetic analysis established the number of aberrant blood cells to be 8.85±1.06 percent, up from the control aberration incidence of 0.75±0.43 percent ($P<0.01$). The TMTD concentration in the workers' breathing area was 1.3 to 3.4 mg/m^3 (MAC=0.5 mg/m^3). Among ziram-exposed employees the proportion of the cells with chromosome abnormalities averaged 5.9±0.59 percent, compared with 1.0±0.31 percent in the control. Its airborne dust concentration exceeded the MAC. The frequency rate of the metaphases with aberrations among persons in occupational contact with zineb amounted, on the average, to 5.98±0.41 percent in the first group (1.0±0.31 percent in the control) and 5.06±0.39 percent in the second group (0.89±0.31 percent in the control). The airborne zineb dust concentration also exceeded the MAC. Thus, the cytogenetic activity of the fungicidal derivatives of dithiocarbamic acid was confirmed both in experiments on bone-marrow cells of laboratory animals and in studies of chromosomal aberrations in vivo and in vitro. These data inputs were applied in setting hygienic standards for these agents, whereafter maneb and ziram were dropped from the list of pesticides registered for use in the USSR and TMTD was permitted for the only use as a seed desinfectant. For zineb, low permissible residual amounts were allowed in plant foods (0.6 mg/kg) and no residual amounts in milk and dairy.

Reports in the toxicological literature concerning mutagenicity of sevin (1-naphthyl-N-methylcarbamate) indicate that its cytogenetic effect was discovered in vitro experiments on plants, insects, and human and animal cells, and in vivo experiments on animals (Vashakidze, 1971; Brzhevsky, 1972; Vasilos et al., 1972). The herbicide 2,4-D was examined by Kurinny and Pilinskaya (1976) in experiments with human lymphocite culture in vitro and bone-marrow cells in mice by injection into the stomach. The highest rate of the metaphases with aberrations in the human lymphocite culture was detected on exposure to high 2,4-D concentrations (0.2 mg/ml and more), representing respectively 8.6±1.25 percent and 9.4±1.34 percent (1.75±0.47 percent in the control). The incidence of chromosomal aberrations was a function of the 2,4-D concentration in the range 0.2 to 0.002 μg/ml. In the bone-marrow cells of mice, the number of metaphases with aberrations correlated with the 2,4-D dose and exceeded the control (0.83±0.26 percent) following injection of 100 mg/kg (1.75±0.38 percent) and 300 mg/kg (3.08±0.5 percent).

Thus, 2,4-D was classed by the authors with mild mutagens on the grounds that in the concentrations that do not harm the lymphocite culture cells it has no cytogenetic effect, whereas its in vivo exposure does produce a cytogenetic effect in bone-marrow cells but only at toxic doses.

The cytogenetic activity of OPP was investigated in bone-marrow cells of mice by oral administration of the chemicals in different doses, and also in human lymphocite culture in vitro. A significant numeric growth of chromosomal aberrations was noted after the animals were

injected a chlorophos dose of 100 mg/kg and phthalophos dose of 20 mg/kg (2.70±0.51 percent versus 0.56±0.19 percent in the control and 2.0±0.46 percent versus 0.9±0.29 percent, respectively). Similar studies of abate, gardona, dibrom, carbophos, metaphos, methylmercaptophos, phosphamide, phosphamidon and cideal reported a significant rise in chromosomal aberrations only for metaphos and phosphamidon. Since the cytogenetic effect resulted from exposures to relatively large quantities of the agents being tested, it seems hardly reasonable to consider it specific.

The literature contains a number of references to mutagenicity in OCP. For aldrin, dieldrin, heptachlor, hexachlorcyclohexane (HCCH) and DDT, cytogenetic activity has been ascertained from experiments in laboratory animals (Markoryan, 1972). With intraperitoneal injection of the agents in the doses representing 4 to 6.7 percent of the LD_{50}, chromosomal anomalies in the bone-marrow cells of mice were several times their rate in the control. The author correlated the cytogenetic effect of DDT with its dose and found the correlation to be quite definitive in the DDT dose range from 1 to 10 mg/kg by intraperitoneal administration. Further increase of the substance's dose affected to a smaller degree the intensity of the mutation process. The author confirmed the cytogenetic potential of DDT in bone-marrow cell nuclei of monkeys giving them orally from 5 to 50 mg/kg of the agent for 26 days. The incidence of chromosomal rearrangements shot up to 2.6±0.7 percent at 12 days and 2.7±0.9 percent at 26 days, from 0.6±0.4 percent in the control. Markoryan's view of the organochlorine insecticides is as relatively mild mutagens for mammals. By the oral entry at doses upwards of 5 mg/kg they can scale up almost four-fold the incidence of chromosomal rearrangements in bone-marrow cells. The cytogenetic effect of polychlorpinene was discovered in the cells of testes in the mouse (Samosh, Kuzmenko, Bokotei, Altareva, 1976). The cytogenetic changes brought about by the agent's inhalation exposure accounted for 7.5 percent (with 0.83 percent in the control) and those by oral exposure for 4.3 percent. The authors indicate that the cytogenetic activity of polychlorpinene rests with its ability to easily pass up the hematotesticular barrier. Samosh (1974) found metaphases with chromosomal aberrations in the peripheral blood lymphocites of eight human subjects and showed them to increase markedly (an average 13.1 percent) following acute polychlorcamphene exposure.

Experiments on bone-marrow cells of mice proved incontrovertibly the cytogenetic impact of the herbicidal triazine derivatives (Kulakov, 1970). Similarly, symazine, etymidine, trietazine, chlorazine, atrazine, ipazine, and promethrin induced a significant increase in the number of cells with chromosomal aberrations — likewise observed in human lymphocites in vitro and in bone-marrow cells of mice on exposure to dinitroorthocresol — as a result of intraperitoneal injections to animals of the doses representing 1/30 LD_{50}.

Population prognosis is as important as individual prognosis to assessing the genetic hazard of pesticides (Bochkov, 1978). The

real genetic hazard of pesticides can be estimated with genetic monitoring as a useful aid. To make such an estimate, one has to bear in mind those 3 to 7 percent of the infants who are born each year in some countries of Europe and America with genetic congenital diseases. Dubinin (1976) cites the data of Sutten and Harris who estimated the spontaneous abortions caused by chromosomal aberrations to account for 30 percent of their total. The difficulty in estimating the speed and consequences of a mutation is that the same manifestation of a disease may be traced to several independent genes. A promising approach to exploring the state of the mutation process was developed by Altukhov (1974). Investigations into the distinctive patterns of protein polymorphism has led to the discovery of electrophoretically invariant monomorphic proteins which are biochemical markers for the genes as they occur in populations at a regular frequency reflecting the natural mutation process. A focused research on the proteins sheds light on the dynamics of mutation in the populations concerned. The method proves of advantage for comparison of genetic burdens between areas of intensive and limited pesticide coverage, with the goal of revealing, and estimating, the respective contribution of each in the rise of the mutation effect.

In assessing the mutagenic contributions of individual compounds and environmental factors in their combined exposure one must be aware that each of them merely adds to already existing background. Metabolic conversions are known to result not just in the formation of inactive metabolites — they can also cause new active mutagens to appear both in the environment and the organism as well. It is therefore not surprising to see researchers lavish attention on the test for mutagenic activity in microorganisms, now known as the host-mediated assay. The basic assumption is of the mutagen undergoing modification in the host organism. With several mutagens acting jointly on the body, their metabolism will likely change because xenobiotics interfere with the activity of oxidative enzymes in the liver cell microsomes to produce more active mutagens and enhance the detoxification processes. Dubinin (1977) refers to the characteristic "coholeness" (integrity) of the biosphere and the integrated impact by all of its constituent mutagenic factors. Here, all kinds of defence, additive, sensitizing, and qualitatively specific effects are possible.

A largely unexplored and fairly complex while also critical area of research in hygiene centers on the problem of chronic effects of minor quantities of mutagens, which for they are most often the case in real-life situations. The sheer urgency of rapid evaluation of the genetic risks from dozens if not hundreds of chemical compounds, new pesticides among them, rules out the possibility of their full-scale investigation within a compressed time schedule and on many test systems. Hence the current emphasis on the selection of several relatively simple test systems permitting at least tentative characterization of the genetic hazard posed by any particular compound. Dubinin (1976) maintains that a full-scale qualitative evaluation of a mutagen as to its degree of hazard for humans calls for the use of

four systems: a. point mutations in microorganisms with metabolic activation; b. dominant lethal mutation in mice; c. chromosomal changes in mammalian bone-marrow; d. chromosome injuries in human leucocytes. If necessary, the list can be restricted to the studies of bacteria with metabolic activation to identify gene mutations and cytogenetic research in human lymphocytes. The metaphase method to estimate chromosome rearrangements was thoroughly elaborated in Bochkov's laboratory (1972). It is important to note that the parallelism between chromosomal aberrations and gene mutations is not always present, still less in cases of exposure to low-level chemical doses, the most common single cause of gene mutations.

Several genetic-hygienic classifications have now been developed that identify pesticides by their potential mutagenic hazard. One such classification was proposed by Pilinskaya and Kurinny (1978).

Its underlying criteria are: 1. multiplicity of excess of the maximum effect induced by the test substance over the spontaneous control level for a single exposure to the substance; 2. minimal effective dose, estimated in weight units and in relation to LD_{50}. If so evaluated, pesticides can be categorized into several conventional groups with different potential hazards of genetic action.

Kuzmenko (1978) defines four hazard groups of substances by their doses as their main criteria, beginning from toxic dose according to toxicologic criteria and all the way to the doses existing in real life. The doses are going to be studied on several test objects in acute and chronic experiments. This offers a more comprehensive profile on chemicals but is also more demanding on time and information.

We consider the Pilinskaya and Kurinny classification suitable for use at the first stage of the research expected to come up with a rapid tentative evaluation of new pesticides for the degree of hazard; the Kuzmenko classification accommodates the second stage of a more detailed description of the chemicals that were cleared after the first stage. The second-stage evaluation will go a long way towards the all-round assessment of genetic pesticide hazard, especially when it employs such test systems as the blood lymphocyte culture of the human workers who were directly exposed to the pesticides concerned.

While examining the sequelae of mutagenic effects possible genetic adaptation must be considered. For all organic forms it proceeds by "dismissal" in natural selection of genetic victims and creation of progressive mutations on the basis of positive ones. However no genetic adaptation of human populations can occur as long as they stand exposed to the rapidly expanding use of industrial chemicals (Dubinin, 1977). The problem lies therefore in the goal-directed modification of the environment so as to make it impervious to contamination by mutagens (Bochkov, 1974). This imparts new urgency to the evaluation of genetic hazard and standardization of new pesticides, particularly those of them whose production in huge tonnages will be bound to affect general public.

Embryotoxic, Teratogenic, and Gonadotoxic Effects of Pesticides. Toxic chemicals become embryotoxic and teratogenic when they have

cleared the placental barrier and converged directly on the embryo tissues. This may be associated alternately with their toxic effect on the maternal organism. The chemicals known to adversely affect the fetus include presently about 600 agents (Kiriushchenkov, 1978).

Especially valuable and meaningful from the hygienic point of view are data on selective embryotoxicity and teratogenicity of pesticides, taken to mean here the effect on the embryo of such doses and concentrations of a substance that have no effect on the maternal organism. The difficulty of evaluating the embryotoxic and teratogenic properties of chemical agents and estimating their magnitude is actually the difficulty of constructing suitable experimental models and extrapolating the model data to man (Dyban, 1968).

With respect to embryotoxic (embryolethal) action, its characteristics include: total embryo mortality, pre- and post-implantation mortality (estimated by counting corpora lutea and live and resorbed embryos), mean litter size, average weight and size of embryos; with respect to teratogenic action, they are external and internal developmental anomalies, the state of ossification areas of the primordia of skeletal bones, and age distribution of offspring. Developmental abnormalities in internal organs are located by inspecting nonfixed embryo sections through a binocular lens and also with Wilson's method. Selective staining of the skeleton makes use of alizarin red, and biochemical changes in the embryo tissues are also integrated if required.

One of the prime objectives in studying the ways in which pesticides affect the reproductive function is to determine thresholds of their harmful effect and the latter's degree of selectivity (Medved et al., 1968; Sanotsky, Fomenko, 1979). Naturally, the most hazardous pesticides are those producing adverse effects in doses approximating their life levels and which selectively affect the fetus once they gain access into the maternal organism in the doses non-toxic to it. With these distinctive patterns in mind, Sanotsky et al. (1978) developed methodological recommendations for experimental procedures to establish thresholds of toxic effects of industrial toxic chemicals on the reproductive function. The thresholds to be determined relate to the total (integral) action on the maternal organism and also on embryogenesis. The ratio of the two thresholds is taken to determine the zone of specific (selective) effect. The recommendations set down the requirements to the selection of laboratory animals.

There is a large body of experimental evidence revealing embryotoxicity and teratogenicity of mercury compounds. The effect of ethyl mercury on pregnant cats is responsible for still-born kittens and compromised activity of the central nervous system (Matsumoto et al., 1965). Vashakidze (1970) found that granosan, if administered to pregnant rats at 0.05 and 0.1 mg/kg, interferes with the implantation of the ovum, thus increasing the still-birth rate. Pregnant rats inhalationally poisoned by mercuran concentrations of 0.03—0.04 mg/m^3 gave birth to pups with inhibited viability and retarted development. With the concentrations raised to 0.4 to 0.9 mg/m^3,

major irregularities were noticed in the course of gestation and resorption of the fetuses (Goncharuk, 1968).

Methyl mercury that enters water bodies in effluents and stores up in fish was responsible for Chisso-Minamota disease in the human persons who consumed the fish. In pregnant women with a previous experience of methyl mercury poisoning, the infants show developmental abnormalities (Harada et al., 1971).

Voronina (1971) established embryolethal action of phthalophos (the ester of phthalimide and dithiophosphoric acid) administered to females of noninbred albino rats every other day throughout gestation in a wide range of doses, from 15 mg/kg (0,1 LD_{50}) to 0.3 mg/kg (0.002 LD_{50}). An injection of 0.06 mg/kg phthalophos (0.0004 LD_{50}) failed to produce marked differences in the control versus experimental animals. There was, however, a distinct relationship between the phthalophos dose and its embryolethal effect. The dams in the test group which had been injected with phthalophos at 0.3 mg/kg gave birth to malformed pups (edematose trunk, constricted pelvis, maxillofacial abnormalities). In order to identify the period of embryogenesis with maximum sensitivity to phthalophos, the animals were exposed to a single 30 mg/kg dose at 3, 5, 7, 9, 11, 13 and 15 days' gestation time. The postimplantation death rate peaked at the 9th day

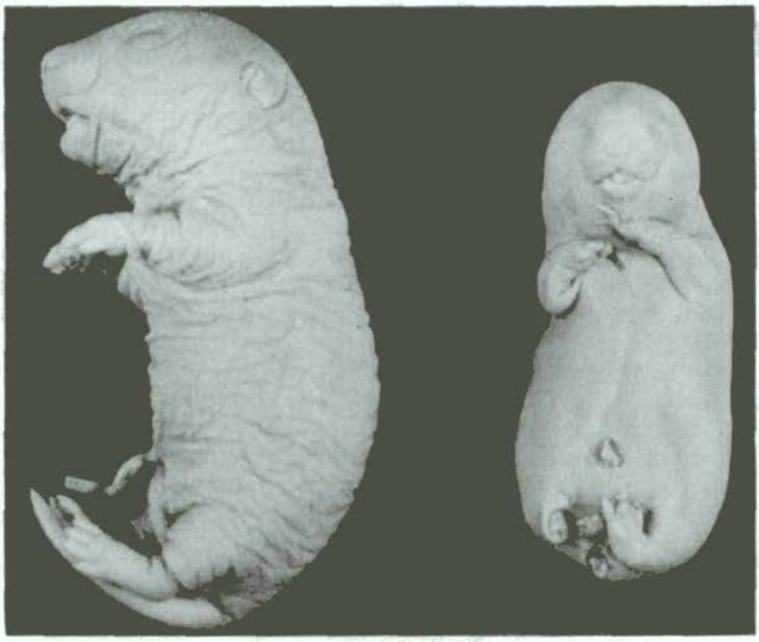


Fig. 11. Teratogenic effect of phthalophos (Voronina's experiment). General truncal edema and hypognathia in the fetus after single intragastric phthalophos injection to pregnant rat at 9 day's gestation time

of gestation when edematose trunks and solitary cases of scoliosis were noted (Fig. 11). Compared with rats, rabbits appeared less sensitive to phthalophos.

The pattern of phthalophos metabolism and the entry of its metabolites (chlormethylphthalimide, oxymethylphthalimide, and phthalimide) across the placental barrier were examined with a method of radioactive tracers.

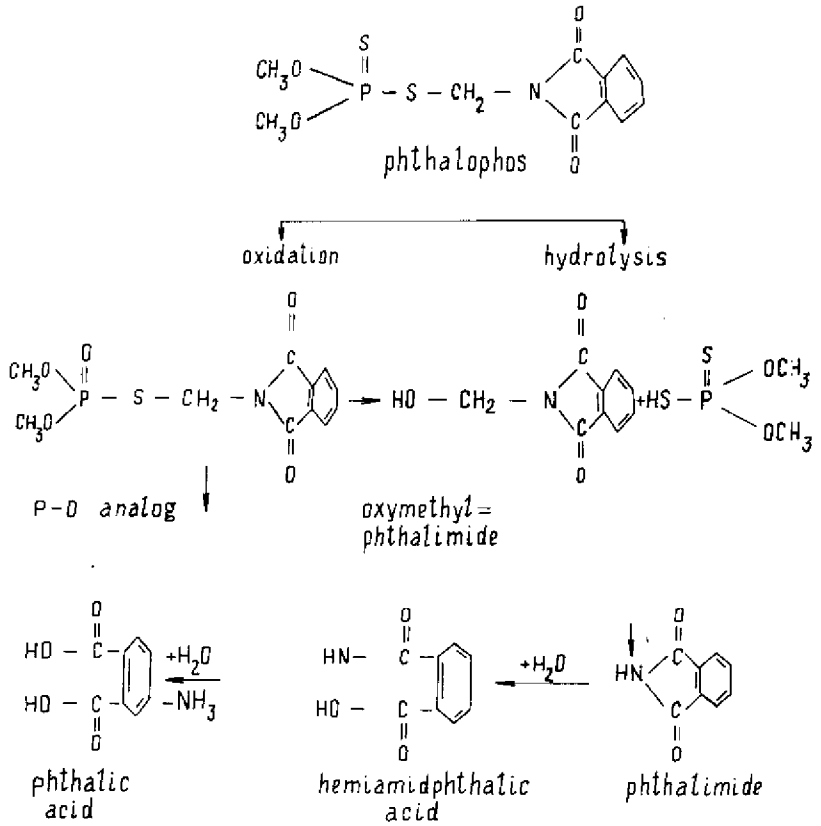


Diagram of metabolism in embryo tissues (Ackerman et al., 1978)

The presence of phthalimide in the rat uterus, placenta, and embryo tissues was conclusively shown by Voronina and Pismennaya (1973). The chemical was also found capable of embryotoxic and teratogenic action.

Data are available suggesting an embryotoxic and teratogenic potential for OCP. Of these, DDT and its metabolites have been proved to move through the placental barrier and DDT and HCCH gamma-isomer to expand the number of stillborn fetuses, retard physical development and cripple the viability of the progeny of the albino rats exposed to them during gestation.

The teratogenic effect of DDT in a dose of 170 mg/kg given to rats at different days through gestation was ascertained by Dinerman et al. (1970): it was manifested by subcutaneous hemorrhages and alteration of the bioenergy patterns in the embryo liver. Fertility disorders in rats were also observed in response to daily DDT administration from the first through 20 days' gestation time, concomitant with a reduction in the embryo weight and size, and hemorrhages and abdominal hernias in 45.9 percent of them.

Exposing pregnant rats to polychlorocamphene in daily doses of 12 mg/kg, Badaeva (1976) has perceived a relationship between structural and enzymatic changes in the neural elements of internal organs and the central nervous system. The chemical's concentrations in the organs of the fetuses attested to punctured permeability of the placental barrier and its suspected influence on the development of the nervous system.

Pentachloronitrobenzene, which involves about 11 percent of hexachlorobenzene, was administered to C 57 B1 mice in large doses (250 to 500 mg/kg) and seen to have caused the cleft palate and renal developmental defects in the embryos (Courtney et al., 1976). Hexachlorobenzene also finds its way into the embryo organism via the placenta (Mahelova et al., 1977).

Embryotoxic and teratogenic properties are known to be present in the derivatives of carbamic and thio- and dithio-carbamic acids. Sevin becomes embryotoxic when administered to albino rats in the dose range of 10 to 40 mg/kg (Voitenko, 1977). The mean embryo mortality per female was 0.9 to 1.4, up from the control 0.2. The progeny exibits functional deficiency and the threshold dose according to embryotoxic effect was 10 mg/kg. This feto-toxic action of sevin was further ascertained from investigations in rhesus monkeys, found to have aborted in response to the sevin dose of 20 mg/kg given after fertilization. At 3 to 50 mg/kg, sevin exposure of dogs throughout the gestation period brought observable malformations in 21 pups out of 181, namely, deformed skeleton, absence of tail, and underdeveloped jaw. The embryotoxic and teratogenic properties of betanal (3-methoxycarbonyl-aminophenyl-N-(3 methylphenyl)-carbamate) become evident after repeated injection to rats of 0.01 LD_{50} dose and inhalation exposure to the concentration of 30 mg/m^3 (Voitenko, 1977).

A comparative embryotoxic study of zineb, ziram and maneb led Martson (1967) to conclude that maneb is the most embryotoxic of them followed by ziram and then zineb. Distinctions between the three agents showed both in the magnitude of doses triggering disease states and the incidence of untoward effects. Embryotoxicity of zineb was registered at 9 and 13 days' gestation time (0.17 LD_{50}), as suggested by resorption of the embryo, death of the progeny, and retarded post-embryonal development. This is further corroborated by Ryazanova's evidence (1967, 1968). Zineb was additionally shown to cause developmental abnormalities, defects of skeletal bones, shortening and curvature of the tail, nanism of the pups; maneb

produced shortening of the tail and non-closure of the hard palate (Kaloyanova-Simeonova et al., 1967). Larsson et al. (1976) relate teratogenicity and embryotoxicity of maneb and mancozeb to the deficit of zinc that the fungicides produce in the tissues of the growing embryo. The teratogenic effect of maneb can be partially offset by simultaneous acetate injection.

The feto-toxic and teratogenic action of polycarbacin was established from administration in large doses (0.1 LD_{50}) to pregnant rats (Martson, 1971). Polymarcine was found feto-toxic at 5 mg/kg (Karpenko, 1977). Biologically active metabolites of dithiocarbamates, ethylenethiourea and ethylenethiuramimmonosulfide, are detectable in the placenta and amniotic fluid (Antonovich, 1977). Captan and phthalan cross easily the placental barrier to attack cells and parenchymatous organs in embryos (Vashakidze, 1976). For the former, fetotoxicity has been recognized from experiments in rhesus monkeys and New Zealand rabbits. The captan metabolite tetrahydrophthalimide is responsible for embryonal resorption (Zakordonets, 1977).

Teratogenic and embryotoxic benzimidazole derivatives include benlat, benomyl, fundazole, uzgene, BMC, derosal, and olgin. Benlat (benomyl), in albino rats, produces embryotoxic, teratogenic, and gonadotoxic effects increasing the total number of fetal deaths by reason of post-implantation embryomortality and the number of hemorrhages in the internal organs (the data of the Kiev Research Institute of Industrial Hygiene and Occupational Diseases; the research work is going on to identify the doses of exposure). Administered to Wistar rats from 7 through 15 days of gestation at 125 mg/kg, the agent induced anomalies of ocular development (micro- and anophthalmia), cranial development (hydrocephalus and encephalocele), and the cleft hard palate and upper lip. A benlat dose of 62.5 mg/kg induced changes in the rat tests (the data of the Nutrition Institute, USSR Academy of Medical Sciences). The evidence relating to its mutagenicity is somewhat controversial in that it has induced extensive gene mutations in microorganisms and some fungus strains and showed cytogenetic activity toward plants, mold fungus and human and animal cell cultures, causing the blockage of cleavage spindles and chromosomal nondisjunction; on the other hand, no dominant lethal mutations were observed in the sex cells of male rats. Usgene, the Soviet analog of benomyl, exerts a cytogenetic effect on the bone-marrow cells of noninbred albino mice.

As regards the teratogenic activity of benlat, Stenberg and Torchinsky (1976) watched it decline markedly when, prior to administration of the teratogenic doses, female rats had been treated with its subthreshold doses or those of DDT. The authors are inclined to attribute it to the induction of microsomal detoxifying enzymes in the liver.

Some herbicides appear to be embryotoxic, particularly the 2,4-D group of chemicals (Aleksashina et al., 1979) capable of making easy progress through the placental barrier (Antonenko, 1975). Buslovich et al. (1976) discovered a pronounced embryotoxic effect in the case

of 2,4-D butyl ester (increased embryo mortality and reduced embryo weight and size) and a somewhat less obvious effect in the case of 2,4-sodium and amine salts. Konstantinova (1975) reported no adverse feto-toxic effects of sodium amides, nor of butyl and octyl esters injected to pregnant rats in doses ranging from 7.1 to 16.5 mg/kg. Sokolova (1975) injected 416 mg/kg of 2,4-dichlorophenoxy-gamma-byturic acid to rats at 4– 5 days of gestation to find a higher incidence of pre-implantation deaths among the fetuses at that time and of pre- and post-implantation embryo deaths at 7 through 14 days. Rats exposed to 2,4-DM at 3.4 mg/kg throughout the gestation period exhibited developmental anomalies of their embryos.

Embryotoxicity was noted after multiple exposure of pregnant animals to 2,4-D at the rate of 5 mg/kg; 2,4-DM at 3 mg/kg, and sodium amides and butyl and octyl esters at 2,4-D at 0.7 to 1.65 mg/kg (Burkatskaya, Diadicheva, 1977). Repeated injection of butyl ester 2,4-D at 1/30 and 1/50 LD_{50} were seen to have caused in-utero deaths, still-births and developmental defects in the offspring. At 9 days' gestation time, rats exposed to the herbicides 2,4-D and 2,4,5-T (trichlorophenoxyacetic acid) showed various disturbances of embryogenesis, specifically hemorrhages into the abdominal cavity and development of hydrocephaluses. Of these, 2,4,5-T apparently owes its teratogenicity to tetrachlorodibenzodioxine present in it as an impurity (Moore, 1977), because its toxicity is acutely reduced by purification. Tetrachlorodibenzodioxine was observed to produce abnormalities in a dose exposure as low as 0.5 γ/kg. In the Soviet Union 2,4,5-T is banned for use.

The herbicide pyramine (1-phenyl-4-amino-5-chloropyridazone-6), as also some derivatives of dinitrophenol, show pronounced embryotoxicity and teratogenicity.

A prime objective of studies on gonadotoxic pesticide effects is to identify the dose — response relationship and determine its thresholds (as much probabilistic as possible). The ratio of the gonadotoxic and general toxic action thresholds suggests the degree of selectivity to be expected from the pesticide being tested.

Vashakidze (1970) presented convincing data about a major gonadotoxic effect of granosan. Chronic intoxication with ethylmercurchloride in doses of 1 and 0.2 mg/kg caused structural abnormalities in the testes and ovaries of experimental animals. Particularly notable were inhibited maturation of the follicules, death of the ova, and an increasing amount of the corpora lutea. In males, these alterations included death of spermatogenous epithelium along with other spermatogenetic injuries. Gonadotoxic properties of mercuran and mercurhexane were detected by Goncharuk (1968) in chronic experiments with inhalation exposure of albino rats to their concentrations of 0.03 to 0.04 mg/m^3. The affected animals had their reproductive function heavily impaired and physical development was retarded in the progeny.

Stenberg and Mametkuliev (1976) investigated, in a chronic nine-month experiment, the impact of HCCH gamma-isomer on the func-

tional state of sexual glands in rats. It transpired that a 0.5 mg/kg dose of the chemical kept down the rats' ability to reproduce. The progeny was underweight at the end of the feeding period. Injection of maneb in different doses was implicated in a variety of biochemical, histological, histoenzymatic and electron-microscopic irregularities of the gonads as well as a change of the reproductive capacity (Ivanova-Chemishanska et al., 1975). Spermatogenesis in rats was likewise affected by exposure to the symtriazine derivatives ipazine, promethrin, and semeron. A symazine dose of 62.8 mg/kg lowers the relative number of normal spermatogonia and heightens the number of tubules with a scaled-off embryonal epithelium.

OCP display more intensive gonadotoxicity than either carbamate or OPP pesticides. The gonads can be affected directly or indirectly — through a modified metabolism of the sex hormones in the liver.

Wozniak (1977) reports that lindane and trichlorfon (chlorophos) at doses equal to 1/15 LD_{50} are associated in rats, singly and in combination, with microstructural changes in the testes and modified activity of the enzymatic systems therein, as well as with spermatogenetic disorders. The author claims that his data is adequate to explain the cause of sterility and impotence in male workers occupationally exposed to these pesticides. It is well-known that DDT is the inductor for mixed-function oxidases vigorously involved in the metabolism of steroid hormones. Kupfer and Bulger (1976) indicate that stepped-up hydroxylation of steroids by chlorinated hydrocarbons of the DDT type is accompanied by a reduced effect of endogenous and exogenous steroid hormones, which interferes with the functioning of gonads. The estrogenous activity and the effect of DDT upon the reproductive function found confirmation in the work of other researchers (McBlain et al., 1977).

Polycystosis of the ovaries with a varying degree of change in the follicules up to the point of complete degeneration was reported by Haralamb (1976) following intraperitoneal 2.5 mg/kg DDT injections to rats five times a week for five months. Dikchith et al. (1978) cite evidence of degenerative injuries developing in the seminal ducts together with necrosis of the spermatogenic epithelium in response to hexabenzene exposure by dermal application to guinea-pigs in the dose range 100 to 500 mg/kg.

Dinerman and Shigan (1975) report DDT accumulation in the sexual organs of birds, assumed to affect their egg-laying capacity. Similarly, DDT and dieldrin act upon hormonal processes in mammals to modify their reproductive function.

The property of gonadotoxicity is presumably inherent also in the derivatives of carbamic, and thio- and dithiocarbamic acids.

Stenberg et al. (1970) established a severe effect of sevin on the state of sexual glands in rats, responsible for the reduced mobility and shorter life of spermatozoids in the males and modification of the estral cycle in the females. Also the males were more sevin-sensitive than the females. In the testis tissues, sevin exposure decreases the DNA content, interferes with enzymatic activity and triggers cytologic

changes. The gonads were seen malfunctioning after comparatively low-level exposure to sevin which, in addition, was more gonadotoxic than DDT. Vashakidze (1970) revealed a relationship between the dose of sevin and its effect on the gonads. She defined the threshold dose of the agent according to gonadotoxic effect (1 mg/kg) in chronic experiment. Daily exposure to this sevin dose resulted six months later in a modified estral cycle and diminished fertility, neither of which was detected in a 0.3 mg/kg dose exposure. The gonadotoxic action threshold for sevin was estimated by Vashakidze to be eight times less than its general toxic threshold, making readily apparent the agent's selectivity towards the gonads.

Ozhovan (1970) experimented with five generations of albino rats to see if sevin affects their reproductive function. At 2 mg/kg, the chemical had no effect in the parental and first generation but did affect adversely the generations that followed.

The severe gonadotoxic action of sevin suggested by these data has been part of the reason for limiting its uses in agriculture.

Lastly, the gonadal function is influenced by the dithiocarbamic acid derivatives: zineb, ziram and maneb, TMTD, etc. (Martson, 1967; Kaloyanova-Simeonova, 1977). The influence becomes manifest in conjunction with other effects — embryotoxic, teratogenic, mutagenic, and carcinogenic.

Long-Term Effects of Pesticides on Body Organs and Systems. Alongside the pesticide toxicity seen to take effect immediately after their single or multiple dose enters into the body, some of them have the ability to give rise to pathological processes (injuries of the nervous system — mental disturbances, pareses, paralyses; and of the cardiovascular system — coronary insufficiency, atherosclerosis, hepatocirrhosis, etc.) obvious in the long term. Such a pathological process may result also as the immune reactivity of the body is waning. Fridlyand (1966, 1971) considers low-intensity chemical exposures to be capable of increasing the incidence of hepatites, exacerbations of chronic diseases (hypertensive crises, peptic ulcers, etc.), and infectious morbidity (influenza, angina, pneumonia, tuberculosis, etc.).

Sós (1976) allocates to pesticides a large role in the causation of quite a few desease states.

In the recent years, information became available about systemic and specific effects of some chemical agents on the nervous system (Vievskaya, 1973). Nervous and mental disorders arose sometimes from OPP intoxications. The affected workers had difficulties of concentration and their speed of psychomotor responses was impaired. The allied mental symptoms may include dejection, schizophrenic syndrome, fear, and erethism. Their EEGs show fast high-voltage activity intermittent with bursts of high-altitude waves (3 to 6 oscillations/sec).

Zilber (1970), Aldridge and Johnson (1972) described OPP neurotoxicity highlighted by developing pareses and paralyses. Mipafox, DFP, triorthocresylphosphate, triorthotolylphosphate, phosvel, chlorophos, dichlorvinyldiethylphosphate and other pesticides affect the

axons of nerve cells to cause their demyelinization and to phosphorylate the protein. In hens per os exposure to phosvel (leptophos) resulted in the swelling of sciatic nerve tissues and fragmentation of the myelinic membrane, followed in 8 to 14 days by ataxia and in 13 to 30 days by paralyses. Similar changes were reported from investigation of the spinal cord in rats. Reduced velocity of impulse conduction via the sciatic nerve in rats was correlated with the destruction rate of its myelinic tunic (Kovtun, Kokshareva, 1980). Neurotoxic activity is known also for fenitrothion.

It seems that ataxia, paralyses and neuropathy 8 to 14 days after an OPP injection are not associated so much with the inhibition of cholinesterase as they are, chiefly, with the phosphorylation of the active center in the enzyme specific for the nerve tissue and now known as neurotoxic esterase. This type of OPP exposure was shown to increase copper concentration in the blood serum of test animals and send up the activity of ceruloplasmin. These data inputs have usefulness in predicting a neurotoxic effect. It is also believed that the OPP-inhibited neural membrane ATP-ase plays a role in triggering the effect.

Both OCP and OPP induce pathomorphological alterations in the thyroid and adrenal glands (Rodionov et al., 1976). Badaeva (1975) noticed phase changes in the thyroid and irritation of the adrenal system of the suprarenal glands in response to tetramethyl thiuram disulfide, lindane, and tigam. The dithiocarbamate zineb also contributed to the enlargement of the rat thyroid gland. Electron-microscopic studies into the gland's ultrastructure revealed its hyperstimulation, namely an 80 percent increase in the incorporation of ^{131}J from the control.

Adenohypophysis contained an increasing number of basophilic cells with PAS-positive granules. The authors perceived these disturbances as a compensatory response to the antithyroid action of zineb. An altered thyroid function was ascertained from data concerning the metabolism and distribution of ^{131}J in the thyroid on exposure to the dithiocarbamates of maneb and ethylenethiourea.

Administration of polychloropinene to experimental animals was followed by expansion in the size and relative weight of the thyroid itself but diminution in the size of its follicules, along with a greater height of the epithelium lining them and several other alterations suggesting in sum an abrupt surge of its functional activity (Grigorieva, 1975).

E. I. Makovskaya et al. (1975) noted, as a result of polychloropinene intoxication, depletion of the adrenal cortical substance, degenerative cell alterations, edema and minor focal hemorrhages, and focal cell death in the fascicular zone.

Several chlorinated hydrocarbons demonstrate the capacity to provoke agranulocytosis and aplastic anemia. Hemolytic anemia was seen to result from direct work exposure to a formulation of heptachlor, dieldrin and toxaphene, and also to zineb; persistent megaloblastic anemia was observed with intoxication by chlordane and porphyria

with intoxication by hexachlorobenzene. Feeding rats on a diet containing 0.2 percent of hexachlorobenzene led to gradually developing porphyria hepatica, with porphyrines and 5-aminolevulinic acid excreted in increasing amounts during the process. This went parallel to growing activity of anilinhydroxylase and cytochrome P-450 in the microsomal fraction of liver cells. The investigators assume the induction of microsomal enzymes to appear first, leading to an ever greater quantity of HCB microsomal oxidation products which are more active porphyrogens than hexachlorobenzene itself.

Komarova (1976) links some blood diseases to the effect of OCP. Among the 1083 patients examined for the causes of the various forms of leukoses, hypoplastic anemia, and diatheses those with a past history of lasting exposure to pesticides accounted for 17.4 percent — way up from the six percent of patients with other than hematologic diseases. In the hemopoietic organs of persons who died of leukoses or hypoplastic anemia DDT and DDE contents were higher than in the hypodermic tela. Among practically healthy producers and applicators of OCP, cytochemical changes in leukocytes were nonetheless detected. The unfavourable effect of DDT exposure on the induction of leukoses was borne out by experiments in rats subcutaneously inoculated with a leukotic cell suspension following chronic administration of the chemical.

Kagan, Lukaneva, and Rodionov (1974) looked at the effects of several pesticides in the progression and origin of several types of experimental cardiovascular pathology. Lukaneva in her experiments kept rabbits long-exposed to small doses of organochlorine (DDT, lindane, aldrin), organophosphorus (chlorophos), and carbamate (sevin) pesticides and then reproduced coronary insufficiency in them using intravenous hypophysin injections in small doses (0.35 units). In the control animals free from pesticide exposure, hypophysin caused the minor short coronary spasm normally observed in the initial forms of coronary insufficiency. Most of the rabbits given during six months a DDT dose of 0.001 LD_{50} were seen to develop in response to hypophysin atrial fibrillation and, in some of the experiments, electrical alternation of the ventricular complexes. When DDT or aldrin were administered in doses representing 0.01LD_{50} hypophysin injection resulted in an acutely enhanced intensity of coronary insufficiency, ventricular fibrillation and occasional animal deaths. Some rabbits emerged with observable evidence of infarction-like myocardial necroses (Fig. 12). Lindane administration in the same doses diminished the voltage of atrial complex waves, with a different shape and height of the P wave. Somewhat less obvious ECG changes were recorded with hypophysin administration to animals given sevin or chlorophos at doses equal to 0.01 LD_{50}. It was thus recognized that organochlorines and especially DDT and aldrin increased cardiac sensitivity on exposure to the coronary spastic agent by prolonged entry into the body. Verich described experiments in which organomercury compounds likewise modified cardiac sensitivity to the coronary spastic agents (hypophysin and vasopressin), and results of similar

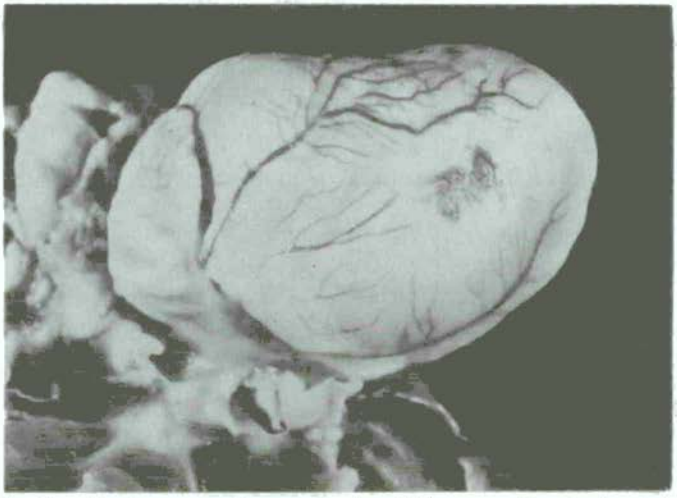


Fig. 12. Infarction-like transmural necrosis of rabbit myocardium induced by intravenous hypophysin injection (0.35 unit/kg) after daily administration of aldrin (Lukaneva's experiment)

experiments were reported in detail by Trakhtenberg in his review (1978).

Another type of a disorder reproduced in experimental animals was adrenal-caffeine myocarditis. Some of the rabbits given DDT daily at 0.01 LD_{50} exhibited more obvious ECG changes (tachycardia, appearance of a Q wave and a split QRS complex). There were differences in survival between the two groups of animals. Chronic DDT intoxication aggravated the course of myocarditis and pushed up the percentage of animal deaths (Lukaneva, Ivanova, 1970).

We, too, investigated the influence of OCP on the initiation of atherosclerosis. The latter was experimentally reproduced in rabbits by giving them cholesterol in sunflower-oil solution at 0.2 mg/kg during 2.5 to 3.5 months. The animals' characteristics that were subsequently estimated included the functional state of the myocardium (ECG), total cholesterol in the blood serum, lecithin, beta-lipoproteids, and total blood lipids. A pathomorphologic research was carried out also and it was found that cholesterol and DDT (1/20 LD_{50} for three months), by combined administration, bring on a higher and more rapid rise of the cholesterol, lecithin and beta-lipoproteid concentrations in the blood.

Chronic exposure to an isotoxic sevin dose of 1/20 LD_{50} was discovered to have produced more apparent alterations of lipid metabolism. A comparative pathomorphologic macro- and micro-examination of the aorta offered evidence on more persistent lipoidosis of the intima in rabbits exposed to cholesterol in combination with DDT and especially with sevin than in those treated with cholesterol singly. A parallel development involved the formation of extensive confluent plaques (Figs. 13, 14). The way in which cholesterol atherosclerosis

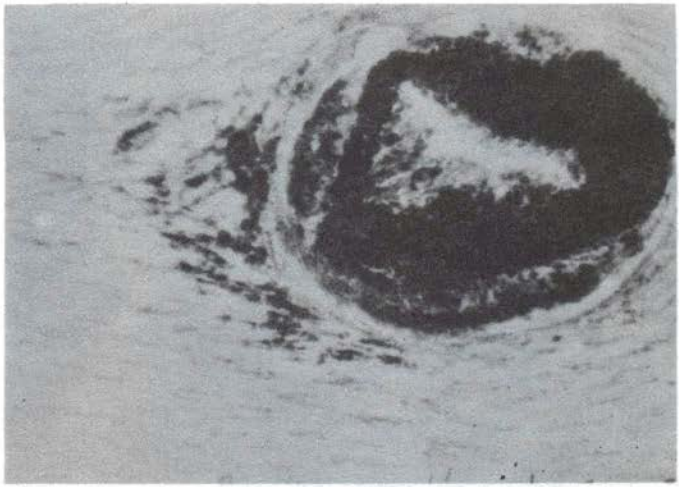


Fig. 13. Intramural branch of coronary heart artery. Marked enlargement of the intima and abundant deposits therein of neutral fat and cholesterol. Deposits of lipids (dark) beyond the internal membrane and in perivascular miocardial stroma. Staining by sudan III. ×90 (Rodionov's preparation)

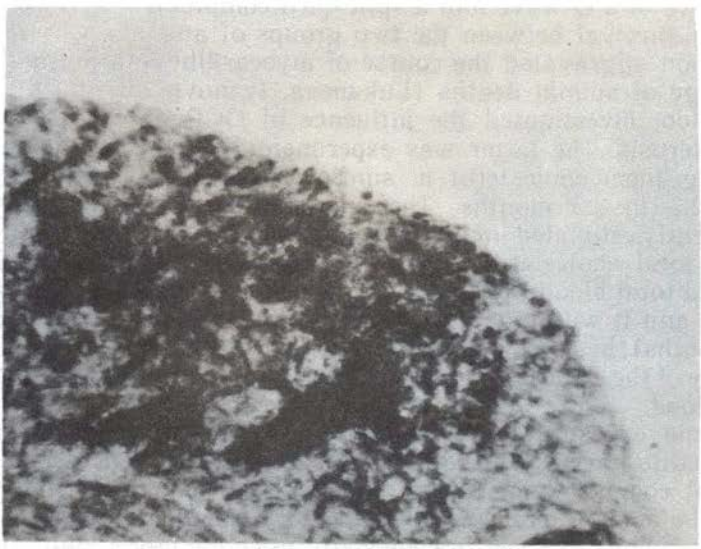


Fig. 14. Rabbit aorta. A large atheromatose plaque deep within the intima. Abundant deposits of neutral fat and cholesterol crystals. Staining by sudan III.×90 (Rodionov's preparation)

in rabbits is affected by DDT, lindane and chlorophos (0.01 LD_{50}) was studied also by their continued administration during eight months. It turned out that DDT had a more severe effect on lipid metabolism than lindane or chlorophos. Atheromatous changes in the intima of the aorta in response to joint DDT — cholesterol exposure were far more evident in its ascending portion than elsewhere. Deformation and luminal constriction of the heart vessels were also observed and necrobiotic alterations similar to infiltration processes were found to have developed in the myocardium. Lindane induced similar pathoanatomical changes though not as pronounced.

Some indices of lipid metabolism such as increased cholesterol and tri-glyceride levels are modified by lengthy polychloropinene exposure (seven months 0.25 mg/kg; Lukaneva, Reisig, 1976).

The findings of numerous toxicological and pathomorphological investigations asserted positively pathogenic effects on the liver from quite a few OPP chemicals. Kagan, Voronina, and Rodionov (1974) showed that in animals receiving orally a chlorophos daily dose of 0.01 LD_{50} for nine months carbon tetrachloride caused more severe changes of their liver, biochemical as well as morphological, than in the control. Grave changes in the antitoxic functions of the liver were identified (by a hexenal test), as was also an increase of alanine aminotransferase and sorbitdehydrogenase activity in the rat blood serum. None of these changes was in evidence on exposure to chlorophos or carbon tetrachloride acting singly. Administration of carbon tetrachloride in conjunction with protracted chlorophos exposure was followed by a decrease of glycogen and RNA quantities in the liver (histochemical studies) and then, in six months' time, distinct and widespread cirrhotic changes, adipose degeneration and adenomas in some of the rats. At nine months, these changes became obvious to a striking degree and destructive-degenerative injuries showed up in the kidneys, a proliferative process in the spleen, and circulatory disorders in the brain.

Attempts at mathematically modelling the behaviour of the liver in the toxicokinetics and toxicodynamics of toxicants are certainly appealing. Popov's studies (1977) with the model he devised of perfused rat liver gave him an opportunity to estimate in dynamics the activity of mixed-function oxidases (MFO) while maintaining the organ's structural and functional organisation sustained throughout; to study metabolisation of toxic substances by the liver to provide a mathematical description of biotransformations for a number of chemicals; and to identify the role of the liver in the processes of adaptation to toxic insults.

Derivatives of ethylenglycol are toxic agents damaging to the kidneys so that the paints containing the monobutyl ester of diethylenglycol cause lesions in the kidneys and liver.

Some compounds selectively affect the lungs, for example, dipyridilium derivatives, e.g. paraquat. Experiments in hamsters simulated pulmonary fibrosis due to paraquat injection. In rats, interstitial inflammation of the lung parenchyma followed a single dose of the agent injected intraperitoneally (15 mg/kg). Histologically, the

lungs were found to have thickened septa infiltrated by lymphocytes, histiocytes, fibroblasts and plasmocytes, and patches of atelectasis. Three paraquat doses administered at a weekly interval (5 to 10 mg/kg) induced observable pulmonary fibrosis.

Therefore, the data available to date give evidence that some organochlorines,. organophosphates, organomercurials and a host of other pesticides can and do influence the development of disease processes in the cardiovascular system, liver, kidneys, and lungs of experimental animals. This fact must be always considered in attempts to evaluate the potential hazard of the common as well as new pesticides, particularly those showing remarkable persistence and cumulativity.

COMBINED, CONCOMITANT, AND JOINT EFFECTS OF PESTICIDES

The system for protecting plants from pests, diseases and weeds employs during the year a large quantity and wide variety of pesticides. Some of these are applied concurrently and others successively, at different dates depending on the emergence timing of pests or weeds. There may be also involved combinations of pesticides acting in concert with chemical or physical environmental factors such as low or high temperatures, solar radiation, humidity, etc.

Combined effect refers to simultaneous or consecutive action upon the body of two or more chemical compounds or other, essentially similar factors. Concomitant effect refers to the entry of one or several different substances, simultaneously or successively, but by different routes (orally or dermally; through the alimentary canal or respiratory system; via the respiratory system or skin; or by all of the above routes at once). Joint effect takes place when the body is exposed to essentially different environmental factors, for example, poisons and high air temperature, or poisons and ultraviolet radiation.

Three distinct patterns are recognized for the combined action of substances: summation, potentiation, and antagonism. "Synergism" is an alternative term used occasionally though improperly, because it only describes an action as unidirectional and says nothing about its magnitude. The term is construed to mean an increasing influence, that is, potentiation though additive action is likewise an alternate type of synergism. References are sometimes made to the so-called independent action which has different points of application but fails to produce either summation or potentiation. The term is justly criticized (Kustov et al., 1975) on the grounds that there can be no independent effects of chemical substances on the organism while the term commonly applied in such cases is to denote actually incomplete summation of these effects, or antagonism.

The above terminology is usually employed in reporting the results of organism's exposure to different environmental factors physical and chemical.

A classic method of research into the combined effect of chemical substances is Loewe's method (Loewe, 1927). It consists essentially in building isodynamic diagrams by plotting on the coordinates of the test substances as fractions of the dose of each one of them that causes some recordable effect, for example, the death of the experimental animals (Fig. 15). Different dose combinations are tested for two substances so that the summary dose should amount, for example, to one LD_{100}. If the summation line coincides with the hypotenuse of the triangle that connects the points representing the LD_{100} of each poison, the action is additive. If two substance doses sum up to less than one LD_{100} but are sufficient to produce lethal effect the points of the effects in combined exposure will fall within the triangle and the action is characterized as potentiation. If, however, lethal effect is achieved with the summary dose of substances greater than one LD_{100}, the points corresponding to their combined exposure occur outside the triangle and their effect is defined as antagonism-relative if the isobol passes inside the triangle and absolute if outside. In some cases combined exposure may be of the synergo-antagonistic type — when some length of the effect's isobol passes inside and the remaining portion of making it outside the triangle. The method has the advantage of making it possible to assess the pattern of combined action by two substances in different combinations. It may be extended to include the effect of three substances but this requires building an isodynamic diagram in a three-dimensional space. Failure to evaluate the combined action of more than three substances and a comparatively large expenditure of laboratory animals constitute the disadvantages of Loewe's method.

In order to appraise the combined effect of two, three, or several chemical agents in any interproportions, it is useful to apply Finney's method (Finney, 1952). If the appraisal relies on the "lethal outcome" criterion, the LD_{50} values need to be determined for each of the test substances and then for their mixture; their respective quantities in the LD_{50} of the mixture are calculated and the percentage each quantity

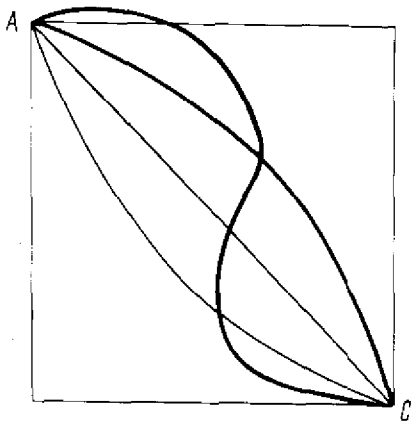


Fig. 15. Evaluation of combined effect by Loewe's method (see text for the explanation)

represents in the LD_{50} of every substance is computed; the percentages are then summated. Should the percentages sum up to more than 100, i.e. the total dose of the agents required to produce lethal effect exceeds the LD_{50}, this is antagonism. If, however, the doses sum up to less than 100 per cent but nonetheless are sufficient to bring the effect, this is potentiation. An effect equal to the calculated one (the percentages total 100) typifies summation.

The calculation by Finney's method can be written thus:

$$\text{if } \frac{A}{LD_{50(a)}} \cdot 100 + \frac{B}{LD_{50(b)}} \cdot 100 + \ldots + \frac{N}{LD_{50(n)}} \cdot 100 = 100\%,$$

then summation takes place;

$$\text{if } \frac{A}{LD_{50(a)}} \cdot 100 + \frac{B}{LD_{50(b)}} \cdot 100 + \ldots + \frac{N}{LD_{50(n)}} \cdot 100 < 100\%,$$

then potentiation is the case; and

$$\text{if } \frac{A}{LD_{50(a)}} \cdot 100 + \frac{B}{LD_{50(b)}} \cdot 100 + \ldots + \frac{N}{LD_{50(n)}} \cdot 100 > 100\%,$$

then antagonism occurs; where

A is the quantity of the substance "a" (mg/kg) in the LD_{50} of the mixture "a+b+...+N";

B is the quantity of the substance "b" (mg/kg) in the LD_{50} of the mixture "a+b+...+N"; and

N is the quantity of the substance "N" (mg/kg) in the LD_{50} of the mixture "a+b+...+N".

Assuming the problem is to estimate the combined action of chemicals A and B, their respective LD_{50} equals to 100 mg/kg for A and 200 mg/kg for B. The LD_{50} of the A and B mixture in the ratio 1:1 equals to 50 mg/kg. We find that the LD_{50} of the mixture comprises 25 mg/kg of substance A and 25 mg/kg of B. For A, 25 mg/kg represents 25% of its LD_{50} (100 mg/kg); for B it is 12.5% because its LD_{50} is 200 mg/kg. In sum the percentages make up 37.5. Consequently, the case here is potentiation with the potentiation coefficient $\frac{100}{37.5} = 2.6$. However when the sum of the percentages approximates 100, the combined effect of the pesticides involved becomes somewhat difficult to identify by type. Svetly (1972) suggested for such cases basic reliance on the confidence limits of the LD_{50} established with a necessary probability (commonly P=0.05). Under this approach it is possible in each specific case to decide at the accepted level of probability between summation, potentiation, or antagonism.

Finney's method is applicable for this kind of calculations provided the combined effect of the factors is expressed in the same measuring units (e.g. the effect of substance doses expressed in mg/kg). We took this method as a basis to develop a universal evaluation technique for a combined or concomitant effect of any two or more injurious environmental factors (Kagan, 1973). In essence, it involves estimating

first the injurious effect of each individual factor and determining its LD_{50} (or ED_{50}), and then evaluating the effect of their combination in any proportions (or of the same factor in simultaneous entry by different routes). The experimental LD_{50} (ED_{50}) values are expressed in fractions of the LD_{50} (ED_{50}) of each constituent factor and compared with the calculated LD_{50} (ED_{50}) for the case of summation. From the comparison emerges a quantitative estimate of the observed effect (the degree of potentiation or antagonism).

Quantitation of combined or concomitant exposure can be based on any quantal or graded index e.g. 50 per cent inhibition of an enzyme's activity. To do so, the dose that reduces by 50 per cent the activity of the enzyme (ED_{50A}) has to be found for the substance A; a similar dose for the substance B (ED_{50B}), and for the sum of the two substances $ED_{50(A+B)}$. The experimental data is then matched against the calculated values. The proposed technique permits quantitation of combined or concomitant effect of any two or several injurious agents.

A method used in the recent years is mathematical experimental design (orthogonal design, analysis of variance, etc.; Sova et al., 1974; Zlatev et al., 1977). Experimental design based on variance (dispersion) sets allows for a variance analysis of the results, separating the effect of each one of the factors being studied and of their interactions. A special difficulty with identifying the type of combined exposure arises in chronic experiments. They are staged in four groups of animals: control — group 1, exposure to the first substance — group 2, exposure to the second substance — group 3, and combined exposure to the two substances — group 4. The experiment in group 4 is designed on the principle of summation of the doses adopted for groups 2 and 3, and not their half-doses. Usually the effect is proportional, not to the doses, but their logarithms and therefore the contention that the added effects of a half-dose of the substance A and a half-dose of the substance B will be equal to the sum of the half-effects of either substance is fallacious.

In order to evaluate the experimental results it is good practice to apply variance analysis of bi-factor proportional sets for qualitative attributes of small groups (Plokhinsky, 1970; Sova et al., 1974). Its use in this application provides information on various aspects of the effects under study, such as their magnitude as coming from either of two isolated factors and from their joint action; the significance of either's action, and so on. In addition to estimating the factor-related effects, it reveals also the random influence of extraneous (non-organized) factors. The farther does the factor-related effect depart from the random effect, the more significant it is. The summary effects of organized plus non-organized factors determine the total variability of an attribute, or its dispersion. The method is advantageous in that, with no increase in the amount of work to do, it provides the investigator with a source of information as to the intensity of the overall effect of these factors on the indexes of concern and the extent of interplay between these indexes. Yet the specific application

of variance analysis to evaluating the results of combined exposure in a chronic experiment disallows him to avoid some difficulties inherent also in the use of other approaches. Since the action of chemical agents is explored at comparatively low levels that fail to cause animal deaths, the criterion in evaluating the effects relies on a variety of physiological and biochemical indexes reflecting the effects of the test substances and their combinations. Not infrequently, changes of these indexes work in different directions.

Still another method, besides analysis of variance, is regression analysis. Multiple regression analysis is of particular value for the toxicological assessment of the combined action exerted by a set of volatile substances when the composition of chemicals released into the atmosphere is not fully known (Nagorny, 1973).

All types of combined effects from pesticides exposure, found in animal experiments can be confined to three basic alternate patterns: summation, potentiation, and antagonism. Summation of toxic effects in warm-blooded animals has been defined in a single exposure to pesticides categorized in the same class e.g. chlorophos and phosphamide (Kagan, 1965), or chlorophos and metaphos (Junusova, 1966) or in different chemical groups, for example, sevin and butyphos or sevin and cyanamide (Demidenko, 1968) or TMTD, HCCH gamma-isomer, and copper trichlorophenolate (Svetly, 1970). Zlatev (1976) observed additive action, according to the lethal outcome criterion, resulting from a single injection to albino mice and rats of the organochloric acaricide kelthane in combination with phthalophos, metaphos or phosphamide or the dinitrophenolic pesticide acrex in concert with phthalophos or phosphamide.

Synerphos-4-nitrophenoxydiethylsulfide, a synergist for some organophosphates on insects, yields the effect of incomplete summation rather than potentiation in warm-blooded animals (Slobodkin, 1975). These differences in the characteristic modes of combined action of pesticides on insects (potentiation) and warm-blooded animals (summation or antagonism) open up a prospect for designing combinations of chemicals having selective insecticidal potential.

Kustov et al. (1975) note with regard to the effects of chemicals from different classes that their summation occurs relatively seldom if at all.

Additive action of pesticides has been established also in chronic experiments. Not infrequently, this involves methodological difficulties, owing to the need to evaluate the pattern and type of combined effects of substances at low levels of exposure. In acute experiments it is possible to estimate such an effect from the lethal outcomes it causes – the most integral of all available criteria. Not so in chronic experiments where one has to record a set of integral and, if possible, specific indexes relating to the effect of substances and draw conclusions carefully weighing the biological significance of the studied indexes. Thus Junusova (1966) found that a chlorophos-metaphos mixture in chronic experiments causes more severe disorders in animal behaviour and the state of several biochemical and morphological

characteristics of the blood than independent administration of either chemical does alone. However the author failed to further qualify that action as summation or potentiation.

Slobodkin (1975) reported a mode of combined action by a mixture of synerphos with methylmercaptophos in chronic experiments consistent with incomplete summation and by a mixture of synerphos with intrathion, with simple summation. With chronic exposure to fenthiuram, the chemical composed of TMTD, HCCH gamma-isomer and copper trichlorophenolate, the changes in the organism of test animals are less obvious than with single exposure to each of the constituents, revealing a tendency towards incomplete summation (less than additive) of the effects, or antagonism (Svetly, 1970). It is appropriate to remind that in acute experiments the same combination produced the summation effect. Zlatev (1976) points out also that in chronic experiments the combined effect of kelthane plus metaphos amounted to summation with some tendency towards antagonism as contrasted from the summation effect identified in acute experiments.

Thus, when summation is registered in acute experiments it may either persist or become incomplete in chronic experiments (the so-called independent action or a tendency towards antagonism)

Sanotsky (1968) reckons that at the level of lethal concentrations, one is likely to observe all types of combined toxic effects — summation, potentiation and antagonism. At threshold-level concentrations summation or "independent" effects will most typically come about. This point of view is not without some theoretical validity, as potentiation or antagonism are most often associated with the effect produced by one of the ingredients on the enzymes metabolizing the other ingredient. For the effect to be tangible, sufficiently severe exposures are required.

For all this, the prediction of chronic effects from low-level combined exposures presents a complex problem whose solution should be approached with care.

Potentiation constitutes the most hazardous type of combined action and its accurate recognition is essential (Kagan, 1977). Cases of potentiation of insecticidal effects on insects have been documented. For example, sesamex inhibits the oxidation of vinylphosphate and enhances its toxic effect on insects (Medved, Kagan, 1962). This is also the key principle for the use of some synergists. Because in warm-blooded animals the processes of pesticide metabolism may be different from those in insects, the use of synergists presents one of the alternatives in increasing selective toxicity of insecticides.

Potentiation of toxicity in case of combined exposure to some pesticides was ascertained in acute experiments on warm-blooded animals. Thus, HCCH and TMTD gamma-isomer, and also HCCH gamma-isomer and heptachlor, were found to produce a combined effect of the potentiation type in experiments on rats and cats (Osetrov, 1962). HCCH gamma-isomer potentiated TMTD in inhalation-exposed rabbits (Voitenko, 1962). Heptachlor is known to transform in the organism

into the more toxic epoxide (lethal synthesis). Presumably, the potentiation of heptachlor toxicity by HCCH gamma-isomer is due to the latter's inductive effect on the mixed-function oxidases present in the microsomal fraction of the liver, which are involved in the epoxidization of the toxic chemical.

There is also evidence for the potentiation of alcohol toxicity by tetramethylthiuram disulfide and tetraethylthiuram disulfide, owing to the inhibitory effect of the dithiocarbamates on the latter's oxidation. In some cases, potentiation identified in acute experiments persists in chronic ones, as in the case of chronic administration to cats and rats of HCCH gamma-isomer and TMTD (Osetrov, 1962) or sevin and butyphos (Demidenko, 1968) or anabasinsulphate and thiophos (Lazutka, 1973).

Antagonism is a rather common alternate pattern of interaction among pesticides. Cases of antagonism were documented in the interaction of organochlorines with organophosphates (Spasovsky, Tsonevsky, 1974; Zlatev, 1976). Zlatev (1976) observed antagonism following exposure of the organism to a combination of organochlorine and organophosphate acaricides (kelthane with phosalone or anthio; milbex with phthalophos, phosalone, phosphamide or anthio). Yakusko and Zlatev (1975) found the organochlorine acaricides kelthane and milbex to be inductors for mixed-function oxidases (MFO), the degree of induction in liver microsomes correlating well with the degree of antagonism between organochlorines and organophosphates. Giving milbex together with phosalone resulted in the coefficient of antagonism by LD_{50} equal to 5.6 ± 0.84 (Fig. 16). Demethylation of aminopyrine, which is the measure of MFO activity in combined milbex-phosalone exposure, increases 5.3 times, thus showing a correlation with their degree of antagonism. Demethylation activity is increased with milbex administration in large doses (1/5 LD_{50} three times), but also with its daily administration to animals at 1/100 LD_{50} (Kagan et al., 1975). This evidence confirms that the antagonism of organochlorines and organophosphates depends on the OCP ability to accelerate OPP detoxification in the organism of warm-blooded animals. Since in the insects other ratios may occur in the interaction of these compounds, simultaneous or alternate application of organochlorine and organophosphate acaricides leads to a system of their use increasing their effectiveness as acaricides (the OCC-OPC alternating use makes less probable the emergence of OPP- or OCP-resistant ticks) and lowering the hazard to humans and animals.

We investigated the impact of phenobarbital and milbex, both of them inductors for mixed-function oxidases, and tetramethylthiuram disulfide (according to Kuzmenko et al., 1977; it inhibits the MFO activity by 50 per cent) on the toxicity of several organophosphates (Kagan et al., 1977). It has been found that the reduction of mixed-function oxidases considerably decreases the toxicity of the organophosphorus compound NSh-6 containing two carboxyl groups in its molecule (the LD_{50} for rats in the control and experiment equal to 95 ± 13 mg/kg and 155 mg/kg respectively). The liver MFO activity

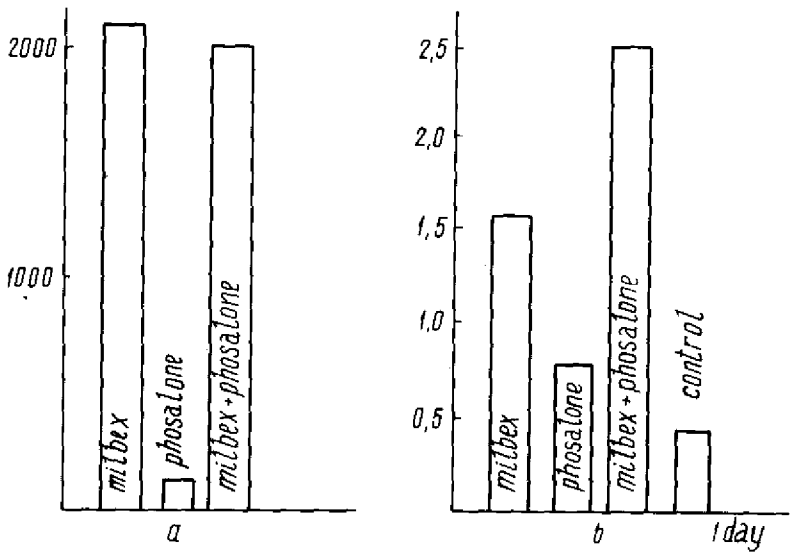


Fig. 16. Milbex/phosalone antagonism in rats and liver MFO activity (Zlatev et al.): *a* — LD_{50} value (mg/kg) (coefficient of antagonism 5.6); *b* — MFO activity (g·m/g) after triple injection of chemical (0.2 LD_{50})

in the control was 0.8μ M/g of the tissue and in the experiment, 2.4μ M/g. Administration to the animals of the MFO inhibitor TMTD had no appreciable effect on the NSh-6 toxicity. Apparently, the induction of mixed-function oxidases intensifies OPP detoxification. Yet its effect on the toxicity of different organophosphates is by no means un-equivocal and neither is the role of MFO in the metabolism of organophosphates. Increased MFO activity promotes detoxification as well as more rapid lethal synthesis (oxidative desulfuration). The relation between these phenomena varies with the OPP involved, and therefore any alternate patterns are possible as concerns the influence of the MFO inductors and inhibitors upon toxicity. For the same reason, all predictive exercises must be designed with due attention to the chemical structure of organophosphates (the presence of the P=S group in the molecule), as well as their metabolic pathways. There may be also wide interspecies differences in metabolism which can well be put to advantage in looking for selective pesticides.

In combined pesticide exposure antagonistic relations show themselves in not just the acute experiment, but in the chronic experiment as well. According to Svetly (1970), HCCH gamma-isomer, copper trichlorophenolate and TMTD, the substances including the combined fungicide fenthiuram, show a moderately antagonistic pattern of behaviour in the chronic experiment. Spasovsky (1962) observed antagonism between the organochlorines DDT and HCCH and the organophosphates systox, ecatin, and parathion. Zlatev (1976) classifies the combined action of milbex and phosalone in the chronic experiment

as antagonism and of kelthane and metaphos as summation biased toward antagonism.

It is hence inferable that, while in acute experiments the type of combined effect that arises is either summation or antagonism, the manifestation of potentiation is hardly likely in chronic exposure. The inference is important hygienically, for it suggests that chronic experiments are mandatory in all those instances when cases of potentiation were recorded in acute experiments.

In order to predict interaction of toxic substances on the potentiation principle it is essential to know their specific patterns of metabolism and identify incidents of one substance inhibiting detoxification of the other constituents in the combination. Less frequent is the type of potentiation through activation by one constituent of another's lethal synthesis. Nevertheless both possibilities must be considered if cases of potentiation in the action of various pesticides on the body are to be anticipated in good time.

An important question centers on quantitative evaluation of environmental status in combined toxic exposure. The USSR National Standard 12.1.005-76 "The Air of the Working Zone" specifies that, with several harmful substances of unidirectional action present at the same time in the air of the working zone, the total ratios of the actual concentrations of each chemical (C_1, C_2 ...C_n) in the indoor air to their respective MACs (MAC_1, MAC_2...MAC_n) should not be greater than unity:

$$\frac{C_1}{MAC_1} + \frac{C_2}{MAC_2} + \ldots + \frac{C_n}{MAC_n} \leqslant 1.$$

Given simultaneous presence in the air of compounds with other than the unidirectional mode of action, the MAC remains the same as it would be for each one of them acting singly. It remains debatable whether correction factors need to be introduced to account for the degree of attenuation of the toxic effect by the phenomenon of antagonism (Kustov et al., 1975). We are inclined to concur with Sanotsky et al. (1968) in that MACs should be adopted for each substance without any corrections for cases of antagonism. Potentiation, of course, is a different matter for then its coefficient needs to be determined and a correction introduced into Averyanov's formula, giving it the form (Kagan, 1973):

$$\frac{C_1}{MAC_1} + \frac{C_2}{MAC_2} + \ldots + \frac{C_n}{MAC_n} \leqslant \frac{1}{K_{potentiation}}.$$

Concomitant and Joint Effects of Pesticides with Physical Environmental Factors. Concomitant effects imply the action of pesticides by entry into the body from different media. The fact is that toxic chemicals can access the organism in air, via the skin, and in food and water. A question arises how to evaluate the concomitant effect of pesticides according to type and whether summation occurs as they enter by different routes or merely deviations from the rule of additivity?

Voitenko (1978) found concomitant entry of lindane into animal organism via the respiratory system and the stomach to bring no major

differences in the variation of the indices being studied as compared with its separate entry by either route. The author proposed further to make summation of the effects the basis for integrated hygienic standardization of the pesticide. But, when studying to concomitant entry of phosphamide via the respiratory system, skin and alimentary canal, Voitenko and Larionov (1976) identified more significant alterations of the morphological and functional characteristics of the peripheral blood than by each route, alone. Also, the anticholinesterase effect of phosphamide was magnified.

Research on the concomitant action of pesticides employs methods of simplex experimental design, and of orthogonal design with processing of the results by the regression method (Spynu et al., 1975, 1978). The authors concluded on the interrelation of the dermal and oral entry of organophosphates into the animal organism. For example, the effect of chlorophos can be characterized as summation with a tendency towards antagonism. Based on such data, it is possible to predict the extent of inhibition of blood erythrocyte cholinesterase by the concomitant action of chlorophos.

The question arises: how does one assess environmental status if the same pesticide can enter the body in different media – air, water, and food? Korbakova (1972) considers it useful, in order to get a notion about the hazard posed by concomitant action of environmental chemicals, to apply the toxicity summation formula:

$$\frac{C_{air}}{MAC_{air}} + \frac{C_{water}}{MAC_{water}} + \frac{C_{food}}{MAC_{food}} \leqslant 1.$$

Should potentiation be detected in the concomitant action of a chemical, the formula must be properly corrected by dividing unity by the potentiation coefficient.

Evaluating the joint effect of pesticides with physical environmental factors presents an important if challenging problem. High temperature elevates pesticide concentrations in the air of the working zone and the absorption rate of chemical substances from the skin surface. Yakubov (1964) notes that a high ambient air temperature (34 to 38°C) increases the toxicity of some organophosphate insecticides (intrathion, methylmercaptophos). Denisenko et al. (1968) indicate that the bodily response to the joint effect of elevated temperature with chlorophos is different in quantity and quality from the response to either factor alone. Their single exposure enhances the toxicity of chlorophos. Two important factors therein are the time of the high-temperature exposure and the intermitting effect of temperature.

Iskanderov (1972) found that with high temperature exposure the toxic effect of methylmercaptophos, butyphos, kilval, anthio, milbex, monuron, and sodium fluosilicate in orally dosed albino mice and rats becomes 1.5 to 2 times higher. The author showed that the effect of temperature creates a new set of conditions, modifying the intensity of pesticide absorption, distribution, metabolism and elimination. In its turn, the toxic effect of the chemical agents may dislocate the tolerance of organism to high temperatures.

Demidenko and Mirgiyazova (1974) reported enhanced toxicity of the organophosphorus pesticide anthio at 36 to 40°C. They consider it necessary, therefore, that the pesticide MAC be lowered 5- to 10-fold in hot climates.

It is generally recognized that the uncoupling of oxidative phosphorylation and subsequent hyperthermia constitute the essential mechanism for the action of dinitrophenolic pesticides. Between them, they trigger the hazard of joint action by dinitrophenol derivatives and high temperature exposure (Burkatskaya, 1974). Panshina (1977) established a 1.5 to 2 times higher toxic effect of herbicides (haloid-substituted anilides of carboxylic acids) with elevated ambient temperature. It is therefore necessary, in setting standards for the pesticides identified for use in high-temperature environments, to incorporate larger safety factors.

Effects of pesticides are susceptible to air humidity, barometric pressure, radiant energy, ionizing radiation, and physical stresses.

Yusupov (1975) investigated ways in which the factors associated with high altitudes influence the toxic effects and cumulative properties of some organophosphates, organochlorines, and organomercurials. Their enhanced toxicity with elevation is explained by the author as being due to hypoxia induced by oxygen deficiency in the mountains and by the action of pesticides. His findings agree with the data of Tiunov and Kustov (1969).

Gabovich et al. (1975) have studied the joint effect of hexachlorobenzene (HCB), temperature and ultraviolet radiation (UV). He found low tolerance of the organism to hexachlorobenzene with overheating and intensive UV-radiation. However, inadequate UV-radiation also increases HCB toxicity, thus leading the authors to believe that maximum tolerance of organism to HCB comes with optimum UV-radiation.

UV-radiation may also affect the photochemical conversion of pesticides in the environment. Klisenko et al. (1977) examined in the laboratory the impact of UV-radiation and temperature on the processes fostering the formation and toxicity of volatile compounds in a soil containing polychloropinene (PHP) and mineral fertilizer. Their conclusion was that the source of highly toxic volatile compounds discharged into the atmosphere comes from utilization of the ammonia water containing iron carbonyl impurities. UV-radiation breaks them down to release carbon monoxide. It is possible for carbonyl iron to react with polychloropinene forming toxic substances.

Involvement of photochemical processes in the conversions of PCP and mineral fertilizer is pointed out also by Kundiev et al. (1975) and Altareva (1975).

Noise and vibration exposure appears to modify the response of the body to the effects of pesticides.

No discussion on combined, concomitant and joint effects pesticides and other environmental factors, chemical or physical, is complete unless, besides their specific patterns of toxicity, it addresses also the long-term effects of chemicals. They are, in fact, intimately related with the concomitant action of chemical and physical environ-

mental factors. Many chemical agents, while neither carcinogens nor mutagens, can nevertheless affect the development of these effects under the influence of other substances. It is necessary, therefore, to detect and predict possible cocarcinogenic and comutagenic effects. Much depends on the ability of substances to intervene into the metabolism of carcinogens, teratogens and mutagens.

HYGIENIC REGULATION OF PESTICIDES

The maximum allowable concentrations of pesticides now effective in the Soviet Union are in most cases several times lower than those current abroad (Table 7).

Table 7

MACs for Pesticides in the Air of the Working Zone Included in USSR (Standard 12.1.005—76) and US Standards, mg/m^3

Pesticide	USSR	USA
Amidophos	0.5	5/20*
Atrazine	2	10
gamma-HCCH	0.05	0.5/1.5
Heptachlor	0.01	0.5-1.5
2,4-D	1	10/20
DDVP	0.2	1/3
DDT	0.1	1/3
Dibrom	0.5	3/6
Dieldrin	0.01	0.25/0.75
Dinitroorthocresol	0.05	0.2/0.6
Dichloropropane	10	350/525
Dichloroethane	10	200/1025
Carbophos	0.5	10
Mercaptophos	0.02	0.1/0.3
Methylmercaptophos	0.1	0.5/1.5
Methylnitrophos	0.1	0.2/0.6
Nicotine-sulphate	0.1	0.5/1.5
Pentachlorophenol	0.1	0.5/1.5
Mercury (organic compound)	0.005	0.01/0.03
Sevin	1	5/10
Carbon disulfide	1	60/90
Thiodane	0.1	0.1/0.3
Thiuram	0.5	5/10
Tributylphosphate	0.5	5
Formaldehyde	0.5	3
Chlorindane	0.01	0.5/2
Chlorophos	0.5	1/3
Free cyanamide	0.5	2

* The numerator and denominator identify the mean and maximum workshift concentrations, respectively.

The MAC specifications for some pesticides in the United States are equal to or higher than the thresholds of chronic effect, indeed higher than thresholds of acute effects for some substances. In the recent years, however, there has been a tendency to lower the MACs and bring them closer to the USSR standards (organomercury compounds, methylnitrophos, thiodane, formaldehyde, chlorophos, etc.).

Soviet legislation rules that a hygienic standard for the concentration of a substance in an environmental media must be such as to guarantee no diseases or impaired health in the short and long term and no harm to future generations.

The standard-setting procedure for pesticides goes through three basic stages:

1. Validation of calculated hygienic standards on the basis of physical and chemical constants for substances and the findings of short-term toxicological experiment.

2. Execution of a full-scale research program complete with establishment of the thresholds of chronic effect and a study of the long-term effects.

3. Hygienic investigation of pesticide concentrations in environmental media; evaluation of their effect on qualitative composition and organoleptic properties of food and water; and drawing of clinico-hygienic parallels in order to adjust the hygienic standards.

Tentative Safe Exposure Levels (TSEL) are used as official hygienic standards. They are proposed for use from the basic inputs of calculation methods (involving a short-term toxicologic experiment). In keeping with the Methodologic Instructions for the Use of the Calculation Method (1977), the TSELs for harmful substances in the air of the working zone are approved to be effective for a limited period of time (two years). It is mandatory in so doing to have available a chemical method to determine the airborne concentrations of these substances. The TSELs for pesticides are usually set at the time their Official (State) Production Tests are in progress in agriculture. With the affirmative results of the tests, the TSELs for common pesticides will further be replaced by MACs. In those cases, however, when a chemical continues to be tested or in limited use, a full-scale toxicological experiment is not practical and the TSEL may be re-endorsed for the next term. The TSEL approval system provides a tool of sanitary control over all pesticides proposed for registration for agricultural uses.

Besides the calculation-based program of hygienic standardization, MACs can be determined under an abridged program by rapid methods. This applies to new pesticides which belong to a well-known class of compounds which have hygienic standards on their content developed for one particular medium but require them to be validated for some other medium.

The complete experimental program includes detection of threshold doses and concentrations in acute experiments (as probabilistic as possible) to provide a guide to selection of the doses and concentrations for determining the thresholds of harmful effect in chronic

experiments. The latter threshold, as defined in the chronic experiment according to the limiting criterion of harm (the basic initial index) is integrated in setting hygienic standards. Its reduction several times over (the safety factor) to allow for the degree of hazard presented by the substance, differences in species sensitivity to it and other pertinent factors is essential to predicting maximum permissible safe exposure levels for man.

On occasion, other criterion than the toxicologic criterion of harm – for example, organoleptic (an altered flavour or smell of food or water) – is assumed as the critical indicator. If so, the hygienic standardization program should take these properties into account.

When a pesticide remains in use over a number of years, revision of the pertinent hygienic standard may be needed to incorporate clinico-hygienic (epidemiologic) research data. Where it is possible to identify the groups of individuals in constant or predominant contact with a given pesticide, as in packing departments, the correlation of their health indices with a pesticide concentrations detected in ambient air may provide useful insights.

Hygienic regulation of pesticides should be an integrated program covering simultaneously different environmental media. Fundamental to such an integrated hygienic regulation program is the establishment of a maximum permissible dose for man on the basis of the subthreshold dose, found in a chronic experiment by selection of suitable safety factors (Methodological Instructions for Hygienic Evaluation of New Pesticides, 1969). The next step estimates possible ratios between the pesticide quantities which enter the human body in food, water, and air and determines the MAC for the chemical so as to keep its total dose entering the body by all routes below the maximum permissible dose and preclude any adverse effects of its concentration specified for each particular environmental medium (for example, to avoid changes in the organoleptic properties and qualitative composition of the food or water concerned). Maximum permissible daily doses of pesticides for man are defined by WHO from their daily intake into the human body or per kg body weight. The threshold principle was discussed earlier (see page 10).

Although hygienic standards are set for each chemical agent on an individual basis, assessing the state of man's environment is a comprehensive venture in terms of covering the entire aggregate of factors to which man is exposed (see page 116).

Preventive sanitary supervision of pesticides' integration into agricultural practices is a most effective means of setting adequate hygienic standards for pesticides. Registration is granted exclusively to the pesticides included in the List of Chemical and Biological Means for Control of Pests, Diseases, and Weeds Recommended for Use in Agriculture. Prepared by the State Commission on Chemical Means for Control of Plant Pests, Diseases and Weeds under the USSR Ministry of Agriculture, the List is updated on an annual basis in coordination and agreement with the Ministry of Health. Pesticides shall not be included in the List which are the most hazardous and

listed as such in the Extremely Toxic Chemicals category or which have the capacity to cause severe disease states, untoward long-term effects, and so on. The List specifies the use rates of the preparation, the agricultural crops to be treated, the replication of the treatments, and the dates of the last treatment before harvesting. It incorporates an inventory of the pesticides currently in the process of experimental commercial testing. Also, the approval of the USSR Ministry of Health is sought for the list of chemicals (pesticides) identified each year for official tests. For a pesticide to be entered into the list, its complete data profile developed in the toxicologic experiment must be available together with all hygienic regulations and standards pertaining to its concentrations in all environmental media.

The list of the pesticides approved for use in personal and cooperative orchards and vegetable fields is reviewed and endorsed by the Chief Sanitary Physician of the USSR.

At the present time, standards are imposed on pesticide concentrations in the air of the working zone, ambient air, foods, livestock forages, water bodies designated for hygienic and domestic uses, and soil.

MACs for Pesticides in the Air of the Working Zone (Airborne Pesticides in the Working Environment). As defined in the National Standard 12.1.005—76, the "working zone is the space no more than two meters above the level of the floor or of the site which is the place where the workers are permanently or temporarily employed. The maximum allowable concentration of a harmful substance in the air of the working zone is the concentration that, in the case of exposure in work conditions for eight hours daily (with the exception of non-working days) or during another period, but not more than 41 hours per week throughout the entire working life, will not cause any disease or deviations from the normal state of health detectable with currently available methods of investigation, either during the work itself or in the long term, in the present and subsequent generations".

To establish MACs for the chemical agents present in the indoor air of production environments at the stage of preventive surveillance is necessary for design of production facilities, calculation of ventilation facilities, and as a means of control over the working conditions at workplaces during scheduled sanitary inspections (Addendum 1). The MACs of pesticides, approved for the air of the working zone are used also for sanitary compliance inspection of the application of toxic chemicals in agriculture. Nevertheless the MACs of pesticides used in agriculture may differ from those for the indoor working environment because of the short duration and multiplicity of exposure and the specific patterns in joint action of pesticides with physical environmental factors.

MACs for pesticides in Ambient Air. Hygienic standards for the levels of harmful substances in the ambient air are set in order to have objective criteria for the cutaway (protection) zones to be allocated around production facilities; to validate the measures designed for neutralization and dilution of toxic substances; and to evaluate the

production processes and units of plant and equipment that are sources of toxic discharges. In agriculture, the MACs for pesticides in ambient air serve likewise to set up cutaway (protection) zones (Addendum 2).

The development of hygienic standards for pesticide concentrations in ambient air has some distinctions. For one thing, such a standard must be thoroughly validated to take into account round-the-clock inhalation exposure to the substance being standardized. For this reason animal experiments too make use of the round-the-clock poisoning parameter. Secondly, while those occupationally exposed to toxic substances are healthy adult persons who undergo pre-employment and then regular medical examinations, the substances in ambient air may affect the children, the aged, and the ailing. Hence follows the necessity to impose more stringent demands on their MAC values in order to take care of age-related reactivity and the responses of sick persons, and also of the persons sensitized to a given chemical agent. In setting standards for substances in ambient air, smell is an essential consideration. Whereas the presence of a mild odour can be tolerated in defining the MAC values for the air of the working zone, none is permissible in setting ambient-air MACs. The hygienist must realize that any stimulus becomes disagreeable and intolerable if omnipresent and obtrusive (Ryazanov, 1954).

Hygienic standardization requires, along with animal experiments, that thresholds be determined for olfactory sensation and irritant action in man. The concentrations tested must be lower than the MAC values for the air of the working zone, and the reflex action is estimated by a change in the bioelectrical activity of the human brain.

With respect to ambient air, two MAC values for chemicals are recognized: maximal single and daily average. For the substances with potential for acute poisoning but without marked cumulative properties the two values are often identical. For the cumulative substances, on the other hand, their maximal single concentrations should be several times their daily averages.

MACs for Pesticides in Water. Standards for harmful substances in water (termed in the USSR "the bodies of water" (are derived in regard of three critical indicators of harm – sanitary-toxicologic, organoleptic, and the effect on the general sanitary condition of a water body. Of these, the organoleptic parameter proves a major constraint and a critical index in the majority of cases and therefore 35 per cent of the pesticides have standards in water established according to the organoleptic criterion.

The MACs of pesticide concentrations in water bodies are applied not merely in validating permissible ceilings on the effluents from pesticide production facilities, designing systems of treatment facilities and neutralization of effluents from production units, but also in order to provide adequate cutaway zones of water bodies from pesticide-treated croplands, as well as to assess the proposed systems for pesticide treatment of farm crops and a host of other uses (Adden-

dum 3. "Maximum Allowable Concentrations of Harmful Substances in Evironmental Media", 1977).

MACs for Pesticides in Food. This hygienic standard has been termed in the USSR "the permissible residual amount (PRA) of a pesticide in foodstuffs". A WHO Conference decided to set similar standards in terms of "maximum permissible levels" (MPL) of toxic chemicals.

Pesticide standardization in foods proceeds in agreement with the Methodological Instructions for Hygienic Evaluation of New Pesticides (1969). It requires a threshold dose to be determined from a chronic experiment in laboratory animals so as to arrive at the maximal safe dose for man (Dm). The factors to consider in so doing include the magnitude of cumulative properties, the resistance of the pesticide concerned to environmental factors, its ability to be excreted in milk and adversely affect the progeny, and data on comparative animal and human sensitivity to the substance. For the agents whose Dm represent decimal fractions of one mg/kg and which show high persistence in the environment and marked cumulative properties, the recommended safety factor is 100. With Dm in the 1 to 5 mg/kg range, the safety factor is selected to be within 50 to 100. Finally, for the chemicals with moderate persistence and low cumulative potential and with the Dm value for animals above 5 mg/kg, the recommended safety factor should fall within 30 to 50. For man, the Dm is calculated by the formula:

$$\text{Dm}^*(\text{man}) = \frac{\text{Dm (animal) mg/kg} \cdot 50 \text{ (body weight in kg)}}{\text{Safety factor}}.$$

After that, according to Spynu's recommendation on integrated standardization for concomitant effects, the Dm is divided by the three media. From our point of view, this standardization procedure fails to consider properly variations in the ratio of the pesticide amounts entering the body in diverse media under its different applications (Addendum 4).

MACs for Pesticides in Livestock Forages. The primary source of pesticides in the foods of animal origin derives from livestock forages and feeds. As a result, strict standards have to be imposed to control their concentrations in the livestock forages if toxic chemical residues in dairy and meat are to be minimized (Addendum 5. The List of Chemical and Physical Means for Control of Pests, Plant Diseases and Weeds Registered for Use in Agriculture, 1979).

MACs for Pesticides in Soil. The practical need to devise standards for harmful substances in soil was recognized by the 16th National Hygienists Convention in 1972; it has been pursued in the recent years with new vigor and emphasis by a number of authors (Spynu, 1972; Goncharuk et al., 1974; Perelygin, 1975; Sidorenko et al., 1978).

The standardization procedure to take care of pesticides in soil is based on close consideration of their transition pathways in the biological chains: soil — plants — man and soil — ambient air — man.

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* Permissible daily intake for man.

The idea is to prevent their build-up, in the media contacting soil in concentrations in excess of the maximum allowable ones, but do so in a way non-damaging to the self-purifying, barrier, and other functions of soil. The duration of a pesticide's persistence and metabolism in a soil appears to be strongly influenced by its microbioceonosis, pH, temperature, and moisture content. The method used to set MACs for pesticides in soil is experimental modeling (Goncharuk et al., 1974; Addendum 6).

Specific Hygienic Standardization Procedures for Pesticides of Biological Origin. Pesticides of biological origin are classified into antibiotics and microbial preparations. The former comprise essentially the same chemical constituents as do most other pesticides so that the same methodological approaches are applied to setting hygienic standards for them. In addition to determining the thresholds of systemic toxic effect one must be sure to identify the minimum doses and concentrations exerting sensitizing, allergic and antibacterial action (the applicable Soviet term is "intestinal dysbacteriosis") since they may and often do turn out to be the critical indicator in the establishment of the MACs.

The strains pathogenic to man are prohibited for agricultural uses. For them, pathogenicity should be assessed in special experiments with intragastric administration to animals of the chemicals being investigated. Microorganisms may be potentially pathogenic, due to their ability to become pathogenic on exposure to environmental factors. To evaluate the likely effects of these preparations on the human population it is required a thorough-going investigation into health status among the persons occupationally exposed to them.

Calculation and rapid methods for hygienic standardization of pesticide concentrations in environmental media. A series of equations have been proposed to calculate toxicities and MACs for volatile and nonvolatile organic compounds. They provide guidance estimates as to lethal and threshold concentration levels of substances and help calculate hygienic specifications for their physico-chemical properties such as molecular mass, density, refractive index, melting and boiling points, etc. TSELs can be calculated for various toxicity indices (CL_{50}, DL_{50}, threshold concentrations) derived in a short-term experiment. The toxicity and TSEL estimation for inorganic compounds presents a more difficult problem as sufficiently close correlations between their structure and toxic action have not been found as yet. In a similar manner, attempts are being made to trace a correlation between toxicity and the electron structure of the atoms in the crystal lattice of a substance, as well as its molecular mass, boiling point and some physico-chemical parameters (Filov, Liublina, 1976).

The rapid and calculation methods of hygienic standardization can be divided into:

1. Methods used to validate calculated standards on the basis of physico-chemical characteristics of substances.
2. Methods used to validate TSELs of substances from the inputs of a short-term toxicologic experiment.

3. Methods used for rapid setting of calculated standards for substances in a particular environmental medium when the standards for their concentrations in another medium are already available.

Most of the methods proposed to date are rested on the common general principle of identifying correlations between the established hygienic standard values and various physico-chemical and biological parameters. Not infrequently, biological parameters are predicted from physico-chemical parameters and complex indices from more simple ones found in experiment. The predictive exercise achieves more success when undertaken within homologous series. Some regression equations are based on correlations between the standards set for different environmental media.

Validation of Calculated Standards on the Basis of Physico-Chemical Characteristics of Substances. The information a hygienic toxicologist gets first from a chemist about a new substance concerns its structure, and physical and chemical properties. Already at that stage and even prior to the start of a biological experiment, there is some possibility to predict in a strictly tentative manner toxic properties and MAC values. Liublina and Golubev (1973) employed correlations to derive a series of regression equations relating MACs in the air of the working zone to molecular mass, boiling and melting points, density, and refractive index. Reliance on the regular patterns derived for homologous series or the compounds similar in chemical structure enhances the accuracy of such calculations. The use of Hammet's correlation equations for the substituents in the benzene ring; Taft's equations for the derivatives of aliphatic compounds; Zabradnik's equations to tie the biological activity of the i-th homologue to that of the ethyl derivative; and Hansch's equations incorporating the effect of steric factors on biological activity, may benefit the prediction of a substance's toxicity and biological activity from data concerning their structure within related compounds. Zaeva (1964, 1970) proposed a MAC calculation method based on assumed additivity of the contributions by chemical radicals, atoms and bonds to the magnitude of biological activity:

$$MAC = \frac{M \cdot 1000}{\Sigma I_i} \ (mg/m^3), \text{ where}$$

M is molecular weight, and

ΣI_i is the sum of biological activities.

An attempt has been made to apply a quantum-chemical approach to predicting toxicity and setting hygienic standards of organophosphorus pesticides (Kagan et al., 1976). It involved computer-aided estimation of quantum-chemical constants for the value of positive charge on the phosphorus atom (Λ^+) and the rupture energy of the ester bond. Using the Del Rae method to obtain data on the electron structure of the molecule, a multiple correlation was identified between quantum- and physico-chemical constants, on the one hand, and the toxic properties of pesticides and their hygienic specifications, on another. The regression equations were checked for their efficacy

through the estimation of CL_{50} and MAC in the air of the working zone for a number of new organophosphorus compounds.

However, the calculated hygienic standards based on physico-chemical characteristics alone are primarily in the nature of tentative guides and therefore cannot as yet be approved as official standards. They can nevertheless be efficiently applied for the design and development of new compounds in the laboratory, as well as in the toxicological experiment.

TSEL Validation from Data of Short-Term Toxicological Experiment. Filov and Liublina (1976) argue that the estimates of substances' toxicities and MACs based on the characteristics of their biological activity (experimental data) are more accurate than those derived from their physico-chemical properties.

Golubev offered several formulas to calculate CL_{50} values from DL_{50}, threshold concentrations from DL_{50} and CL_{50}, and TSEL in the air of the working zone from DL_{50}, CL_{50}, and threshold concentrations. The formulas are listed in the book (1976) titled "Fundamentals of General Industrial Toxicology" (Osnovi obshchei promyshlennoi toksikologii).

Kagan, Sasinovich and Ovseenko (1971, 1972) developed on computer paired and multiple regression equations to permit TSEL calculation for the air of the working zone using for the purpose pesticide toxicity and cumulation indices. The formulas were then put to advantage to estimate the TSELs in the air of the working zone for all of the pesticides currently used in the USSR (Kagan, Sasinovich, 1973). Likewise, paired and multiple correlations between the permissible residual doses (PRD) of pesticides in food and their toxicity indices were established and regression equations constructed on that basis. The latter can serve to arrive at tentative PRD estimates for new pesticides in foods (Kagan et al., 1972). Using the formulas, Antonovich (1976) defined PRDs for 26 new toxic chemicals.

In the drive towards specialization and unification of methodological approaches to design of a toxicologic experiment, new information has been accumulated over the recent years, sufficient to examine the correlations of the MACs with the toxicity parameters of substances and their physical properties not only for the pesticide category in its entirety, but also for the latter's more common chemical classes — organophosphates, organochlorines, and derivatives of carbamic and thio- and dithiocarbamic acids.

They provided new inputs for updating the formulas and nomograms (Figs. 17–21) used to identify the pesticide TSELs in the air of the working zone (Kagan, Sasinovich, Ovseenko, 1976). The highest accuracy of the calculated standards is achieved by determining the MAC multifactorial dependence on the degree of toxicity by the oral and dermal routes, along with the integration of their cumulative properties.

The nomograms thus obtained save the need to resort to elaborate calculations in the tentative estimation of TSEL values for the pesticides present in the air of the working zone.

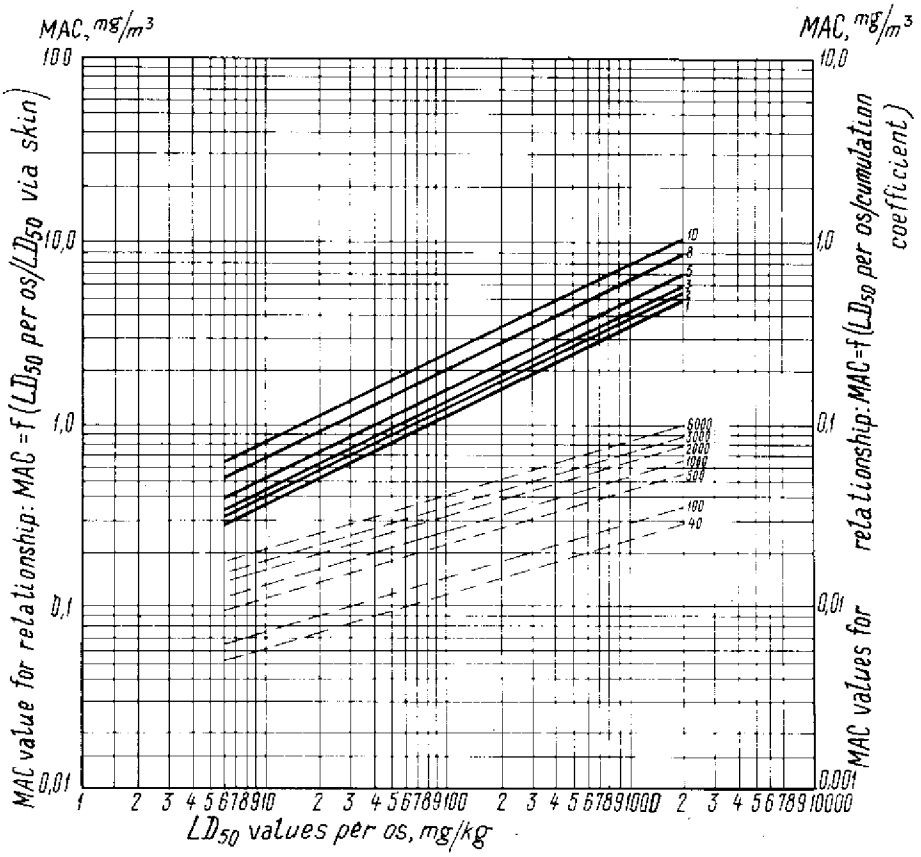

Fig. 17. Nomogram to define tentative safe exposure levels (TSELs) for OPP, using two parameters (Kagan, Sasinovich, Ovseenko):
LD_{50} via skin, ———— cumulation coefficient

An example from the regression equations for the organophosphates is given below:

$$\ln y = 0.517 \ln x_1 \quad 3.928 \qquad (1)$$

$$\ln y = 0.299 \ln x_1 + 0.255 \ln x_2 - 4.458 \qquad (2)$$

$$\ln y = 0.483 \ln x_1 + 0.086 \ln x_3 - 4.347 \qquad (3)$$

$$\ln y = 0.284 \ln x_1 + 0.237 \ln x_2 + 0.08 \ln x_3 - 4.811 \qquad (4)$$

where y — TSEL, x_1 LD_{50} per os, x_2 — LD_{50} via skin, and x_3 — the cumulation coefficient. The same principle was applied in deriving the similar equations for the organochlorines, derivatives of carbamic

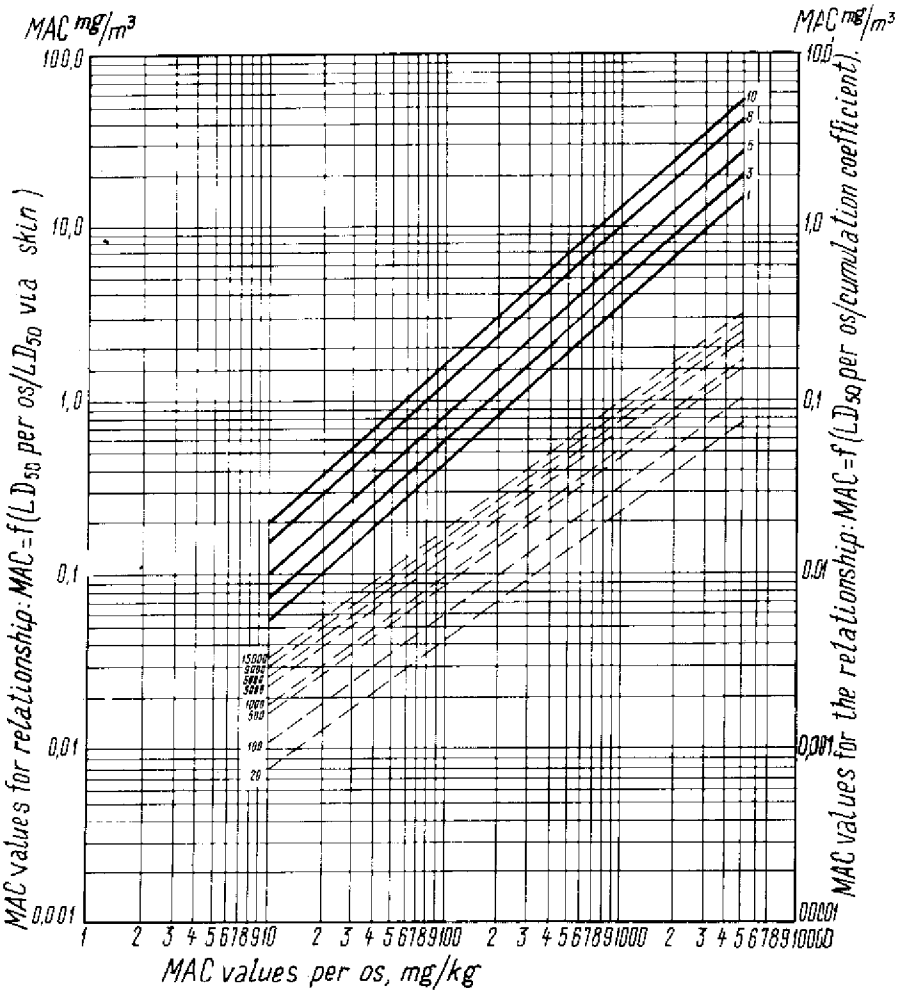

Fig. 18. Nomogram to define OCP TSELs, using two parameters (Kagan, Sasinovich, Ovseenko):

LD_{50} via skin, ——— cumulation coefficient

and thio- and dithiocarbamic acids, and several other pesticide categories. Also, nomograms have been constructed for all variants. Differentiated TSEL calculation with respect to individual pesticide classes has increased their consistency with the relevant hygienic standards developed under the complete validation procedure.

For some pesticides, the critical indicator of their effects on the organism considers primarily their irritant properties and not systemic toxic effect. Where this is the case, one can apply the irritant action threshold to estimate the TSEL values (Maximov, 1969; Golubev et al., 1973).

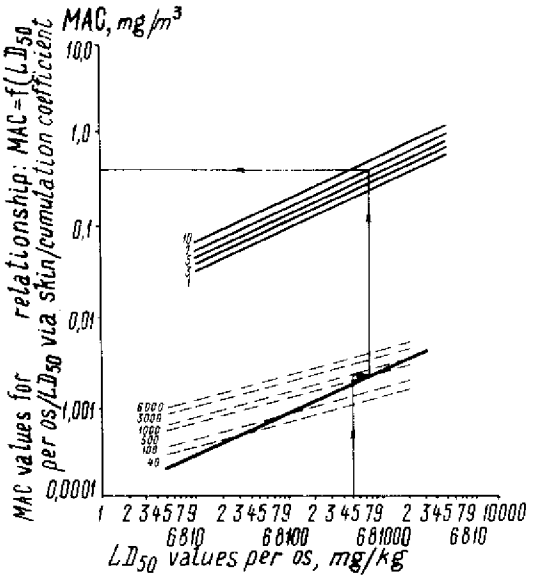

Fig. 19. Nomogram to define OPP TSELs, using three parameters (Kagan, Sasinovich, Ovseenko): — — LD_{50} via skin, ——— cumulation coefficient, ——— auxiliary line

Krotov (1971) examined closely the correlation between olfactory sensation thresholds, photosensitivity of the eye, bioelectrical activity of the brain cortex and maximal single (one-time) ambient-air MACs and deduced several regression equations.

Andreeshcheva (1971, 1976) has found the correlation between the accepted maximal single ambient-air MACs and olfactory sensation thresholds for 101 chemicals. The maximal single MACs fell below the olfactory sensation threshold by a factor of 3.8 on the average, and by a factor of 5 when the triple error of the coefficient was considered. On that basis, the author proposed a rapid method to set maximal single MACs for airborne substances, using their olfactory threshold with the safety factor of 5. Because for most pesticides the threshold concentration according to systemic toxic action is above their olfactory threshold and because two thirds of the 700 substances now standardized for the air of the working zone provenly possess that threshold, the author thinks it quite reasonable to employ a method for rapid standardization of the slightly toxic substances (hazard classes 3 and 4) centered on the criterion of smell.

Zaugolnikov et al. (1975) derived several equations to determine by calculation hygienic standards for pesticides in ambient air and water (with the sanitary toxicological index as the critical indicator) according to their acute toxicity indices and some physico-chemical properties. The authors additionally plotted nomograms to establish MACs for the air of the working zone, ambient air and water bodies from the CL_{50} and DL_{50} of respective pesticides.

Setting Rapid and Calculated Standards for Substances in a Particular Environmental Medium from Available Standards for Their

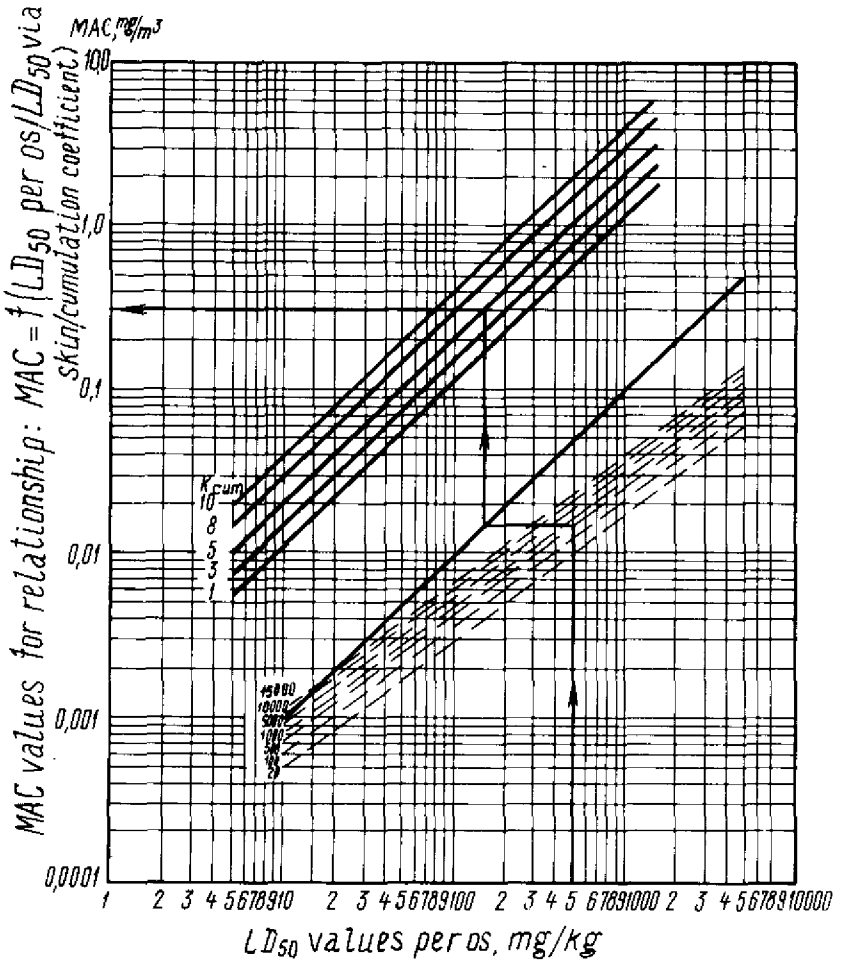

Fig. 20. Nomogram to define OCP TSELs using three parameters (Kagan, Sasinovich, Ovseenko):

- LD_{50} via skin, ——— cumulation coefficient, ——— auxiliary line

Concentrations in Another Medium. The relation between the MAC of a chemical in the air of industrial areas and its ambient-air MAC was determined by Tolokontsev (1967), yet the correlation coefficient has not been significant (+0.49). Spynu and Ivanova (1969) obtained a significant correlation coefficient ($p=0.001$) between the pesticide MAC in the air of the working zone and their maximal single (+0.63) and mean daily (+0.69) ambient-air concentrations. They came up with a set of regression equations relating the pesticide ambient-air MACs with their MAC in the air of the working zone, and the daily average with maximal single ambient-air MACs:

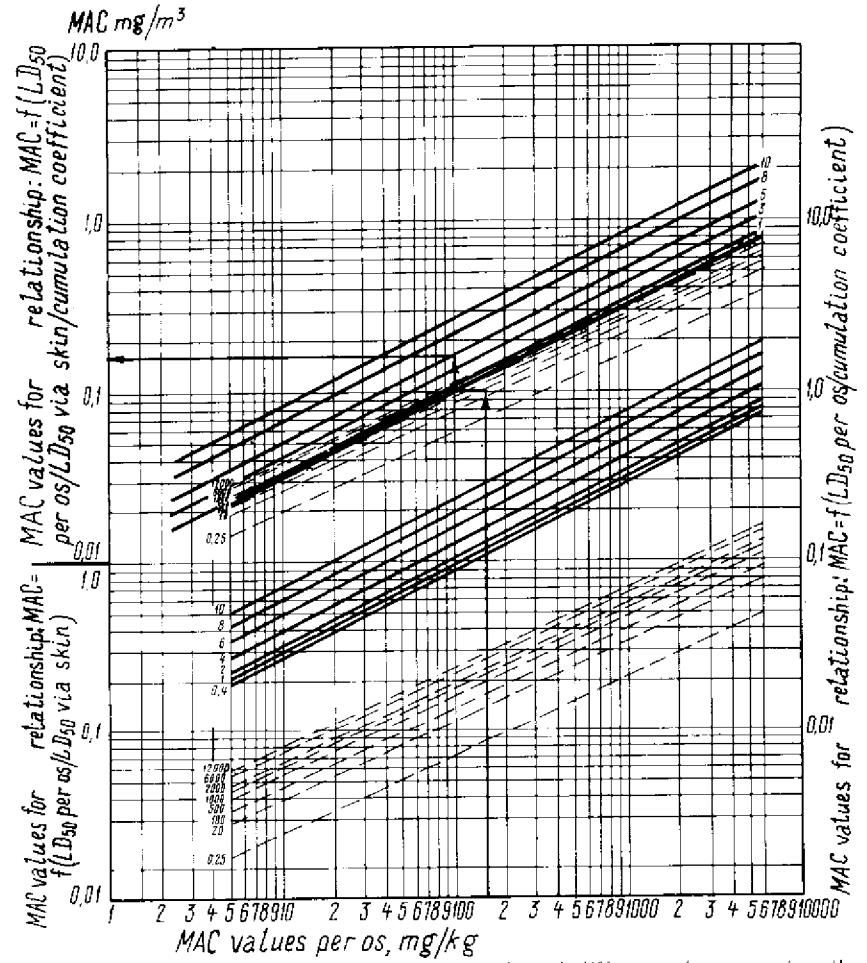

Fig. 21. Nomogram to define TSELs for pesticides of different classes, using three parameters (Kagan, Sasinovich, Ovseenko):
LD_{50} via skin, ——— cumulation coefficient, ——— auxiliary line

$$\lg y_{m.d.} = (-2.16 \pm 0.28) + (0.88 \pm 0.16) \lg x \quad (r = +0.69),$$
$$\lg y_{m.s.} = (-1.777 \pm 0.3) + (0.55 \pm 0.09) \lg x \quad (r = +0.63),$$
$$\lg y_{m.d.} = (-0.15 \pm 0.03) + (1.02 \pm 0.2) \lg y_{m.s.} \quad (r = +0.94),$$

where $y_{m.d.}$ is the mean daily MAC in ambient air,
$y_{m.s.}$ is the maximal single MAC in ambient air, and
x — is the MAC in the air of the working zone.

The somewhat lower correlation of the maximal single MACs of chemicals in ambient air than of the daily averages with their MACs in the air of the working zone can be accounted for by the way they have been obtained: whereas the former were established primarily on the basis of reflex action indices, the mean daily MACs as also the

MACs in the air of the working zone are most typically rested on the threshold concentration value for the skin resorptive effect. In the analysis of the causes for this disparity, one must bear in mind that some ambient air MACs have been defined by the organoleptic criterion.

A series of regression equations to estimate ambient-air MACs of substances from their respective MACs in the air of the working zone was proposed by Zaugolnikov et al (1975):

$$\lg \text{MAC}_{\text{m.d.}}\ (\text{mg/m}^3) = -2.0+0.86 \lg \text{MAC}_{\text{w.z.}}\ (\text{mg/m}^3) \quad (r=+0.65),$$

$$\lg \text{MAC}_{\text{m.s.}}\ (\text{mg/m}^3) = -1.78+\lg \text{MAC}_{\text{w.z.}}\ (\text{mg/m}^3) \quad (r=+0.65),$$

$$\lg \text{MAC}_{\text{m.d.}}\ (\text{mg/m}^3) = 0.47+0.86 \lg \text{MAC}_{\text{m.s.}}\ (\text{mg/m}^3) \quad (r=+0.88).$$

Golubev and Subbotin (1968), having performed a correlation analysis on the data secured from a survey of 37 substances, suggested the following equation for the estimation of MACs in bodies of water:

$$\lg \text{MAC}\ (\text{mg/l}) = 0.61 \lg \text{MAC}_{\text{w.z.}} - 0.1\ (\text{mg/m}^3).$$

The equation is applicable to setting tentative standards for water concentrations of those substances whose critical indicator is the toxicological principle of harmfulness.

ADDENDA

Addendum 1

MACs of Pesticides in the Air of the Working Zone (USSR National Standard 12.1.005—76)

Trade name	Chemical name	Concentr. mg/m³	State of aggreg.	Class of hazard
Abate *	0,0,0,0-tetramethyl-0,0-thio-di-p-phenylenethio-phosphate	0.5	v+a	2
Acylate	Acetoxyisopropylphenylcarbamate	2.0	v+a	3
Afugan *	0,0-diethylthiophosphoryl-5 methyl-6-carboethoxypyrazolylpyridyl	0.05	v+a	1
Aldrin *	Hexachlorohexahydro-dimethanonaphthalene	0.01	v+a	1
Amidophos *	0-(4-terthutyl-2-chlorophenyl)-0-methyl-N-methyl-amidophosphate	0.5	v+a	2
Amiphos *	0,0-dimethyl-S-2 (acetylamino)-ethyldithiophosphate	0.5	v+a	2
Anilate	Monoethanolamine salt of sulfanilic acid	1.0	a	3
Anthio *	0.0-dimethyl-3-(N-methyl-N-formylcarbamoylmethyl)-dithiophosphate	0.5	v+a	2
Arsenite chemicals*	by As	0.3	a	2
Atrazine	2-chloro-4-ethylamino-6-isopropylamino-sym-triazine	2.0	a	3
Basudine *	0,0-diethyl-0-(2-isopropyl-4-methyl-6-pyrimidyl) thiophosphate	0.2	v+a	2
Baytex *	0,0-dimethyl 0-(4-methylmercapto-3-methylphenyl) thiophosphate	0.3	v+a	2
Betanal	3-methoxycarbonyl-aminophenyl-N-(3-methylphenyl) carbamate	0.5	a	2
Boverine	Fungal drug	0.3	a	2
Bromophos *	0,0-dimethyl-0-(2,5-dichloro-4-bromphenyl) thiophosphate	0.5	v+a	2
Butylcaptax	2-butylthiobenzthiozene	2.0	v	3

Addendum 1 (contd.)

Trade name	Chemical name	Concentr. mg/m³	State of aggreg.	Class of hazard
Carbathione	Sodium N-methyldithiocarbamate	0.1 (by methyl-isothiocyanate)	a	1
Carbin	4-chlorobutin-2-yl-N-(3-chlorophenyl) carbamate	0.5	a	2
Carbon disulfide *		1	v	2
Carbophos *	0,0-dimethyl-S-(1,2-biscarbethoxyethyl) dithiophosphate	0.5	v+a	2
Carpene (melprex)	N-dodecylguanidineacetate	0.1	a	2
Carbon tetrachloride *		20.0	v	2
Chloramp	3,5,6-trichloro-4-aminopicolinic acid	10.0	a	3
Chlorindan *	Octachloroendomethylentetrahydroindan	0.01	v+a	1
Chlor IPC	Isopropyl-N-(3-chlorophenyl) carbamate	2.0	a	3
Chlorophos *	0,0-dimethyl-(1-oxy-2,2,2-trichloroethyl phosphonate)	0.5	v+a	2
Cidial *	Ethyl ether of 0,0-dimethyl dithiophosphoryl-l-phenylacetic acid	0.15	v+a	2
Copper vitriol	Copper sulfide	0.3	a	2
Cotoran	3-trifluoromethylphenyl-N, N-dimethylurea	5	a	3
Cuprosan	Zinc ethylene-N,N-bis-dithiocarbamate and copper chloroxide	0.5	a	2
Cuprocine	Mixture: zinc and copper ethylene-bis-dithiocarbamates	0.5	a	2
Cyanamide (free)		0.5	v+a	2
Cyanox *	0,0-dimethyl-0-4-cyanophenyl-thiophosphate	0.3	v+a	2
2,4-D	2,4-dichlorophenoxyacetic acid	1.0	a	2
2,4-D amide	Amide 2,4-dichlorophenoxyacetate	1.0	a	2
2,4-D, butyl ether	Butyl ether of 2,4-dichlorophenoxyacetic acid Chloro	0.5	v+a	2

Trade name	Chemical name	Concentr. mg/m³	State of aggreg.	Class of hazard
2,4-D, crotyl ether	Crotyl ether of 2,4-dichlorophenoxyacetic acid	1.0	v+a	2
2,4-D, sodium salt	Sodium 2,4-dichlorophenoxyacetate	1.0	a	2
2,4-D, octyl ether	Octyl ether of 2,4-dichlorophenoxyacetic acid	1.0	v+a	2
Dalapon	Sodium α,α-dichloropropionate	5.0	v+a	3
DDB	Mixture: 1,2-dichloroisobutane, 1,3-dichloroisobutylene, 3,3-dichloroisobutylene	0.3 (by dichloroisobutylene)	v	2
DDVP *	0,0-dimethyl-0.2.2-dichlorovinylphosphate	0.2	v	2
DDT	4,4-dichlorodiphenyltrichloroethane	0.1	v+a	1
Dianate	2-methoxy-3,6-dichlorobenzoic acid	0.1	a	2
Dibrom *	0,0-dimethyl-0(1,4-dibromo-2,2-dichloroethyl)-phosphate	0.5	v	2
Dicotex	2-methyl-4-chlorophenoxyacetic acid	1.0	a	2
Dicresyl	Dicresyl ether of methylcarbamic acid	0.5	v+a	2
Dieldrin *	1,2,3,4,10,10-hexachlor-6,7-epoxy-1,4.5,8-diendomethylene-1,4,4a,5,6,7,8,8a-octahydronaphthalene	0.01	v+a	1
Dinitroorthocresol *	2,4-dinitro-6-methylphenol	0.05	v+a	1
Dinitrorodanbenzene *	2,4-dinitrophenylthiocyanate	2	a	3
Dichloronaphthoquinone (Phygon)	2,3-dichloro-1,4-naphthoquinone	0.5	a	2
Dichloropropyonic acid	α,α — dichloropropyonic acid	1.0	a	2
Dichloroethane *	1,2 dichloroethane	10.0	v	3
Entobacterine	Bacterial drug	3.0	a	3
Eptam	S-ethyl-N, N-dipropylthiocarbamate	2	v+a	3
EMP *	Ethylmercury phosphate	0.005	v	1
EMC *	Ethylmercury chloride	0.005	v	1

Trade name	Chemical name	Concentr. mg/m³	State of aggreg.	Class of hazard
Euparen	N,N-dimethyl-N-phenyl-N-fluorodichloromethylthio-sulfamide	1.0	v+a	3
Ester sulfonate	p-chlorophenyl-p-chlorobenzenesulfonate	2	v+a	3
Gamma HCCH *	Gamma-isomer of hexachlorocyclohexane	0.05	v+a	1
Gardona *	Trans-isomer-2-chloro-1 (2, 4,5-trichlorophenyl) - vinyldimethylphosphate	1.0	a	2
Gliftor	70% 1,3-difluoropropanol-2+30% 1-fluoro-3-chloropropanol	0.05	v	1
Granosan (by Hg) *	Active ingredient ethylmercury chloride	0.005	v+a	1
Hexachloran *	1,2,3,4,5,6-hexachlorocyclohexane	0.1	v+a	2
Hexachlorobenzene *	Hexachlorobenzene-(1,2,3, 4,5,6)	0.9	v+a	2
Hexachlorobutadiene *	Hexachlorobutadiene-1,3	0.01	v+a	1
Heptachlor *	1,4,5,6,7,8,8-heptachloro-4, 7-endomethylene-3a,7,7a-tetrahydroinden	0.01	v	1
Herban	N-(tetrahydrodicyclopentadienyl)-N',N'-dimethyl-urea	5.0	a	3
Hydrocyanic acid *	Hydrogen cyanide	0.3	v	2
Hydrochloric acid	Hydrogen chloride	1	v	2
Iodofenphos *	0,0-dimethyl-0-(2,5-dichlor-4-iodophenyl) thiophosphate	0.5	v+a	2
IPC	Isopropyl-N-phenyl-carbamate	2.0	v+a	3
Karathane	2,4-dinitro-6-(2-octyl)-0,2-phenyl-crotonate	0.2	a	2
Kilval *	0,0-dimethyl-S-2-(1-methyl-carbomoylethylthio) ethyl-thiophosphate	0.1 *	v+a	2
Linuron	N-(3,4-dichlorophenyl)-1-N-methyl-N-methoxy-urea	1	a	2
Melprex	Dodecylgranidinacetate	0.1	a	2

Addendum 1 (contd.)

Trade name	Chemical name	Concentr. mg/m³	State of aggreg.	Class of hazard
Mercuran * (by Hg)	Mixture of ethylmercury-chloride and gamma-isomer of hexachlorcyclohexane	0.005	v+a	1
Metaphos *	0,0-dimethyl-0-(4-nitrophenyl) thiophosphate	0.1	v+a	1
Methylacetophos *	0,0-dimethyl-S-(carbaethoxymethyl) thiophosphate	1	v+a	2
Methyl bromide	Methyl bromide	1,0	v	2
Methylmercaptophos *	Mixture: 0,0-dimethyl-0-(2-ethylthioethyl)-thiophosphate and 0,0-dimethyl-S-(2-ethylthioethyl)-thiophosphate	0.1	v+a	1
Methylethylthiophos *	0-methyl-0-ethylnitrophenylthiophosphate	0.03	v+a	1
Monuron	N-(4-chlorophenyl)-N',N'-dimethylurea	2	a	3
M-81 *	0,0-dimethyl-S-ethylmercaptoethyldithiophosphate	0.1	v+a	1
Octamethyl *	Octamethyltetramide pyrophosphate	0.02	v+a	1
Pentachloronitrobenzene *	Pentachloronitrobenzene	0.5	v+a	2
Pentachlorophenol *	Pentachlorophenol	0.1	v+a	1
Phenazon	1-phenyl-4-amino-5-chloropyridazon	0.5	a	2
Phosalon *	0,0-diethyl-S-(6-chlorobenzoxazoniline-3-methyl) dithiophosphate	0.5	v	2
Phosphamide *	0,0-dimethyl-S-(N-methylcarbamidomethyl) dithiophosphate	0.5	v+a	2
Phthalophos *	0,0-dimethyl-S-(phthalimidomethyl) dithiophosphate	0.3	v+a	2
Phytobacteriomycin	Streptotricin antibiotic	0.002	a	1
Polycarbacin	Zinc polyethylenthiuramdisulfide	0.1	v+a	2
Polymarcin	Mixture: zinc ethylene-bis-dithiocarbamate, ethylenthiuram disulfide and manganese ethylene-bis-dithiocarbamate	0.5	a	2

Addendum 1 (contd.)

Trade name	Chemical name	Concentr mg/m³	State of aggreg.	Class of hazard
Polychlorocamphene *	Mixture of chlorinated camphenes	0.2	v+a	2
Polychloropinene *	Complex mixture of chlorinated bicyclic compounds	0.2	v+a	2
Prometrin	2-methylthio-4,6-bis-(isopropylamino)-sym-triazine	5	a	3
Propanide	3,4-dichloropropionanilide	0.1	a	1
Propasin	2-chloro-2,6-bis-isopropylamino-sym-triazine	5	a	3
Ramrod	2-chloro-N-isopropylacetanilide	0.5	a	2
Ronit	S-ethylcyclohexylthiocarbamate	1	v+a	2
Sayphos *	0,0-dimethyl-S-(4,6-diamino-1,3,5-triazin-2-yl-methyl) dithiophosphate	1 *	v+a	2
Sevin	1-naphthyl-N-methylcarbamate	1	a	2
Simazine	2-chloro-4,6-bis-ethylamino-symtriazine	2	a	3
Solan	N-(3-chloro-4-methylphenyl)-2-methyl pentanamide	1.0	a	2
Sodium trichloroacetate	Sodium trichloroacetate	5.0 *	a	3
Tillam	S-propyl-N-ethyl-n-butylthiocarbamate	1	v+a	2
Thiodan *	1,2,3,4,7,7-hexachlorobicyclo-(2,2,1)-heptene-5,6-bis-oxymethylensulfite	0.1	v+a	1
Thiophos *	0,0-diethyl-0-(4-nitrophenyl) thiophosphate	0.05	a	1
TMTD	Tetramethyl thiuram-disulfide	0.5	a	2
Trichloro-metaphos-3 *	0-methyl-0-ethyl-0-trichlorophenylthiophosphate	0.3	v+a	2
Trolen *	0,0-dimethyl-0-(2,4,5-trichlorophenyl) thiophosphate	0.3	v+a	2
Valexon *	0,0-diethyl-thiophosphoryl-α-oxymino-phenylnitryl acetate	0.1	v+a	1
Vitavax	2,3-dihydro-6-methyl-5-phenyl-carbamoyl-1,4-oxathiin	1.0	a	2

Trade name	Chemical name	Concentr mg/m³	State of aggreg.	Class of hazard
Yalan	S-ethyl-N-hexamethylent-hiocarbamate	0.5	v+a	2
Zineb	Zinc ethylene-N, N-bis-dithiocarbamate	0.5	a	2

* Hazardous by entry through skin.

Addendum 2

MACs of Pesticides in Ambient Air, mg/m³

Pesticide	Maximal single concentr.	Mean daily concentr.
Anthio	0.02 *	0.006 *
Arsenites	By arsenic	0.003
Atrazine	0.02 *	0.02 *
Basudine	0.01	0.01
Carbathione	0.005 *	0.001 *
Carbin	0.01 *	0.006 *
Carbophos	0.015	0.006 *
Chlor IPC	0.02 *	0.02 *
Chlorophos	0.04	0.02
Cidial	0.006 *	0.002 *
Copper vitriol	0.009 *	0.004 *
Cyanamide free	0.01 *	0.006 *
Cuprosan	0.01 *	0.006 *
2,4-D amide	0.02 *	0.01
2,4-D butyl ether	0.01 *	0.006 *
DDB	0.009 *	0.004 *
DDVP	0.007 *	0.002 *
DDT	0.005 *	0.001 *
Dinitroorthocresol	0.003 *	0.0008 *
Dichloronaphthoquinone	0.05	0.05
Dichloroethane	3.0	1.0
Eptam	0.02 *	0.02 *
Ethylmercury chloride (by Hg)	0.0009 *	0.0001 *
Ether sulfonate	0.02 *	0.02 *

Addendum 2 (contd.)

Pesticide	Maximal single concentr.	Mean daily concentr.
Gardona	0.007	—
Granosan (by Hg)	0.0009 *	0.0001 *
Hexachloran	0.03	0.03
Hexachlorobutadiene	0.001 *	0.0002 *
Heptachlor	0.001	0.0002
IPC	0.02 *	0.02 *
Kilval	0.005 *	0.001 *
Mercuran (by Hg)	0.0009 *	0.0001 *
Metaphos	0.008 *	0.001 *
Methyl bromide	0.02 *	0.01 *
Methylmercaptophos	0.005	0.001 *
Monuron	0.02 *	0.02 *
M-81	0.001	0.001
Nitraphen	0.02*	0.01*
Octamethyl	0.002*	0.0004*
Pentachloronitrobenzene	0.01 *	0.006 *
Pentachlorophenol	0.005 *	0.001 *
Phosalon	0.01	0.01
Phosphamide	0.003	0.003
Phthalophos	0.009 *	0.004 *
Polycarbacin	0.002 *	0.0005 *
Polychloropinene	0.005	0.005
Prometrin	0.04 *	0.04 *
Propasin	0.04 *	0.04 *
Propanide	0.005 *	0.001 *
Sevin	0.02 *	0.02 *
Simazine	0.02*	0.02*
Tillam	0.02*	0.01*
Thiodan	0.005*	0.001*
TMTD	0.01*	0.006*
Trichlorometaphos-3	0.009*	0.004*
Yalan	0.01*	0.006*
Zineb	0.01*	0.006*

* Calculated values (Maximum Allowable Concentrations of Harmful Substances in Environmental Media, 1977).

MACs of Pesticides in Bodies of Water for Domestic, Public, and Recreational Uses ("Sanitary-Domestic")

Pesticide	MAC, mg/l	Limiting criteria
Acrex	0.2	Sanitary-toxicologic
Aldrin	0.002	Organoleptic
Anthio	0.004	General sanitary
Arsenites	0.05	Sanitary-toxicologic
Atrazine	0.5	General sanitary
Basudin	0.3	Sanitary-toxicologic
Baytex (sulfidophos)	0.001	Organoleptic
BMC	0.1	"
Captan	2.0	"
Carbathione	0.02	"
Carbin	0.03	"
Carbon disulfide	1	"
Carbophos	0.05	"
Carpene	0.005	"
Chlorophos	0.05	"
Copper vitriol	0.1	"
2,4-D, amide	0.2	Sanitary-toxicologic
2,4-D, butyl ether	0.5	"
2,4-DM	0.01	"
2M-4CD	2.0	"
2,4-D, octyl ether	0.2	Organoleptic
Dalapon	2.0	"
DD	0.4	Sanitary-toxicologic
DDB	0.4	"
DDVP	1.0	Organoleptic
DDT	0.1	Sanitary-toxicologic
Dicotex	0.25	Organoleptic
Dilor	0.1	"
Dinitroorthocresol	0.05	Toxicologic
Dinitrorhodan benzene	0.5	General sanitary
Diuron	1.0	Organoleptic
Diphenamide	1.2	Sanitary-toxicologic
Dichloronaphthoquinone	0.25	"
Dichloroethane	2.0	Organoleptic
EF-2	0.01	General sanitary
Eptam	0.1	Organoleptic
Gardona	0.3	General sanitary
Gliftor	0.006	Sanitary-toxicologic
Granosan (by Hg)	0.001	"
Hexachloran	0.02	Organoleptic
Hexachlorobenzene	0.05	Sanitary-toxicologic
Hexachlorobutadiene	0.01	Organoleptic

Addendum 3 (contd.)

Pesticide	MAC, mg/l	Limiting criteria
Hexilur	0.2	Sanitary-toxicologic
Heptachlor	0.05	"
Herban	2.0	"
Isophos	0.4	"
IPC	0.2	Organoleptic
Kotoran	0.3	"
Melprex	0.005	"
Mercaptophos	0.01	"
Metazin	0.3	"
Metaphos	0.02	"
Methylacetophos	0.03	"
Methylmercaptophos	0.01	"
Methylnitrophos	0.25	"
Monuron	5.0	"
Morocide	0.03	Sanitary-toxicologic
M-81	0.001	Organoleptic
OP-7 (OP-10)	0.1	Sanitary-toxicologic
Pentachlorophenol	0.3	Organoleptic
Phosalon	0.001	"
PDN	0.003	Sanitary-toxicologic
Phenazon	2.0	Organoleptic
Phenuron	0.2	General sanitary
Phthalan	0.04	Organoleptic
Polycarbacin	2.0	"
Polychlorocamphene	0.004	"
Polychloropinene	0.2	Sanitary-toxicologic
Prometrin	3.0	"
Propasin	1.0	Organoleptic
Propanide	0.1	General toxicologic
Ramrod	0.01	General sanitary
Sayphos	0.1	Sanitary-toxicologic
Sevin	0.1	Organoleptic
Simazine	Not permitted	
Sodium trichloroacetate	5.0	General sanitary
Solan	0.1	Organoleptic
Tillam	0.01	"
Thiophos	0.003	"
TMTD (thiuram)	1.0	Sanitary-toxicologic
Trichlorometaphos-3	0.4	Organoleptic
Yalan	0.07	"
Zineb	0.03	"

Maximum Permissible Pesticides Residues in Foods (Approved by Deputy Chief Sanitary Physician of the USSR) (after Burkatskaya et al; Sanitary Inspection of Pesticide and Mineral Fertilizer Applications, 1979).

Pesticide	Food	MPRQ, mg/kg
Abate	Sugar beet, vegetables, citruses, cotton oil	0.3 *
Agelon	Corn	0.2 *
Acrex	Cucumbers, apples, citruses	0.05 *
Aldrin **	All foods	Not permitted
Amiben	Cabbage, tomatoes	0.25
Amiphos	Sugar beet and other foods of plant origin	0.1 0.3
Anthio	Apples, pears, plums, citruses, grapes, cabbage and other vegetables	0.2
Arezine	Potatoes	0.1
Arsenites **	Meat and plant products	Not permitted (evaluated by natural contents: up to 0.5 mg/kg for fruit, veget., meat and milk and up to 1.0 mg/kg for cereals)
Atrazine	Cereals, fruit, vegetables	0.1
	Meat and eggs	0.02 *
	Milk	Not permitted *
Baytex (leybacide, sulfidophos)	Grain	0.15
	Meat and meat products	0.2
	Milk and dairy products	Not permitted *
Bayalan	Cereals	0.2 *
Basudin	Cabbage, onion, potatoes	0.1
	Tomatoes, beet, cucumbers	0.5
	Grain	1.0
	Meat fat	0.7
	Carrots, milk, dairy products	Not permitted *
Benlat	Sugar beet, wheat	1.0
	Fruit, cucumbers, tomatoes, cabbage	0.5
Betanal	Beet	0.2
Bromophos	Cabbage, stone fruit, apples, hop	0.5 *
	Berries	0.1 *
Bromtan	Vegetables, melons	3.0
Bordeaux liquid	Fruit and vegetables	5.0 *
	Meat and eggs	2.0 *
Captan	Stone and seed fruit, grapes, veget.	0.35

Pesticide	Food	MPRQ, mg/kg
Caragard	Stones and grapes	0.1
Carbin	Vegetables and fruit	0.1 *
	Cereals	1.0
Carbon disulfide	Grain	10.0
	Flour and groats	1.0
	Bread and other cereal products	0.006
Carbon tetrachloride	Flour and groats	10.0 *
	Cereals	50.0 *
	Bread and other cereal products	0.05 *
Carbophos	Vegetables, fruit and other plant foods	1.0
	Flour and groats (less semolina), and pulses	3.0
Carpene	Fruit	0.6
Chlorophos	Herbs, fruit, cabbage	0.1 *
	Other vegetables	0.2
	Meat, milk and dairy products	Not permitted
Chlor IPC	Carrot	0.05
Cidial	Stones, grapes, citruses	0.1
Ciodrin	Milk and dairy products	Not permitted
	Meat	0.005
Copper vitriol	Fruit	5.0 *
Co-ral	Milk, dairy products, eggs	Not permitted
	Meat, meat products	0,2
Cotoran	Cotton oil	0,1
Cuprosan	Vegetables, fruit, grapes, melons	5.0 *
	Cereals	1.0 *
Cupronaphth	Grapes	4.0
Cuprocine	Potatoes, tomatoes, grapes	1.0
Dacthal	Plant foods	3.0
Delapon	Fruit, grapes, vegetables	1.0 [2]
DDVP	Flour, groats, milk, meat	Not permitted
DDVP	Bran, grain	0.3
	Stone and seed fruit, berries, grapes	0.05
DDT and its metabolites (the chemical banned for use in agriculture)	Fruit, vegetables, potatoes	0.1
	Fish	0.2 (temporarily) together with PCB

Pesticide	Food	MPRQ, mg/kg
	Canned fish	0.2 (temporarily)
	Cereals	0.02
	Milk, baby-food and dietetic dairy, meat, eggs	0.005 (temporarily)
	Milk-processing products (curds, cream and sour cream, butter)	1.25 mg/kg in terms of fat (CMEA standard)
DDT and its metabolites (DDE, DDD) **	Tobacco and tobacco products	0.7
Dilor	Grapes, potatoes	0.15
	Tomatoes and other vegetables, sugar beet	0.2
Diuron	Cotton oil	0.05 *
Dicresyl	Milk and dairy products, eggs	Not permitted *
Dicotex (metaxon)	Grain	0.05 *
Dinitroorthocresol	All foods	Not permitted *
Diphenamide	Vegetables	0.15
Dichloralurea	All foods	Not permitted *
Dibrom	Potatoes	0.1 *
Dichloroethane	Grain	7.0
	Flour	5.0
Dinitrorodan benzene	Vegetables, fruit, grapes	0.2 *
Dosanex	Vegetables, cereals	0.1
Editon	Plant foods	1.0
Eptam	Beet	0.05
Ethylene urea	Plant foods	0.02
Ethylenethiuramammonosulfide		0.3
Euparene	Plant foods	1.3
	Strawberry	Not permitted
Ether sulfonate	Apples, citruses, grapes	3.0
Galecron **	Apples, citruses	0.1
Gardona	Stone and seed fruit, hop, cabbage	0.8
	Berries	0.01
Gamma-isomer of hexachlorocyclohexane (lindane)	Potatoes, peas, cereals	0.5
	Butter, fat	0.2
	Fish	0.2
	Milk, dairy, meat (muscular tissue), eggs, sugar	0.005

Pesticide	Food	MPRQ, mg/kg
Hexachloran (sum of isomers)	Potatoes and vegetables	0.5
	Cereals	0.2
	Butter and fat	0.2
	Fish	0.2
	Milk, dairy, meat, eggs, sugar	0.005
Hexachlorobutadiene	Grapes, grape wine	0.01
	Grape juice	Not permitted
Hexachlorobenzene	Wheat (grain)	0.01
Herbicides from 2,4-D group	All foods	Not permitted
Heptachlor	All foods	Not permitted
Herban	Plant foods	0.1
Inorganic bromides	Grain	35.0
	Flour	10.0
	Vegetables, dry fruits, potatoes, fish	14.0
	Fruits	5.0
	Bread	3.0
Isophos	Rice	0.1 *
Karathane	Melons	1.0
Kelthane	Fruits, vegetables	1,0
Kilval	Vegetables	0,2
Lenacil	Garden beet	0.5
Linuron	Potatoes, pulses, corn	0.1
	Carrot	Not permitted
MG-sodium	Potato tubers, onions	14.0
Mezaronil	Vegetables	0.2
Mercaptophos ** (Systox)	Grain, cotton oil	Not permitted
Mercury-containing pesticides	All foods	Not permitted (evaluated by natural Hg contents in livestock liver — not more than 0.03 mg/kg, and kidneys — not more than 0.05 mg/kg)
Methylmercaptophos	Apples, sugar beet, hop	0.7
Methylnitrophos	Stone, seed, and citric fruits	0.1
Methallyl chloride	Legumes	3.5
Methaldehyde	Vegetables, fruits	0.7
Metaphos	All foods	Not permitted
Methoxychlor	All foods	14.0
Miltox-special	Vegetables, fruits, grapes, melons	0.5 * (by zineb)
	Grain	1.0 * (by zineb)

Pesticide	Food	MPRQ, mg/kg
Monuron	Stones, grapes, citruses, vegetables	0.05 *
Morocide	Fruits, citruses	0.002
M-81 (intrathion) **	Apples	0.5
Morestan	Stones, grapes	Not permitted
2M-4CM	Cereals	0.1
2M-4CD	Cereals	0.25
Neoron	Cotton oil, oil cake	0.02 *
Nitraphen	All foods	Not permitted
Octamethyl **	Mulberries	Not permitted
Pentachloronitrobenzene	Cereals	1.0 *
Phenocapton	Apples	0.3
Phosalon	Stone and seed fruits, citruses, grapes, cereals, potatoes	0.2
Phosphamide	Fruits, citruses, potatoes, vegetables, cereals	1.0
Phthalan	Stone and seed fruits, grapes, vegetables, potatoes	2.0
Phthalophos	Sugar beet	0.25
	Potatoes	Not permitted
Phostoxin	Cereals	0.01 (by hydrogen phosphide)
Phytobacteriomycin	Haricot, soya	Not permitted
Polymarcin	Apples, grapes, tomatoes, potatoes	0.1 *
Polycarbacin	Vegetables, fruits, berries	1.0
Polychlorocamphene	Potatoes, sugar beet	0.1
	Green pea, sugar, milk, meat, eggs	Not permitted
Polychloropinene	Potatoes, sugar beet, peas, sugar, milk, meat, eggs	Not permitted
Preparation 242	Flour	Not permitted
(Chloropicrin)	Grain for processing	2.0
Propanide	Rice	0.3
Propasin	Legumes	0.2 *
Prometrin	Vegetables, potatoes	0.1
	Carrot	Not permitted
Ramrod	Cabbage and other vegetables	0.2
Reglon	Plant foods	0.05
Ronit	Sugar and garden beet	0.3 *

Pesticide	Food	MPRQ, mg/kg
Sayphos	Vegetables, potatoes, sugar beet, melons, pulses, stones and other fruits	1.0
Sevin	Fruits, berries, corn, cotton seeds	Not permitted
Semeron	Cabbage	0.05
Simazine	Fruits	0.2
	Grapes	0.05
	Cereals	1.0
Sodium silicofluoride	Meat	0.4
Sodium trichloroacetate	Vegetables, fruits, grain	0.01 [2]
Solan	Tomatoes	1.5
Tedion	Vegetables, fruits	0.7 *
Tenoran	Carrots	0.02 *
Terbacil	Apples, citruses, grapes, peaches	0.05
Thiazone	Potatoes, cucumbers, other vegetables, fish	0.5
Tillam	Vegetables, tomatoes, sugar and garden beet	0.05 *
Thiophos**	All foods	Not permitted
TMTD	"	"
Trichlorometaphos-3	Fruits, vegetables	1,0
	Grain	0.5
Trichothecin	Cucumbers	1.0
Trolene	Milk, dairy products, eggs	Not permitted
	Meat and meat products	0.3
Yalan	Rice	0.2
Zineb	Fruits, vegetables	0.6
	Cereals	1.0
	Milk and dairy products	Not permitted

Notes. The term "not permitted" denotes failure to detect the chemical at the sensitivity level of the officially designated method;

* identifies a standard set by calculation;

** identifies pesticides currently banned for production and use in the Soviet Union.

Maximum Permissible Pesticide Residues in Livestock Forages (approved May 17, 1977 by the Chief State Veterinary Inspector of the USSR)

Pesticide	MPRQ, mg/kg	
	Dairy cattle, egg-laying poultry	Fattening livestock and poultry
Aldrin (dieldrin)	Not permitted	Not permitted
Anthio	2.0	2.0
Arsenite pesticides	Not permitted (evaluated is natural arsenic content in forages, to be not more than 1.0 mg/kg)	Not permitted
Atrazine	1.0	1.0
Butyphos	3.0	3.0
Carbon disulfide	10.0	10.0
Carbon tetrachloride	50.0	50.0
Carbophos	2.0	5.0
Chlorophos	1.0	3.0
DDT (sum of isomers and metabolites)	0.05	0.05
2,4-D (all derivatives)	0.1	0.6
Dinitroorthocresol	Not permitted	Not permitted
Dursban	0.2	0.2
HCCH (sum of isomers)	0.05	0.2
Heptachlor (heptachlor epoxide)	Not permitted	Not permitted
Inorganic bromides	35.0	35.0
Mercury-containing pesticides	Not permitted (evaluated is natural mercury content, to be not more than 0.02 in green, succulent and grain forages and not more than 0.05 mg/kg in mixed feeds containing fish flour)	Not permitted
Metaphos	Not permitted	0.5
Methylmercaptophos	1.0	1.0
Methylnitrophos	1.0	2.0
Phosphamide	2.0	2.0
Phthalophos	1.0	2.0
Polychlorocamphene	Not permitted	0.25
Polychloropinene	"	0.25
Sevin	1.0	1.0
Sodium rhodanide	Not permitted	0.5
TMTD	"	Not permitted
Valexon	—	0.6

MACs for Pesticides in Soil
(approved by Deputy Chief Sanitary Physician of the USSR; Nos 1134—73 of December 19, 1973 and 1496—76 of August 11,1976)

Pesticide	MAC, mg/kg	Pesticide	MAC, mg/kg
Carbophos	2.0	DDT	1.0
Chlorophos	0.5	Polychloropinene	0.5
Chloramp	0.05	Polychlorocamphene	0.5
Hexachloran	1.0	Prometrin	0.5
Hexachloran gamma-isomer	1.0	Sevin	0.05

TSELs for Pesticides in the Air of the Working Zone
(Standard 12.1.005—76)

Pesticide	Chemical name	Concentration, mg/m^3	State of aggreg.	Class of hazard
Acrex	Isopropyl-2(1-methyl-n-propyl)-4,6-dinitrophenyl carbonate	0.2	a	2
Afugan	2-(0,0-diethylthiophosphoryl)-5-methyl-6 carboethoxy-pyrazole-(1,5a) pyrimidine	0.5	v+a	2
Basudin	0,0-diethyl-0-(2-isopropyl-4-methyl-pyrimidyl-6) thiophosphate	0.2	v+a	2
Baytex	0,0-dimethyl-0-(4-methyl-mercapto-3-methylphenyl) thiophosphate	0.3	v+a	2
Benlat	Methyl-1-(butylcarbamayl)-2-benzimidizole-carbamate	0.01	a	1
Cupronaphth	Copper naphthenate	2	a	3
CGA-18809	S-(6-chloroxyazolopyridinyl)-2-yl-3-methyl-(0,0-dimethylthiophosphonate)	0.5	v+a	2
Demuphos	0,0-dimethyl-N-methylisopropylurethanphosphate	1	v+a	3
Dicryl	3,4-dichloromethylacrylanilide	0.1	a	1

Addendum 7 (contd.)

Pesticide	Chemical name	Concentration, mg/m³	State of aggreg.	Class of hazard
Dilor	2-exo-4,5,6,7,8,8-heptachlor-3',4',7,7'-tetrahydro-4,7-methanoindene	0.1	v+a	2
Dursban	0,0-diethyl-0-(3,5,6-trichloropyridine) thiophosphate	0.3	v+a	2
Ethaphos	0-ethyl-S-propyl-2,4-dichlorophenyl-phosphate	0.3	v+a	2
EF-2	2-dichloromethylene-3a-7a-dichloro-3a,4,7,7a-tetrahydro-4,7-methanoindan-1,3-dion	1	a	3
Euparene	N,N-dimethyl-N-phenyl-N-fluorodichloromethylthiosulfamide	1	a	3
Chloreth	Ethyl-2,4,5-trichlorophenyl-ß-chloroethylphosphate	0.2	v+a	2
Hydrel	Hydrazinite ß-chloroethylphosphonate	1	a	3
Hostaquick	0,0-dimethylphosphoryl-6-chlorobicyclo (3,2,0-hepta-1,5, diene)	0.3	v+a	2
Hostathion	1-phenyl-3-(0,0-diethylthiophosphoryl)-1,2,4-thiazole	0.2	v+a	2
Kitazin	0,0-diisopropyl-S-benzylthiophosphate	1	v+a	3
Lasso	N-methoxymethyl-2,6-diethylchloroacetanilide	0.5	a	2
Maloran	N-(4-bromo-3-chlorophenyl)-N-methoxy-N'-methylurea	1	a	3
Menide	3-chloro-4-methylpropionanilide	1	a	3
Orthene	O-S-dimethyl-N-acetylphosphoramidothionate	0.7	v+a	2
Pesticide 228-F	O-butyl-S-methylbenzyldithiophosphonate	0.3	v+a	2
Prefar	S-(0,0-diisopropyldithiophosphate) benzenesulfamidoethyl	1	v+a	3
Roundup	N-(phosphonomethyl) glycin	1.5	v+a	3
Reglone	1,1-ethylene-2,-2dipyridyl-bromide	0.2	v+a	2

Pesticide	Chemical name	Concentration, mg/m^3	State of aggreg.	Class of hazard
Rycide	0,0-diethyl-S-benzylthiophosphate	0.3	v+a	2
San 197	0,0-dimethyl-0-(2-ethyl-4-ethoxypyrimidinyl 6-(thiophosphonate)	0.5	v+a	2
Sayphos	0,0-dimethyl-S-(4,6-diamino-1,3,5-triazinyl-2)-methyl-dithiophosphate	1	v+a	3
CP-52 223	N-isobutoxymethyl-2'-chloroacetate-2,6-dimethylanilide	0.3	a	2
Suffix	Ethyl ether-N-benzoyl-N-(3,4-dichlorophenyl) 2-aminopropionate	0.5	a	2
Tachigaren	3-oxy-5-methyl-isoxazole	1	a	3
Thiocron	0,0-dimethyl-S-(2-methoxyethylcarbamoylmethyl) dithiophosphate	0.15	v+a	2
Topsin-M	1,2-bis-3-(methoxycarbonyl-2-thiourea)-benzene	1.5	a	3
Trichothecin	Antibiotic	0.2	a	2
Valexon	0,0-diethylthiophosphoryl-2-oxyminophenylnitryl acetate	0.3	v+a	2

Addendum 8

The List of Principal Antidotes for Pesticide Intoxications

Intox.-causing pesticides	Antidotes and their dosing
Organophosphorus pesticides: methyl mercaptophos, phosphamide, chlorophos, metaphos, DDVP, etc.	*Atropine sulfate* — in the early stage of OPP intoxication (nausea, vomit, bronchospasm, psychic and motor excitation) 2—3 ml of 0.1% atropine sulfate injected intramuscularly; in the stage of muscular fibrillation and convulsions and in the state of coma 4—6 ml of 0.1% atropine sulfate solution injected intravenously and then 2—3 ml intramuscularly every 8—10 min; after that 20—25 mg of atropine sulfate injected within the hour and another 50 mg or so per day (24 hrs) in the period of supportive atropinization (Luzhnikov, 1977). The atropinization maintained until normal breathing resumed, bronchorrhea halted and tachycardia seen to appear. The antidote injection combined with

Intox.-causing pesticides	Antidotes and their dosing

the use of cholinesterase reactivators, resuscitative measures and means of pathogenetic and symptomatic therapy. Cholinolytic drugs also antidotal for the anticholinesterase derivatives of carbamic acid (Buslovich, Zakharov, 1972).

Dipyroxime (TMB-4, trimedoxime bromide). Chemically, it represents 1,3-trimethylene-bis-(4-pyridinaldoxime) dibromide: a yellowish fine-crystalline powder, odourless, with bitter taste and readily soluble in water; a cholinesterase reactivator applied in combination with atropine and other cholinolytic drugs to treat OPP poisoning; with early signs of OPP intoxication, 1 ml of 15% dipyroxime solution injected subcutaneously or intramuscularly in 2–3 ml of 0.1% atropine sulfate; if necessary, the dipyroxime injection repeated after 2–3 hours; prescription of the drug for the later stages of OPP poisoning only marginally effective and hardly advisable for fear of probable toxic side-effects.

Isonitrozine-1-dimethylamino-2-isonitrosobutanone-3-chloride- a white crystalline powder soluble in water; a cholinesterase reactivator with the ability for CNS penetration, applied routinely in conjunction with atropine and dipyroxime at 3 ml of 40% solution intramuscularly; in case of severe poisoning (convulsions, coma) 3 ml of 40% solution injected intravenously and then repeated every 30–40 min until fibrillation stopped and consciousness regained in the total dose of 8–10 ml (3–4 g).

Diethyxime — hydrochloride-N-diethylaminoethyl ether of n-brombenzthiohydroximic acid; a fine-crystalline powder, odourless with bitter taste and readily soluble in water; the water solution can withstand sterilization with fluid steam at 100°C for 30 minutes; supplied in 5 ml ampoules of 10% water solution for intramuscular injection (the therapeutic dose 7–10 mg/kg); for joint use with dipyroxime the latter's dose reduced to 1.5 mg/kg, and of diethyxime up to 5 mg/kg; diethyxime rapidly reactivates AChE in the brain and eliminates symptoms of OPP poisoning; it is 5–7 less toxic than dipyroxime; stored with precautions (List B) in a place protected from light at minimum temperature of +8°C.

Dialcob — bis-(N-allyldiethanolamine)-cobalt chloride: a fine-crystalline powder, odourless, pale-pink colour, readily soluble in water (about 20% at 18°C); unstable in water and oxidized when exposed to atmospheric

Intox.-causing pesticides	Antidotes and their dosing
	oxygen; sterilized by irradiation from bactericidal lamp; dialcob restores OPP — inhibited cholinesterase activity; applied to treat acute OPP intoxications in combination with atropine by intramuscular injection, mild intoxications (excitement, myosis, hidrosis, salivation, initial symptoms of bronchorrhea) treated with single intramuscular dose of 0.3 g (2 ml of 15% solution), and moderate and severe intoxications (muscular fibrillation, convulsions, comatose state) with repeated intramuscular injections every 2—3 hours in the same dose as supplement to intensive atropinization; in especially severe cases the dose per course increased to 1.2 g (four injections at 4—6 hour interval); prescription of dialcob contraindicated in allergic states; delivered in 0.3 g hermetically sealed vials; if needed, dissolved in ampulled 0.1% atropine sulfate solution; stored with care (List B).
Halogen-containing hydrocarbons of aliphatic series	*Cysteine* — L-cysteine-1-amino-2-mercapto propionic acid: a white crystalline powder, slightly odorous and soluble in water; water solutions unstable and oxidized by atmospheric oxygen with precipitation; degraded if sterilized; supplied in powder form and recommended for use at 0.5 g the first day 2—3 times every 3 - 4 hours, the next 2—3 days 2 times at 4—6 hours (Miziukova, Bakhishev, 1971). *Acetylcysteine* — N-acetyl-L-cysteine: a white or yellowish crystalline powder, emits slight smell; readily soluble in water; recommended to treat dichloroethane poisoning (Miziukova et al., 1978); supplied in powder form or 5 and 10 ml ampoules of 20% solution as a mucolytic means and 10 ml of 5% solution (for intravenous injections in dichloroethane intoxications); stored in a place protected from light at temperatures from 0 to 5°C; sterilized acetylcysteine solutions reduced by addition of sodium hydroxide solution to pH 7—7.5; keep acetylcysteine solution from contact with metal and rubber to prevent formation of sulfates; as a stabilizer, use 0.04% disodium EDTAte; acetylcysteine solution is a colourless liquid with characteristic smell; after opening the ampoule may turn pink but remains usable as before; acetyl cysteine injection used to treat dichloroethane intoxications: intravenously at 1.25—1.5 g (25—30 ml of 5% solution) as soon as possible after poison gains access into the body; 4—5 injections every 3—4 h the first day and 2 injections every 6—8 h the second day; after the

Intox.-causing pesticides	Antidotes and their dosing
	third injection change to intravenous injection dropwise if necessary; prescription of acetylcysteine is not to the exclusion of other pathogenetic and symptomatic remedies.
Diene-synthesis organochlorines (aldrin, dieldrin, heptachlor, etc.)	*Tocopherol acetate* (vitamin E acetate) — alfa-tocopherol-6-acetoxy-2 methyl-2 (4,8,12 trimethyltridecyl)-chroman: a light-yellow transparent oily liquid, slightly odorous and darkening when oxidized in light; an active antioxidant, it prevents oxidation of various substances — the property utilized in treatments with drugs subject to lethal synthesis by oxidation; prescribed intramuscularly in oil solutions; delivered in 1 ml ampoules; contains 50, 100 and 300 mg per ml; the dose is 100—300 mg/day; contraindications include severe cardiosclerosis and myocardial infarction.
Arsenites and compounds containing mercury and other heavy metals	*Unithiol* — sodium 2,3-dimercaptopropanesulfonate: a white or cream-shaded fine-crystalline powder emitting a weak mercaptane smell; readily soluble in water; used to treat acute and chronic intoxications by compounds of arsenic, mercury and other heavy metals; injected intramuscularly or subcutaneously in 5% water solution at 5—10 ml every 6—8 h the first day, every 8—12 h the second day, and 1—2 injections daily the following 6—7 days; for chronic intoxications, unithiol is prescribed internally at 0.5 g (pills or capsules) two times daily in courses of 3—4 days each; in inhalational poisoning injected as aerosols in a 10-day course of two 20-minute inhalations/day (at 5 ml of 5% solution), to remove attendant hydrogen-sulfide smell add 1—2 drops of menthol oil; in efficacy, the inhalational unithiol therapy not inferior to intramuscular injection (Sidorenko, 1978). *Calcium tetacine* — calcium-disodium ethylenediaminetetraacetate; delivered in 10% solution for injections; a colourless transparent liquid; a complex-forming compound; calcium can be substituted by metal ions to form water-soluble low-toxic compounds rapidly excretable in urine; used to treat acute and chronic intoxications by heavy metals (lead, mercury, cadmium, cobalt, etc.) through intravenous injection dropwise in isotonic sodium-chloride or glucose solutions; a single dose is 2 g (20 ml of 10% solution) and daily dose 4 g; if injected twice a day, the minimum interval must be at least three hours; routinely injected daily (3—4 days) 3—4 day interval; the course is one-month-long; in case of chronic intoxication by heavy metals use 0.4 g pills (four times) day or 0.25 g (eight times) day every other day during a 20—30-day course, totalling 20 g or above for

Intox.-causing pesticides	Antidotes and their dosing
	the course but not more than 30 g (Mashkovsky, 1977); contraindicated in nephrites, nephroses and hepatic conditions involving functional disorders.
	Disodium ethylenediaminetetraacetate: forms complex compounds with various cations; used to treat heavy-metal poisoning by intravenous injection in 5% glucose solution dropwise at 2—4 g daily during a 3—6-day course;
	Penicillamine — 3,3-dimethycysteine; prescribed in intoxications by copper, mercury, arsenic, lead, and other metal compounds; the daily dose is 2 g; complications possible including leukopenia, thrombocytopenia, agranulocytosis, anemia, alimentary canal disorders, etc.; supplied in 0.15 and 0.25 g capsules and pills and prescribed internally.
Hydrocyanic acid and its salts	*Sodium thiosulfate* — sodium hyposulfite: colourless crystals soluble in water; injected intravenously at 5—10 ml of 30% solution, and up to 50 ml in acute intoxications by hydrocyanic acid; delivered in 5, 10 and 50 ml ampules of 30% solution.
	Methylene blue (trimethylthionine chloride); dark-green crystalline powder or dark-green crystals with a bronze shine; water solutions blue; endowed with oxidative and reductive properties and can serve as donor and acceptor of hydrogen in the body; its water solution injected intravenously to persons poisoned by hydrocyanic-acid pesticides, at 50—100 ml or as 1% solution in 25% glucose solution (chromosmone); has the ability to convert hemoglobin in part to methemoglobin whose trivalent iron binds the hydrocyanic acid and diminishes its interaction with tissue respiratory enzymes.
	Sodium nitrite — white tinged yellow crystals soluble in water; injected intravenously to cyanide poisoning cases at 10—20 ml of 1—2% solution (methemoglobin former); stored in places protected from light.
	Amylnitrite — isoamyl ester of nitrous acid: a transparent yellowish volatile substance with a characteristic fruit smell; inhalation of amylnitrite vapors causes dilatation of coronary and cerebral vessels; the effect is soon to appear but also soon to subside; has the ability to convert blood hemoglobin in part to methemoglobin, the property utilized to treat cyanide intoxications; used inhalationally by applying 2—3 drops on handkerchief, cloth or wad; the maximal single dose is 6 drops and daily dose 30 drops; delivered in 0.5 ml darkglass ampules and stored in a cool place protected from light.

REFERENCES

Abramova Z. I. Use of natural antioxidants to prevent and treat intoxications by poisons with different modes of action. Obshchiye voprosi promyshlennoi toksikologii. Moscow, 1967, 146—149 (in Russian).

Ackerman N., Faust N., Kagan Y. S., Voronina V. M. Metabolism and toxic behaviours of phthalimide derivatives in albino rat. Arch. Toxicol., 1978, 40, 255—261.

Albert A. Selective toxicity and related topics. London, 1971.

Aldridge W. N. Mechanism of interaction of organophosphorus compounds and carbamates with esterases. WHO Bull., 1972, 44, No. 1—2—3, 27—33 (Russian transl.).

Aldridge W. N., Johnson M. C. Side-effect of organophosphorus compounds: neurotoxic action. WHO Bull., 1972, 44, No. 1—2—3, 271—279 (Russian transl.).

Aleksashina Z. A., Buslovich S. Y., Kolosovskaya V. M. Embryotoxic action of the diethylamine salt 2,4-D. Gigiena i sanitariya, 1973, 100—101 (in Russian).

Alexeeva O. G., Dueva L. A. Allergy to industrial chemical compounds. Moscow, 1978, 271 (in Russian).

Altareva L. A. Work hygiene and prevention of intoxications in sugar-beet crop management. In: Current Questions of Pesticide Use. 5th National Conference. Abstr. Rep. Kiev, 1975, 66—67 (in Russian).

Albert R., Altshuler B., Friedman L., Yanysheva N. Y., Antomonov Y. G. Approaches to standard-setting for carcinogens in the environment. Gigiena i sanitariya, 1975, 10, 84—90 (in Russian).

Andreeshcheva N. G. Prediction of biological effect according to chemical structure and principal physicochemical properties of organic substances. Proc. Summing-Up USSR—USA Symposium on Environmental Hygiene, December 9—14, 1974. Moscow, 1975, 32—35 (in Russian).

Anina I. A. Possible use of indices of nucleic acid metabolism to predict long-term effects of some pesticides. Gigiena truda i profzabolevaniya, 1975, 3, 51—53 (in Russian).

Archakov A. I. Microsomal oxidation. Moscow, 1975, 324 (in Russian).

Arkhipova O. G., Zorina L. A., Sorkina N. S. Complexones in the clinical course of occupational diseases. Moscow, 1975 (in Russian).

Ashbel S. I., Stroikova G. D., Khil R. G., Yavorskaya Y. S. Prevention of occupational mercurialism among granosan production workers by means of the new Soviet antidote D-penicillamide. Gigiena truda i profzabolevaniya, 1974, 7, 25—28 (in Russian).

Badaeva L. N. Systems structural criteria to judge neurotoxic effects from pesticides (polychlorocamphene and valexon) in experiment. Ukraine Academy of Sciences Reports, 1978, B, 3, 262—267 (in Russian).

Baskakov Y. A. New synthetic plant growth regulators and herbicides. J. Nat. Mendeleev Chem. Soc. 1978, 23, 2, 149—160 (in Russian).

Bakhishev G. N. State of some parameters of carbohydrate metabolism during acute intoxication of rats by methyl bromide. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1970, 392—398 (in Russian).

Beznosko B. K., Voronov I. B., Feigman O. E., Krivenchuk V. E. Central action of the new cholinesterase reactivator diethyxime. Farmakologiya i toksikologiya, Moscow, 1977, 5, 531 (in Russian).

Bezugly V. P., Kaskevich L. M., Ilyina V. I., Sivitskaya I. I., Gorskaya N. Z., Soboleva L. P., Komarova L. I., Fokina K. V., Yurchenko I. V. Methodologic approaches to designing methods for diagnosis of pre-clinical and early clinical symptoms of pathologic states of chemical etiology. In: Pesticide Use Hygiene for Different Climate-Geographic Zones. Yerevan, 1976, 149 (in Russian).

Belenky I. L. Elements of quantitative evaluation of pharmacologic effect. Leningrad, 1963, 149 (in Russian).

Bochkov N. P. Basic principles governing quantitative evaluation of chemical mutagenesis in man. In: Questions of Hygienic Standardization in the Study of Long-Term Effects of Industrial Chemical Exposures. Moscow, 1972, 13—16 (in Russian).

Bochkov N. P., Shram R. Y., Kuleshov N. P., Zhurkov V. S. System of evaluation of chemicals for mutagenicity to man: general principles, practical recommendations and further elaboration. Genetika, 1975, II, 10, 156—169 (in Russian).

Brestkin A. P. Role of hydrophobic interaction in cholinesterase inhibition. In the book: Chemistry and Use of Organophosphorus Compounds. Moscow, 1973, 322—333 (in Russian).

Broocs G. T. Chlorinated insecticides. CPC Press. 1976, 249.

Burkatskaya E. N. Toxicology of dinitrophenol pesticides and prevention of intoxications by occupational exposure. Kiev, 1974, 52 (in Russian).

Burkatskaya E. N., Rudnichenko M. F. Study of allergic pesticide properties, a current problem of agricultural toxicology. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1969, 166—172 (in Russian).

Burkatskaya E. N., Ivanova Z. V., Lysina G. G. Medical examination of persons occupationally exposed to pesticides. Kiev, 1978, 184 (in Russian).

Burkatskaya E. N., Lysina G. G., Karpenko V. N. Laboratory diagnosis of pesticide intoxications. Moscow, 1978, 128 (in Russian).

Buslovich S. Y., Zakharov G. G. Clinical features and treatment of acute intoxications by poisonous chemicals. Minsk, 1972, 128 (in Russian).

Casida Y. E. Pyrethrum. The natural insecticide. London, 1973, 329.

Chaklin A. V. A problem of the century. Moscow, 1976, 143 (in Russian).

Chepinoga O. P. Pesticides: investigation for blastomogenic, mutagenic, embryotoxic and gonadotoxic properties and its results. In the book: Pesticide Use, Hygiene Toxicology and Clinical Features of Poisonings. Kiev, 1970, 30—39 (in Russian).

Chepinoga O. P., Zastavnik N. T., Zadorozhnaya N. L. Blastomogenic hazard of some dithiocarbamic acid derivatives and approaches to its detection. Kiev, 1969, 153—166 (in Russian).

Cherkes A. I. Fundamentals of the toxicology of chemical warfare agents. Kiev, 1939, 387 (in Russian).

Courtney K. D. Bull. Environ. Control and Toxicology, 1976, 16, 6, 674—681.

Dauterman W. S. WHO Bull., 1972, 44, 1—3, 135—152.

Deichman B. C., Le Blanc T. J. Industrial hygiene and toxicology, 1943, 25, 9, 415.

Demidenko N. M. Toxicity studies of poisonous chemicals in regard of their combined and consecutive action. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1968, 718—723 (in Russian).

Denisenko P. P., Ostrovsky M. M., Lisitsina K. A. Characteristics of chlorophos toxicity to mice under different conditions of heat exposure and physical burden. Gigiena i sanitariya, 1968, 8, 14—18 (in Russian).

Dinerman A. A. Some points concerning teratogenic action of chemicals. Gigiena i sanitariya, 1968, 7, 77—82 (in Russian).

Dinerman A. A., Shigan S. A. Sources of some common environmental pollutants and long-term effects of their biological impact. In the review: Long-Term Effects from Biological Impact of Some Chemicals. 1975, 43—94 (in Russian).

Dishovsky H. D. Experimental studies on the mechanism of action of some cholinesterase reactivators in the intoxication by dimethylchlorvinylphosphate. 1971, 31, (in Russian).

Dishovsky H. D., Kagan Y. S., Kovtun S. D., Kokshareva N. V., Rodionov G. O., Sasinovich L. M. The physiologic mechanism for the therapeutic effect of dipyroxime. Fiziologicheskii zhurnal AN Ukr.SSR, 1973, XXIX, 3, 310—314 (in Russian).

Dishovsky H. D., Kagan Y. S., Kovtun S. D. Electrophysiologic analysis of the action of the reactivator dipyroxime in the acute intoxication by O,O-dimethyl-2,2-dichlorvinylphosphate. Farmakologiya i toksikologiya, 1974, 3, 365—366 (in Russian).

Dixon M., Webb E. Enzymes. Moscow, 1966, 816 (Russian transl.).

Dubinin N. P. Environmental mutagens and human heredity. Genetic aftermaths of environmental pollution. Moscow, 1977, 3—20 (in Russian).

Dubinin N. P. Genetics. Moscow, 1978 (in Russian).

Dunaisky V. B. Polychloropinene. A report of toxicologic and hygienic evaluation of new pesticides. Moscow, 1973, 56—67 (in Russian).

Dzhunusova R. M. Hygienic investigation of chlorophos, metaphos and their combined effect on animal organism. Moscow, 1966, 21 (in Russian).

Elizarova O. N. Determination of threshold doses of industrial poisons by oral administration. Moscow, 1971, 192 (in Russian).

Engst R., Macholz R. M., Kukawa M. Z. Gesamte Hyg., 1976, **22**, 7, 495—498.

Eto M. Organophosphorus pesticides. Organic and biological chemistry. CRC Press, 1974, 369.

Evreev V. N., Golub V. A. Cobalt with N-alkyl derivatives of diethanolamine. Necrganicheskaya himiya, 1976, 21, 2, 465—467 (in Russian).

Ershova E. A. A comparative toxicologic study of new organophosphorus insecticides and acaricides containing residual aminoacids in their structure. Kiev, 1972 (in Russian).

Faerman I. S. Functional state of the liver and stomach among organophosphorus insecticide production employees. Ter. arch., 1965, 37, 51—54 (in Russian).

Fomenko V. P., Strekalova E. E. Development of methods to predict mutagenic activity of industrial poisons in experiment. In: Questions of Hygienic Standardization in the Study of Long-Term Effects of Industrial Chemical Exposures. Moscow, 1972, 16—24 (in Russian).

Fridlyand I. G. Non-specific manifestations of the action of chemical agents. Pharmacology, chemotherapeutic drugs, toxicology. 1969. Problemi toksikologii. Moscow, 1971, 29—67 (in Russian).

Gabovich R. D., Murashko V. A., Timoshenko Y. G. Tolerance of organism to hexachlorobenzene for different combinations of air temperature and UV-irradiation. Gigiena i sanitariya, 1975, 8, 17—21 (in Russian).

Gadaskina I. D., Filov V. A. Transformation and determination of industrial organic poisons in the body. Leningrad, 1971, 304 (in Russian).

Gadaskina I. D., Levina E. N. General regular patterns in the entry, transport, distribution and elimination of industrial toxic agents from the body. In the book: Fundamentals of General Industrial Toxicology. Leningrad, 1976, 136—159 (in Russian).

Giebel W., Lohs K., Wiedner G. P., Zieberath D. Experimentalle Untersuchungen über die hepatoxische und kanzerogene Wirkung phosphororganischer Verbindungen. Arch. f. Geschwulstforschung, 1971, 37, 4, 303—312.

Giebel W., Lohs K., Wiedner G. P. Über die kanzerogene hämatoxische und hepatoxische Wirkung pestizider organischen Phosphorverbindungen. Arch. f. Geschwulstforschung, 1973, 4, 311—328.

Glozman O. S., Kasatkina A. P., Detoxifying therapy in acute poisonings and autointoxications. Tomsk, 1970, 320 (in Russian).

Golikov S. N. Prevention and treatment of organophosphorus pesticide poisonings. Moscow, 1968, 168 (in Russian).

Golikov S. N. Emergency aid in acute intoxications. Moscow, 1978, 311 (in Russian).

Golikov S. N., Rozengart V. I. Cholinesterases and anticholinesterase agents. Leningrad, 1964, 384 (in Russian).

Golikov S. N., Zaugolnikov S. D. Cholinesterase reactivators. Leningrad, 1970, 166 (in Russian).

Golikov S. N., Sanotsky V. I. Disfunction of presynaptic nerve terminals in response to anticholinesterase agents. Abstr. Rep. 4th National Pharmacol. Congr. Leningrad, 1976, 48—49 (in Russian).

Goloma E. A. A contribution to clinical characteristics of the intrauteral intoxication by granosan. Gigiena i sanitariya, 1962, 9, 101—103 (in Russian).

Golubev A. A., Subbotin V. G. Possible applications for calculation techniques to establish tentative maximum permissible concentrations of harmful substances in bodies of water. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1968, 284—288 (in Russian).

Golubev A. A., Liublina E. I., Tolokontsev N. A., Filov V. A. Quantitative toxicology. Leningrad, 1973, 285 (in Russian).

Goncharuk G. A. Impact of the organomercury pesticides mercuran and mercurhexane on progeny of albino rats. Gigiena i sanitariya, 1968, 6, 111—113 (in Russian).

Goncharuk G. A. An experimental study on the effects of pesticides from the group of organomercury compounds on generative function and progeny. Gigiena i sanitariya, 1971, 7, 32—35 (in Russian).

Gostinsky V. D., Maeva T. E. Comparative sensitivity of humans and animals to the entry of chemicals via skin. The dermal route of entry of industrial poisons into the body and its prevention. Moscow, 1977, 12—17 (in Russian).

Gofmekler V. A. Embryotropic effect of chemical air pollutants. Gigiena i sanitariya, Moscow, **12**, 9, 7 (in Russian).

Gofmekler V. A., Khuriev B. B. An experimental research into the embryotropic action of methylmercaptophos by the inhalational entry into the body. Gigiena i sanitariya, 1971, 1, 27—32 (in Russian).

Gubler E. B., Genkin A. A. Use of non-parametric statistical criteria in medical-biological research. Leningrad, 1973, 144 (in Russian).

Hochacka P. W., Somero L. N. Strategies of biochemical adaptation. Philadelphia-London-Toronto, 1973.

Hollingworth R. M. Insecticides: Biochemistry and Physiology, New York-London, 1976.

Ivanova-Chemishanska L. A contribution to the toxicology of some dithiocarbamates. Gigiena i sanitariya, 1971, 11, 95—98 (in Russian).

Iskanderov T. I. Current questions relative to the combined effect of high temperature and pesticides. Problemi gigieni i organizatsii zdravoohraneniya v Uzbekistane. 1977, 6, 38—44 (in Russian).

Ivanov N. G. A rapid method for determination of safe chemical levels based on recording specific symptoms of intoxication. Proc. 2d Nat. Symp. Kiev, October 12—14, 1976, 81—83 (in Russian).

Kabachnik M. I. Impact of organophosphorus substances on conduction of neural excitation. Vestnik AN SSSR, 1968, 5, 86—94 (in Russian).

Kabachnik M. I., Brestkin A. P., Mikhelson M. Y. The mechanism for physiological action of organophosphorus compounds. Proc. Mendeleev Congr. Gen. Appl. Chemistry. Moscow, 1965, 32, (in Russian).

Kagan Y. S. Toxicology of organophosphorus insecticides and work hygiene in their use. Moscow, 1963, 327 (in Russian).

Kagan Y. S. The mechanism for selective toxicity of organophosphorus insecticides. Gigiena truda, 1963, p. 38—43 (in Russian).

Kagan Y. S. Cumulation coefficient as a quantitative measure of cumulative action of toxic chemicals. In the book: Pharmacology and Toxicology. Kiev, 1964, 231—237 (in Russian).

Kagan Y. S. Quantitative criteria of harmfulness for chemical substances. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1965, 46—60 (in Russian).

Kagan Y. S. Cumulation, criteria and evaluation methods. Prediction of chronic intoxications. In: Principles of Maximum Allowable Concentrations. Moscow, 1970, 49—65 (in Russian).

Kagan Y. S. Method for quantitative evaluation of combined and complex effects on the organism from chemical and physical environmental factors. Gigiena i sanitariya. 1973, 12, 89—91 (in Russian).

Kagan Y. S. Current problems of toxicologic pesticide research. In the book: Plant Protection, vol. I, 1972, 285—316 (in Russian).

Kagan Y. S. Key trends in research on pesticide toxicology and prevention of their harmful effects. J. Mendeleev Nat. Chem. Soc., XIX, 1974, 2, 192—200 (in Russian).

Kagan Y. S., Stankevich V. V. Cumulation coefficient as a quantitative criterion for evaluation of cumulative action of toxicants. In: Current Problems of Work Hygiene, Industrial Toxicology and Occupational Diseases in Petroleum and Petrochemical Industries, V, 1964, 48—49 (in Russian).

Kagan Y. S., Liublina E. I., Sanotsky I. V. et al. Principles of research into combined effects of chemicals and their hygienic regulation methods. Proc. 15th Nat. Congr. Hyg. and Sanitary Physicians, May 23—27, 1967, Kiev, 133—135 (in Russian).

Kagan Y. S., Miziukova I. G., Tarakhovsky M. L., Terekhov I. T. Treatment of acute intoxications. Kiev, 1973, 227 (in Russian).

Kagan Y. S., Lukaneva A. M., Rodionov G. A. Impact of pesticides on development of some experimental pathologic states in the cardiovascular system. Farmakologiya i toksikologiya. Kiev, 9, 1974, 189—195 (in Russian).

Kagan Y. S., Kokshareva N. V., Sasinovich L. M., Krivenchuk V. E. Experimental data on a new cholinesterase reactivator from the group of thiohydroxylic esters. Farmakologiya i toksikologiya, 1975, 3, 294—298 (in Russian).

Kagan Y. S. Some problems of theory concerning disease states of chemical etiology. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1976, 60—69 (in Russian).

Kagan Y. S. Toxicology of organophosphorus pesticides. Moscow, 1977, 296 (in Russian).

Kagan Y. S., Petrunkin V. E., Evreev V. N. Complex cobalt compounds with monoethanolamine as potential reactivators of cholinesterase inhibition. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1968, **6**, 433—437 (in Russian).

Kagan Y. S., Rodionov G. A., Kulagin O. M., Voronina L. Y., Velichko L. S., Peremitina A. D. Effect of DDT on functional state and liver structure in experiment on animals. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1970, 69—82 (in Russian).

Kagan Y. S., Sasinovich L. M., Ovseenko G. I. Computer-aided application of correlation analysis to toxicity and cumulation indices to develop hygienic standards. Proc. 1st Symp. Use of Math. Methods for Eval. and Predict. Real Haz. Pesticide Accumul. in Env. and Organ. Kiev, 1971, 40—42 (in Russian).

Kagan Y. S., Sasinovich L. M., Ovseenko G. I. A tentative calculation method of hygienic pesticide specifications for foods based on correlation analysis of their toxicity and cumulation indices. Gigiena i sanitariya, 1972, 7, 44—45, (in Russian).

Kagan Y. S., Sasinovich L. M., Danilenko L. P. Comparative evaluation of the therapeutic effect of dipyroxime in laboratory animals poisoned by some organophosphorus compounds. Farmakologiya i toksikologiya, 1971, 3, 359—362 (in Russian).

Kagan Y. S., Shtabsky B. M. Molecular mechanisms of cumulative effect. Gigiena i sanitariya, 1974, 11, 69—72. (in Russian).

Kagan Y. S., Sasinovich L. M., Ovseenko G. I. Use of computer-aided correlation analysis of toxicity and cumulation indices for hygienic regulation of pesticides in the air of the working zone. Gigiena truda, 1972, 8, 21—25 (in Russian).

Kagan Y. S., Sasinovich L. M. Use of calculation methods to recommend tentative maximum allowable pesticide concentrations in the air of the working zone. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1973, 10, 31—40 (in Russian).

Kagan Y. S., Trakhtenberg I. M., Rodionov G. A., Lukaneva A. M., Voronina L. Y., Verich G. E. Impact of pesticides on the origin and development of some pathologic processes. Gigiena i sanitariya, Kiev, 1974, 3, 23—27 (in Russian).

Kagan Y. S., Sasinovich L. M., Ovseenko G. I. A differentiated approach to setting calculated TSELs for individual pesticide classes. Proc. 2 Nat. Symp. Kiev, October 12—14, 1976, 77—78 (in Russian).

Kagan Y. S., Ershova E. A., Mintzer O. P., Larionova E. P. Forecasting toxic properties and hygienic standards of organophosphorus pesticides from data inputs

on their quantum-chemical and physicochemical parameters. Proc. 2d Nat. Symp. Kiev, October 12–14, 1976, 83–86 (in Russian).

Kagan Y. S., Ershova E. A., Klisenko M. A., Popov T. A., Chmil V. D., Reisig I. S. Some questions concerning toxicodynamics and metabolism of new thioorganophosphorus acid derivatives. Novosibirsk, 1977, I, 61—87 (in Russian).

Kagan Y. S., Voronina V. M., Akkerman G. Phthalophos: impact on embryogenesis and metabolism in the organism of albino rats and their embryos. Gigiena i sanitariya, 1978, 9, 28–31, (in Russian).

Kagan Y. S., Katznelson B. A., Kurliandsky V. A. Some ways to increase the efficacy of research in hygienic standardization of harmful substances. Gigiena i sanitariya, 1979, 8–11 (in Russian).

Kaloyanova-Simeonova F. Pesticides: toxic effects and prophylaxis. Sofia, 1977, 308 (in Bulgarian).

Kaloyanova-Simeonova F. et Fournier. Les pesticides et l'homme (Etude generale de la toxicologie humaine des pesticides modernes). Lyon, 1971, 166.

Karpenko V. N., Voitenko G. A., Polchenko V. I. Role of pesticides in the development of circulatory disorders. Problemi gematologii i perelivaniya krovi. 1972, 17, 8, 44—47 (in Russian).

Katznelson B. A. Deposition, elimination and retention of dust in the lungs. Toksikologiya. Moscow, 1976, 7—24 (in Russian).

Kachalai D. P. Functional state of gastric-intestinal tract among toxic chemicals warehouse employees. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1968, 523—528 (in Russian).

Khalimov E. M. Modern perceptions of drug metabolism in the body. In: Abridged Course in Molecular Pharmacology. Moskow, 1975, 51–74 (in Russian).

Khovaeva L. A., Stenberg A.I. Immunologic state of the organism exposed to low doses of chlorophos and methylnitrophos. Gigiena i sanitariya, 1976, 1, 98—100 (in Russian).

Khodosova I. A. A biochemical perspective on carcinogenesis. Moscow, 1976, 208 (in Russian).

Khochachka P., Somero J. Strategy of biological adaptation. Moscow, 1977, 399 (Russian transl.).

Kiriushchenkov A. P. Impact of harmful factors on fetus. Moscow, 1978, 215 (in Russian).

Klisenko M. A., Yakhim V. S. Sevin: accumulation in and elimination from the warm-blooded organism. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1966 (in Russian).

Klisenko M. A., Voitenko G. A., Kiselyova N. I. Toxicologic and chemical investigation of the volatile compounds that form in a soil containing polychloropinene and mineral fertilizer. Gigiena truda i profzabolevaniya. 1977, 10, 32–35 (in Russian).

Knizhnikov V. A. Some general problems and possibilities inherent in hygienic standardization of carcinogens of physical and chemical nature. Gigiena i sanitariya, 1975, 3, 96–100 (in Russian).

Kokarovtseva M. G. Some aspects to the mechanism for toxic action of dichloroethane. In: Use Hygiene of Polymer Materials. Stankevich K. I. (Ed.). Kiev, 1976, 54—56 (in Russian).

Kokarovtseva M. G. Effect of acetylcysteine on some biochemical indices of hepatic and renal disorders in rats during acute intoxication by dichloroethane. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1977, 113—116 (in Russian).

Kokshareva N. V. Evaluation of therapeutic action of S-diethyl ethyl ether of β-brombenzthiomerioxylic acid in the intoxication by DDVP phosphoethane. Farmakologiya i toksikologiya. Kiev, 10, 1975, 129—132 (in Russian).

Kokshareva N. V. Effectiveness and some physiologic mechanisms of action of diethyxime in DDVP poisonings. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1977, 109–112 (in Russian).

Kokshareva N. V., Kagan Y. S., Sasinovich L. M., Krivenchuk V. E. Therapeutic effectiveness and physiologic mechanism of action of the new cholinesterase reactivator diethyxime. In: Distinctive Resuscitation Patterns for Acute Intoxications. Proc. Russ. Sci. Conf. Ivanovo, 1975, 146–148 (in Russian).

Kokshareva N. V., Kovtun S. D., Sasinovich L. M., Krivenchuk V. E. Effectiveness of a new reactivator from the group of thiohydroxylic esters. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1976, 124 (in Russian).

Kokshareva N. V., Kovtun S. D., Kagan Y. S., Miziukova I. G., Medvedev B. M. Effect of the new cholinesterase reactivator diethyxime on the central nervous system. Bull. Exp. Biol. Med., 1977, 1, 29–31 (in Russian).

Kokshareva N. V., Kovtun S. D., Djura D. Electrophysiologic analysis of the effect of cholinesterase reactivators on the peripheral nerve function and myoneural junction in the intoxication by dimethyldichlorvinylphosphate. Farmakologiya i toksikologiya, 1978, 13, 82–86 (in Russian).

Komarova L. I. Accumulation of organochlorine pesticides in human embryo and fetus tissues. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1970, 61–70 (in Russian).

Komarova L. I., Batueva L. N. Some data relating to circulatory diseases to the effect of organochlorine pesticides. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1973, 389–392 (in Russian).

Komarova L. I. Influence of organochlorine pesticides on development of circulatory diseases. Problemi genatologii i perelivaniya krovi, 1976, 21, 11, 46–50 (in Russian).

Komissarenko V. P. Inhibitors of the adrenal cortex function and their importance for endocrinology and oncology. In: New Data on Hormones and Their Mechanism of Action. Kiev, 1977, 12–27 (in Russian).

Kondratiev Y. A., Lebedeva K. V., Pyatnova Y. B. Outlooks for practical use of insect pheromones. J. Nat. Mendeleev Chem. Soc. 1978, XXIII, 2, 179–189 (in Russian).

Konstantinova T. K. Effect of the herbicide butyl ether 2,4,5-T on the maternal organism and fetus development in experiment. Gigiena i sanitariya, 1974, 8, 101–102 (in Russian).

Konchalovskaya N. M. Cardiovascular system exposed to occupational factors. Moscow, 1976 (in Russian).

Konchalovskaya N. M., Rashevskaya A. M., Manaenkova A. M. Changes in the cardiovascular system exposed to lead, carbon disulfide and benzene. Long-term effects of chemical factors on the cardiovascular system. Moscow, 1972, 10–36 (in Russian).

Korablyov M. Dithiocarbamic acid derivatives. Minsk, 1971, 152 (in Russian).

Korolkov A. A., Petlenko V. P. Philosophical problems in the theory of norm in biology and medicine. Moscow, 1977, 391 (in Russian).

Kosarev V. A. Effects of specific treatment by cholinesterase reactivators on clinical symptoms of acute intoxications by organophosphorus pesticides. In: Distinctive Resuscitation Patterns for Acute Intoxications. L., 1975, 149 (in Russian).

Kosarev V. A. Liver abnormalities in acute intoxications by organophosphorus pesticides. Trans. Sklifosovsky Res. Inst. for Emerg. Aid. 1976, **22**, 134–135 (in Russian).

Koshland D. E. Conformation changes at the active site during enzyme action. Fed Procs, 1964, **23**, 315–323.

Kovtun S. D., Kokshareva N. V. Impact of the new ChE reactivator LA-54 on spontaneous activity of the neuromuscular synapse in acute DDVP intoxications. Bull. Exp. Biol. and Med. 1974, **1**, 24–25 (in Russian).

Kravkov N. P. Fundamentals of pharmacology. Moscow-Leningrad, 1927 (in Russian).

Krasovsky G. N. General pattern of dependence of biological parameters in mammals on body weight and some practical aspects of its use. New Developments in Research Methods, Diagnosis, Treatment and Prevention of Major Diseases. Moscow, 1972, 27–29 (in Russian).

Krasovsky G. N. Some research results on the problem of man vs animal sensitivity to toxic substances. In: Organism and Environment. 1970, 1, 59–64 (in Russian).

Krasovsky G. N. Intoxication modeling and rationale for experimental data extrapolation from animals to man in the solution of hygienic standardization problems. Moscow, 1973 (in Russian).

Krasovsky G. N. Problem of experimental data extrapolation from animals to man. Proc. I. Summing-Up USSR–USA Symp. on Environmental Hygiene, December 9—14, 1974. Moscow, 1975, 60—72 (in Russian).

Krasovsky G. N., Korolyov A. A., Belyaeva N. G., Varshavskaya S. N., Kutakov K. V., Malikova R. T., Trakhtman M. B. On comparative man vs test animal sensitivity to chemical factors (acetophos) in experiment. Gigiena i sanitariya, 1969, I, 45—49 (in Russian).

Krasovsky G. N., Sobinyakova O. R. Comparative man vs animal sensitivity to various substances according to acute toxicity indices. Gigiena i sanitariya, 1970, 4, 29—34 (in Russian).

Krasovsky G. N., Shigan S. A., Vitvitskaya B. R., Arsenieva M. V. On evaluation methods of species-specific differences. In: Hygienic Science for Practice. Moscow, 1972, 64—68 (in Russian).

Krasovsky G. N., Avilova G. G. Species-, age- and sex-related sensitivity to toxins. J. Nat. Mendeleev Chem. Soc., 1974, XIX, 2, 159—162 (in Russian).

Krasovsky G. N., Shigan S. A., Egorova N. A. Current problems of communal toxicology in view of chemical pollution of water bodies. Pharmacology, chemotherapeutic remedies, toxicology and toxicologic problems. Moscow, 1974, 15—83 (in Russian).

Krivenchuk V. E., Petrunkin V. E. Thiohydroximic esters I.S-diethylamine esters of thiohydroximic acids and their derivatives. Himiko-farmazevticheskii zhurnal, 1973, **VII**, 3, 13 (in Russian).

Krieger L. Y., Clark C. R. Toxicology and applied pharmacology, 1976, 38, 2, 315—323.

Krivenchuk V. E., Bryzgailo L. I., Petrunkin V. E. Thiohydroximic esters. Constarts for the reactivation rate of serous cholinesterase inhibited by GD-42. Biohimiya, 39, 6, 1163—1167 (in Russian).

Krivenchuk V. E., Kokshareva N. V., Sasinovich L. M., Petrunkin V. E., Kagan Y. S. Thiohydroximic esters 2.S-diethylaminoethyl ether of pyridine-4-thiohydroximic acid and its physiologic activity. Himiko-farmazevticheskii zhurnal, 1975, II, 18—20 (in Russian).

Krivoglaz B. A. Clinical features and treatment of intoxications by poisonous chemicals. Leningrad, 1965, 221 (in Russian).

Krotov Y. A. Use of calculation methods to set tentative maximum momentary permissible concentrations of air pollutants. Gigiena i sanitariya, 1971, 12, 8 (in Russian).

Kriuchkova V. A. Work hygiene in mercaptophos production and agricultural use. In: Hygiene, Toxicology and Clinical Features of New Insectofungicides. Moscow, 1959, 93—98 (in Russian).

Kryzhanovskaya M. V. Current questions of research on chemical allergens as part of the problem of sanitary environmental protection. In: Pesticide Use Hygience, Toxicology and Clinical Features of Poisonings. Kiev, 1968, 147—160 (in Russian).

Kuzmenko N. M. Hygienic principles and approaches in studying long-term effects of exposure to chemical substances. In: Current Questions of Pesticide Use Hygiene for Different Climate-Geographic Zones. Yerevan, 1976, 84—87 (in Russian).

Kuzminskaya U. A. Biochemical characteristics of subcellular hepatic structures on exposure to pesticides. A contribution to the mechanism of action of organochlorine and carbamate pesticides. Kiev, 1975, 42 (in Russian).

Kuzminskaya U. A. Importance of biochemical research methods for evaluation of effects of chemical substances on the organism. In: Current Questions of Pesticide Use Hygiene for Different Climate-Geographic Zones. Yerevan, 1976, 79—84 (in Russian).

Kuzminskaya U. A., Novachik V., Klisenko M. A. Polychlorocamphene distribution in lipid tissue fractions. Gigiena i sanitariya, 1972, 12, 96—97 (in Russian).

Kuzminskaya U. A., Yakushko V. E., Bersan L. V. Influence of some pesticides on microsomal mixed-function oxidases' activity. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1976, 11, 1, 69–73 (in Russian).

Kuzminskaya U. A., Yakushko V. E., Svetly S. S. Activity of the detoxifying microsomal liver system on exposure to some combinations of organochlorine, organophosphate, and dithiocarbamate pesticides. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1977, 76—81 (in Russian).

Kulagin O. M. Vitamin therapy of organochlorine pesticide intoxications. Kiev, 1974 (in Russian).

Kundiev Y. I. A hygienic perspective on the problem of pesticide absorption via skin. In the book: Questions of Pesticide Hygiene and Toxicology. Moscow, 1970, 51—55 (in Russian).

Kundiev Y. I. Pesticide absorption through skin and prevention of intoxications. Kiev, 1975, 199 (in Russian).

Kundiev Y. I., Voitenko G. A., Khokholkova G. A., Alexandrova L. G. Influence of pesticide metabolism and routes of entry into the body on magnitude of cumulative action. Proc. Nat. Conf. Scient. Basics of Modern Methods for Hygienic Regulation of Environmental Chemicals. Moscow, 1972, 118–123 (in Russian).

Kundiev Y. I., Nikitin D. P., Erman M. I., Khokholkova G. A. New hygienic problems due to widescale introduction of agricultural chemicals. Gigiena i sanitariya, 1975, 12, 6—10 (in Russian).

Kundiev Y. I., Burkatskaya E. N. Questions of toxicology of the pesticidal derivatives of carbamic, thio- and dithiocarbamic acids. In: Guidelines of Research on the Development of Plant Protection and Weed Control Chemicals, Moscow, 1976, 19 (in Russian).

Kustov V. V., Tiunov L. A., Vasiliev G. A. Combined effects of industrial toxicants. Moscow, 1975, 256 (in Russian).

Kurganov B. I. Allosteric enzymes. Moscow, 1978, 278 (in Russian).

Kurinny A. I., Pilinskaya M. A. Pesticides as mutagenic environmental factor. Tsitologiya i genetika. Kiev, 8, 4, 342–374 (in Russian).

Kurinny A. I., Pilinskaya M. A. Investigation of pesticides as environmental mutagens. Kiev, 1976 (in Russian).

Kurliandsky B. A. On body adaptation to chemical exposures. Gigiena i sanitar.ya, 1973, 7, 95—97 (in Russian).

Kurliandsky B. A., Nevzorova N. I. Regulation of chemical blastomogenic agent concentrations in human environment. Toksikologiya, Moscow, 1979, 9, 88—109 (in Russian).

Kurchatov G. V., Miziukova I. G., Vasilieva Z. A., Petrunkin V. E. A study of thiol-compounds as possible detoxifying agents for 1,2-dichloroethane poisoning. In: Physiologically Active Substances. Kiev, 7, 101—104 (in Russian).

Lambert Y., Broden J. Rev. Can. Biol, 1976, **35**, 1, 33–41.

Larionov L. V., Grishko V. A. Contemporary problems of pharmacology. Moscow, 1963, 358 (in Russian).

Larssen K. S., Arnauder C., Ce Kapova E., Kjellberg M. Teratology, 1976, **14** 2, 171–183.

Levina E. N. Combined action of industrial toxic agents. A Manual of Work Hygiene. Moscow, 1963, 154—164 (in Russian).

Leibovich D. L. Evaluation of embryotropic effects from low doses of the organophosphorus toxic chemicals chlorophos, metaphos, and carbophos. Gigiena i sanitariya, 1973, 8, 21—24 (in Russian).

Leliukh V. M., Maliuk V. I. Origination of bronchial asthma by protracted DDT exposure. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1966, 330–334 (in Russian).

Liublina E. I. Correlations of physicochemical properties of hydrocarbons with their toxicity. Gigiena i sanitariya, 1969, 7, 20–25 (in Russian).

Liublina E. I. Distinctive correlation patterns between molecular weight and MACs of some classes of organic compounds as a function of their state of aggregation Gigiena truda, 1973, 4, 37—40 (in Russian).

Liublina E. I. Acute and chronic intoxications. Threshold toxic action. In: Fundamentals of General Industrial Toxicology. Leningrad, 1976, 48—63 (in Russian).

Liublina E. I., Rabotnikova L. V. Possibility of predicting toxicity of volatile organic compounds from physical constants. Gigiena i sanitariya, 1971, 8, 33—37 (in Russian).

Liublina E. I., Minkina N. A., Rylova M. L. Adaptation to industrial toxic agents as a phase in intoxication. Leningrad, 1971, 206 (in Russian).

Liublina E. I., Mikheev M. I. A way to validate the equations proposed for calculation of toxicity indices in regard of maximum allowable concentrations, safety factors, etc. Gigiena i sanitariya, 1974, 12, 77—80 (in Russian).

Liublina E. I., Mikheev M. I. Scientific bases of setting tentative safe exposure levels for man. J. Nat Mendeleev Chem. Soc., 1974, XIX, 2, 142—146 (in Russian).

Litvinov N. N. Hygienic standardization of chemical substances showing blastomogenic properties. Gigiena i sanitariya, 3, 1973, 95—103 (in Russian).

Loewe S. Ergebn. Physiol., 1927, **27**, 47—187.

Lohs K., Dedek N., Giebel W. Studies on the alkylating properties of organophosphorus pesticides in mammals in vivo 3rd Intern. Congr. Pest. Chemistry. Helsinky, 1974, Abstr. book, 328.

Lubianova I. P. Comparative effectiveness of succimer, unithiol, and D-penicillin in treating microsaturnism. Kiev, 1979, 28 (in Russian).

Lugansky N. I., Miziukova I. G., Lokantsev D. S. A profile on the mechanism for antidotal action of unithiol in arsenite poisonings. In the book: Thiol Compounds in Medicine. Kiev, 1957, 48—50 (in Russian).

Lugansky N. I., Rodionov P. V., Cherkes A. I. (ed.) Toxicology manual of toxic chemicals. Kiev, 1964, 463 (in Russian).

Lukaneva A. M. Effect of DDT as a low-intensity factor on coronary insufficiency in experiment. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1968, 731—738 (in Russian).

Lukaneva A. M. Impact of protracted lindane exposure on functional state of the myocardium and development of coronary insufficiency in experiment. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1973, 240—243 (in Russian).

Lukaneva A. M. Influence of low pesticide doses on development of atherosclerosis in experiment. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1976, II, I, 90—92 (in Russian).

Lukaneva A. M., Kokshareva N. V. Modification of rabbit ECG in intoxication by DDVP and treatment by the cholinesterase reactivator, 5-diethylaminoethyl ether hydrochloride of β-brombenzthiohydroximic acid. In: Farmakologiya i toksikologiya. Kiev, 1975, 10, 127—129 (in Russian).

Lutsoya H. I. Experimental inputs for hygienic standardization of residual components of some pesticides in view of their combined use for fruit plant treatment. 1966, 16 (in Russian).

Luzhnikov E. A. Principal pathologic syndromes of acute poisoning. In: Emergency Care in Acute Intoxications. Moscow, 1977, 27—58 (in Russian).

Luzhnikov E. A. General principles underlying diagnosis of acute intoxications. In: Emergency Care in Acute Intoxications. Moscow, 1977, 24—26 (in Russian).

Luzhnikov E. A. General principles of emergency care in cases of acute intoxication. In: Emergency Care in Acute Intoxications. Moscow, 1977, 59—75 (in Russian).

Luzhnikov E. A., Dagaev V. N., Firsov I. N. Fundamentals of resuscitology in cases of acute intoxication. Moscow, 1977 (in Russian).

Lykken L., Casida J. E. Metabolism of organic insecticide chemicals. J. Canad. Med. Ass., 1969, 100, 145—179.

Makovskaya E. I. Pathologic anatomy of intoxications by poisonous chemicals. Moscow, 1967, 348 (in Russian).

Makovskii V. N. Toxicologic-hygienic investigation of the dipyridyl herbicides diquat and paraquat. Vinnitsa, 1972, 23 (in Russian).

Matsumoto H., Koya J., Takeuchi T. J. Neuropath. exp. neurol., 1965, 24, 4, 563—574.

Maximov G. G. Primary response of the body as the basis for prediction of the poisoning hazard presented by industrial toxic agents in the environment. Proc. 1st Symp. Dec. 14—15, 1971. Use of Math. Meth. Eval. and Predict. Real. Haz. Pesticide accumul. in Env. and Org. Kiev, 1971, 76—81 (in Russian).

Markoryan D. S. Cytogenetic effect of some organochlorine insecticides on bone-marrow cell nuclei in mice. Moscow, 1966, 130—137 (in Russian).

Markoryan D. S. A study on mutagenic properties of organochlorine insecticides in primates and small test animals. In: Questions of Hygienic Standard-Setting in the Study of Long-Term Effects of Industrial Chemical Exposures. Moscow, 1972, 24—40 (in Russian).

Martson V. S. A contribution to the toxicologic profile of the new fungicide polycarbacin. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1968, 593—602 (in Russian).

Martson L. V., Voronina V. M. An experimental investigation of the impact on embryogenesis by several organophosphorus pesticides (dipterex and imidan). Proc. 1st Summing-Up USSR-USA Symp. Environ. Hygiene. Moscow, 1975, 168—173 (in Russian).

Mastriukova T. A., Kabachnik M. I. Some ways to rational synthesis of new organophosphorus insecto-acaricides. J. Nat. Mendeleev Chem. Soc., 1978, XXIII, 2, 160—170 (in Russian).

Martyshenko N. V., Mironets N. V., Shigan S. A. Use of calculation methods to develop hygienic standards for chemicals in water. Proc. 2nd Nat. Symp. Kiev, October 12—14, 1976, 87—95 (in Russian).

Medved L. I. Summary results and future objectives of research in hygienic and toxicologic evaluation of insectofungicides. In: Hygiene, Toxicology and Clinical Features of New Insectofungicides. Moscow, 1959, 5—22 (in Russian).

Medved L. I. Work hygiene in the use of organomercury fungicides. Moscow, 1961, 34 (in Russian).

Medved L. I. (Ed.) Hygiene of pesticide use, toxicology and clinical features of poisonings. Kiev, 1968, 6, 864 (in Russian).

Medved L. I. Summary results and prospective goals of research in pesticide hygiene and toxicology. In: Pesticide use hygiene, toxicology and clinical features of poisonings. Moscow, 1976, 11, 1, 13—17 (in Russian).

Medved L. I., Kagan Y. S. Some theoretical issues of pesticide toxicology. In: Issues of Industrial and Agricultural Toxicology. Kiev, 1964, 43—53 (in Russian).

Medved L. I., Kundiev Y. I., Kagan Y. S. On methods to study penetration of chemicals through unimpaired skin. Gigiena i sanitariya, 1964, 10, 71—75 (in Russian).

Medved L. I., Kagan Y. S., Spynu E. I. Pesticides and public health problems. J. Nat. Mendeleev Chem. Soc., 1968, 3, 263—271 (in Russian).

Medved L. I., Spynu E. I., Kagan Y. S. The method of conditioned reflexes in pesticide toxicology. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, Zdorovie, 1966, 4, 5—34 (in Russian).

Medved L. I., Kagan Y. S., Spynu E. I., Buryi V. S. Present requirements for medical investigation of pesticides to assure their safe use. J. Nat. Mendeleev Chem. Soc., 1978, XXIII, 123—130 (in Russian).

Melnikov N. N. Pesticides and environmental control. J. Nat. Mendeleev Chem. Soc., 1973, 5, 570—576 (in Russian).

Melnikov N. N. Pesticide chemistry and engineering. Moscow, 1975, 766 (in Russian).

Melnikov N. N. Pesticides and environmental control. J. Nat. Mendeleev Chem. Soc., 1978, XXIII, 208—215 (in Russian).

Melnikov N. N. Key trends in the discovery of new pesticides. J. Nat. Mendeleev Chem. Soc., 1978, XXIII, 136—143 (in Russian).

Melnikov N. N., Volkov A. I., Korotkova O. A. Pesticides and the Environment. Moscow, 1977, 240 (in Russian).

Menn I. I. Comparative aspects of pesticide metabolism in plants and animals. Environ. Health Perspective. 1978, 27, 113—124 (in Russian).

Methodologic Instructions for Hygiene Evaluation of New Pesticides. Kiev, 1969, 160 (in Russian).

Methods of Hygienic and Toxicologic Evaluation of Biological Effects from Pesticides. Shitskova A. P., Elizarova O. N., Zhidkova L. V. et al. (Ed.). Moscow, 1977, 199 (in Russian).

Miziukova I. G., Petrunkin V. E. An approach to the discovery of drugs for antidotal treatment of intoxications of chemical ethiology. Moscow, 1967, 153—158 (in Russian).

Miziukova I. G., Bakishev G. N. Specific treatment of acute intoxications by methyl bromide. Vrachebnoye delo, 1971, 7, 149—152 (in Russian).

Miziukova I. G., Petrunkin V. E. Discovery of antidotes to treat poisonings of chemical ethiology. Trans. VIIIth Hygien. Congr. Ukr.SSR, 1971, 238 (in Russian).

Miziukova I. G., Bakhishev G. N., Kurchatov G. V., Petrunkin V. E. Discovery of specific remedies to treat intoxications by haloid-containing hydrocarbons. Trans. IInd Pharmac. Cong. Ukr.SSR. Kiev, 1973, 153 (in Russian).

Miziukova I. G., Petrunkin V. E. Unithiol and mercaptide as antidotes for arsenite poisonings. Vrachebnoye delo, Kiev, 1974, 2, 126—129 (in Russian).

Miziukova I. G., Bakhishev G. N. A drug against fumigant intoxications. Arstr. Rep. Soviet particip. IIIrd Intern. Plant Protec. Congr. Moscow, 1975, 127—128 (in Russian).

Miziukova I. G., Petrunkin V. E., Bakhishev G. N., Kokarovtseva M. G., Kurchatov G. V., Vasilieva V. A. Thiol compounds as specific treatment drugs for intoxications by haloid-containing hydrocarbons of aliphatic series. Farmakologiya i toksikologiya. 1978, 13, 103—106 (in Russian).

Monoenkova A. M. Cardiovascular system in intoxications by industrial toxic agents. In the book: Effects of Occupational Factors on Cardiovascular System. Moscow, 1975, 46—99 (in Russian).

Nagorny N. A. Toxicity of the volatile products resulting from thermal oxidative destruction of anthracenetolueneformaldehyde resin and the fibrogenic products of its dust. Gigiena truda i professionalniye zabolevaniya. 1971, 11, 21—24 (in Russian).

Nemirovsky O. N. Industrial toxicology. In: Methodologic Instructions for Practical Studies in Work Hygiene. Leningrad, 1970, 102—127 (in Russian).

Nakatsugawa T., Morelli M. In the book: Insecticide Physiochemistry and Physiology. New York-London, 1976, 61—114.

Odintsov V. S. Biochemical basics of organophosphorus insecticide application. Kiev, 1972, 175 (in Russian).

Olefir A. I. The effect of pesticides on immunogenesis. Vrachebnoye delo. 1978, 5, 14—16 (in Russian).

Osetrov V. I. On combined toxic action of thiuram with hexachlorocyclohexane gamma-isomer and heptachlor. In: Hygiene and toxicology of new pesticides and clinical features of intoxications by them. Moscow, 1962, 396—401 (in Russian).

O'Brien R. D. Insecticides, action and metabolism. New York, 1967, 289.

O'Brien R. D. Toxic phosphorus esters: chemistry, metabolism and biological effects. New York and London, 1960.

Panshina T. N. Toxicology of the herbicides, haloid-substituted anilides of carbonic acids, and hygienic aspects of their agricultural use. Kiev, 1977, p. 49 (in Russian).

Parke D. V. The Biochemistry of Foreign Compounds. Oxford-London-New York-Paris, 1967.

Perelygin V. M., Bykovskaya T. K. Hygienic approaches to the standardization and limitation of harmful substances arrival into soil. Gigiena i sanitariya. 1978, 11, 22—26 (in Russian).

Petkov V. Medicine. Organism. Pharmacologic effect. Medicine and Physical Culture. Sofia, 1974, 351 (in Russian).

Pinigin M. A. Current problems of communal toxicology in view of chemical air pollution. Pharmacology. Toxicology. Toxicologic problems. Moscow, 1974, 83–139 (in Russian).

Pinigin M. A. Biological equivalence in the solution of methodological problems in imposing hygienic standards on atmospheric pollutants. Moscow, 1977, 44 (in Russian).

Pliss G. B., Zabezhinsky M. A. Carcinogenic properties of symmetric triazine derivatives. Voprosi onkologii, 1970, **16**, 1, 82—85 (in Russian).

Pliss G. B. The threshold nature of the blastomogenic effect and the hygienic standardization of blastomogens. In: Questions of Hygienic Standardization in the Study of Long-Term Effects of Industrial Chemical Exposures, 1972, 141—147 (in Russian).

Plokhinsky N. A. Biometry. Novosibirsk, 1961, 364 (in Russian).

Pokrovsky A. A. The enzyme mechanism for some intoxications. Uspekhi biol. himii, 1962, 4, 61–81 (in Russian).

Polikar A. Elements of cell physiology. Leningrad, 1977, 390 (in Russian).

Polchenko V. I. Pesticide poisonings from data of world literature. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1968, 6, 61–71 (in Russian).

Polchenko V. I. Domestic pesticide intoxications abroad. In: Pesticide use hygiene, toxicology and clinical features of poisonings. Kiev, 1969, 7, 470–562 (in Russian).

Polchenko V. I. Hygienic evaluation of contributory factors to pesticide toxicity persistence. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1973, 10, 22–28 (in Russian).

Polchenko V. I., Tkach L. I., Gumenny V. S., Sokolovsky N. V., Baida L. K. An analysis into distribution patterns of some diseases in areas of intensive pesticide use. In: Pesticide Use Hygiene for Different Climate-Geographic Zones. Yerevan, 1976, 175—177 (in Russian).

Polchenko V. I., Tkach L. I., Gumenny V. S., Sokolovsky N. V., Baida L. K. An epidemiologic perspective on the study of morbidity due to pesticide use in agriculture. Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1976, 11, 1, 125–128 (in Russian).

Popov T. A. Biological and mathematical modeling of intoxication. Kiev, 1977, 50 (in Russian).

Popov T. A., Evreev V. N., Kagan Y. S. Use of rat liver perfusion to investigate the mechanism for the therapeutic action of complex cobalt compounds in phosphamide intoxication. In: Current Questions of Pesticide Use Hygiene for Different Climate-Geographic Zones. Yerevan, 1976, 140—143 (in Russian).

Popov T. A., Kaskevich L. M. The possibilities to identify preclinical forms of toxic heparatitis by evaluation of the activity of liver mixed-function oxidases. Vrachebnoye delo, 1977, 3, 126–131 (in Russian).

Popov T. A., Kotova A. B., Antomonov Y. G. Mathematical modeling of individual liver functions in pesticide poisonings. In the book: Mathematical Methods for Research Planning, Data Analysis and Prediction in Hygiene. Kiev, 1977, 89—95 (in Russian).

Pravdin N. S. A guide in industrial toxicology. 1934, **1** (in Russian).

Pravdin N. S. A methodology in minor toxicology of industrial poisons. Moscow, 1947, 217 (in Russian).

Promonenkov V. K., Korotkova O. A. New trends in pyrethroid synthesis and application. In: J. Nat. Mendeleev Chem. Soc., 1978, XXIII, 170–179 (in Russian).

Prozorovsky V. B. Application of the least-square method for probit-analysis of lethality curves. Farmakologiya i toksikologiya, 1962, I, 115—120 (in Russian).

Prozorovsky V. B. Intoxication by organophosphorus compounds. In: Poisoning at the Juvenile Age. Leningrad, 1977, 155—166 (in Russian).

Prozorovsky V. B., Savateyev N. V. Non-anticholinesterase mechanisms of action of anticholinesterase drugs. Leningrad, 1976, 160 (in Russian).

(Quantitative Toxicology. Leningrad, 1973, 285. Golubev P. P., Liublina E. I., Tolokontsev N. A., Filov V. A. (Authors).

Rapoport I. A. In: Problems of Medical Genetics. Moscow, 1970, 249—266 (in Russian).

Rashevsky N. Some medical aspects of mathematical biology. Moscow, 1966, 244 (in Russian).

Rodionov G. A. A contribution to the experimental pathology of the heart and blood vessels, caused by pesticides. 2nd Pathologoanat. Congr. Ukr. SSR. 1976, Chernovtsy, 184—185 (in Russian).

Rodionov G. A., Voronina L. Y. Impact of chlorophos on the development and course of pathologic states in experiment. Vrachebnoye delo, 1973, 11, 54—58 (in Russian).

Roll D. P. Problem of extrapolation of toxicologic data from test animals to man. Gigiena i sanitariya, Moscow, 1973, 4, 73—77 (in Russian).

Roslavtseva S. A. Problem of resistence in arthropods and ways to overcome it. J. Nat. Mendeleev Chem. Soc., 1978, XXIII, 196—202 (in Russian).

Roslavtseva S. A., Popov P. V. Synergistic action of organophosphorus pesticide mixtures. Himiya v selskom khozyaistve, 1965, 9, 29—32 (in Russian).

Roshchin A. V., Sanotsky I. V. (ed.). Long-term effects of chemical factors on cardiovascular system. A review. Moscow, 1972 (in Russian).

Rozengart V. I. Cholinesterases: functional role and clinical importance. In: Problems of Medical Chemistry. Moscow, 1973, 66—106 (in Russian).

Rozengart V. I. Metabolism of organophosphorus insecticides in insects. Nalchik, 1977, 1, 47—61 (in Russian).

Rozengart V. I., Sherstobitov O. E. Selective toxicity of organophosphorus insecto-acaricides. Leningrad, 1978, 176 (in Russian).

Rubenchik B. L. Biochemistry of carcinogenesis. 1977, 192 (in Russian).

Ryazanova R. A. Impact of the poisonous chemicals ziram and zineb on generative function in test animals. Gigiena i sanitariya, 1967, 2, 26—30 (in Russian).

Ryazanova R. A., Gafurova T. V. Mutagenic effect of the zineb-ziram pesticide mixture on bone-marrow cells in mice. In: Data Inputs for Hygienic Evaluation of Pesticides and Polymers. Moscow, 1977, 33—36 (in Russian).

Rylova M. L. Methods to investigate the chemical impact of harmful environmental factors in experiment. Leningrad, 1964, 228 (in Russian).

Savitsky I. V., Balashov V. E., Svetly S. S., Slobodkin V. I. Combined action of pesticides: criteria and evaluation methods. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1966, 703—710 (in Russian).

Sadykov A. S., Rozengart E. V., Abduvakhabov A. A., Aslanov H. A. Cholinesterases: active center and mechanism of action. Tashkent, 1976 (in Russian).

Sanotsky I. V. (ed.). Methods to determine toxicity and hazard of chemicals (Toxicometry). Moscow, 1970, 343 (in Russian).

Sanotsky I. V. Gonadotropic action of chemical compounds (questions of methodology and methods). In: Pharmacology, Chemotherapeutic Drugs, Toxicology. In: Problems of Toxicology, Moscow, 1971, 107—130 (in Russian).

Sanotsky I. V. Basic trends of preventive toxicology in the year 1970. In: Pharmacology, Chemotherapeutic Drugs, Toxicology. Problems of Toxicology, Moscow, 1971, 17—26 (in Russian).

Sanotsky I. V. Major questions concerning the problem of long-term effects of occupational toxic exposures. In: Questions of Hygienic Standardization in the Study of Long-Term Effects of Industrial Chemical Exposures. Moscow, 1972, 5—13 (in Russian).

Sanotsky I. V. Summary results and prospects of research on physiologic adaptation and temporary compensation of the chronic pathologic process in response to minimum chemical exposures. In: Pharmacology, Chemotherapeutic Drugs, Toxicology. Moscow, 1973, 41—48 (in Russian).

Sanotsky I. V., Grodetskaya N. S., Bazarova A. A., Sheveleva G. A. Methods of experimental investigation of the cardiovascular system exposed to chemicals used

in industry. In: Long-Term Effects of Chemical Factors on Cardiovascular System. Moscow, 1972, 67—88 (in Russian).

Sanotsky I. V., Fomenko V. N., Zaeva G. N., Grodetskaya N. S. The problem of long-term effects from industrial toxic agents. J. Nat. Mendeleev Chem. Soc., 1974, XIX, 2, 146—159 (in Russian).

Sanotsky I. V., Ulanova I. P. Criteria of harmfulness in hygiene and toxicology for hazard evaluation of chemical substances. Moscow, 1975, 328 (in Russian).

Sanotsky I. V., Sidorov K. K. Current views at safety factor for the validation of MAC values for the harmful substances present in environmental media. Gigiena i sanitariya, 1975, 12, 93 (in Russian).

Saralmanova Z. S. Effects of some toxic agents in the sexual sphere. Tashkent, 1971, 23 (in Russian).

Sasinovich L. M., Evreev V. N., Golub V. A., Petrunkin V. E., Kagan Y. S. Polynucleus compounds of transition metals with aminoalcohols and their therapeutic properties in acute poisonings by organophosphorus pesticides. In: Current Questions of Pesticide Use Hygiene for Different Climate-Geographic Zones. 1976, 136—140 (in Russian).

Seifulla R. D. Question of theory bearing on pharmacologic reception. In: Abridged Course in Molecular Pharmacology. Moscow, 1975, 77—143 (in Russian).

Semencheva E. M. On the mechanism of action of DDT and some pesticides on immunohematologic indices of the organism. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1968, 805—815 (in Russian).

Semencheva E. M., Gorbachevskaya E. F., Komarova L. I. The pattern of the autoimmune process in adults and newborns with manifested and latent DDT intoxication of the maternal organism. In: Pesticide Use Hygiene for Different Climate-Geographic Zones. Yerevan, 1976, 105—108 (in Russian).

Semencheva E. M. Changes of hemoglobin synthesis accompanied by the autoallergic process in cases of hypochromic anemia caused by propazine. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1969, 172—182 (in Russian).

Semencheva E. M., Gorbachevskaya E. F. Antibody activity in the blood and milk of rabbit females on exposure to HCCH — isomer, relative to tissue antigenes in newborns. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1976, 122 (in Russian).

Sergeev P. V. Molecular pharmacology (Its Origins and future prospects). In: Abridged Course in Molecular Pharmacology. Moscow, 1975, 7—50 (in Russian).

Shabad L. M. Endogenous blastomogenic substances. Moscow, 1969 (in Russian).

Shabad L. M. Methods for determination and investigation of blastomogenicity of chemical substances. Moscow, 1978 (in Russian).

Shabad L. M. Some observations on organ cultures subject to transplacental action of blastomogenic substances. In: Questions of Hygienic Standardization in the Study of Individual Effects of Industrial Toxic Exposures. Moscow, 1972, 123—130 (in Russian).

Shabad L. M. Blastomogenic hazard of human habitat pollutants. Toksikologiya. Moscow, 1978, 9, 7—59 (in Russian).

Shabad L. M., Sanotsky I. V., Zaeva G. N., Bruevich T. S., Katznelson B. A., Yanysheva N. Y., Shugaev B. B. The possibility of establishing maximum allowable concentrations for benz(a) pyrene in the air of work rooms. Gigiena i sanitariya, 1973, 4, 78—80 (in Russian).

Shabad L. M., Ilnitsky A. P. Summing-up the discussion on hygienic standardization of carcinogenic substances. Gigiena i sanitariya, 1976, 5, 91—93 (in Russian).

Shimanko I. I. Nephropathy in acute exogenous poisonings, its conservative treatment and surgery. Moscow, 1972 (in Russian).

Shitzkova A. P. Some aspects to the evaluation of specific effects of pesticides on the organism. Gigiena i sanitariya, 1974, 8, 76—78 (in Russian).

Shitzkova A. P., Ryazanova R. A. Pesticide toxicology and hygiene. Moscow, 1975, 190 (in Russian).

Shitzkova A. P., Elizarova O. N., Zhidkova L. V. et al. Methods of hygienic and toxicologic evaluation of biological pesticide effects. Moscow, 1977, 199 (in Russian).

Shtabsky B. M. On the cumulation coefficient and the zone of cumulative action of chemicals. Gigiena i sanitariya, 1971, 4, 91–93 (in Russian).

Shtabsky B. M. On correlation between the phenomena of material and functional cumulation. Proc. 1st Symp. Use Math. Meth. for Eval. and Predic. Real Haz. Pesticide Accumul. in Environ. and Organism. Kiev, 1971, 43–47 (in Russian).

Shtabsky B. M. Quantitative evaluation of cumulation phenomena. Gigiena i sanitariya, 1973, 8, 24–28 (in Russian).

Shtabsky B. M., Kagan Y. S. Evaluation of cumulative properties of chemicals according to the index and standardized coefficient of cumulation. Gigiena i sanitariya, 1974, 65–67 (in Russian).

Shtabsky B. M. Methodologic bases of cumulation studies as part of a toxicologic-hygienic research. Lvov, 1975, 30 (in Russian).

Sidorenko E. N. Pesticide poisoning. Kiev, 1978, 128 (in Russian).

Sidorenko G. I. Current problems of environmental hygiene. Gigiena i sanitariya, 1978, 10, 9—15 (in Russian).

Sidorenko G. I., Gofmekler V. A., Khuriev B. B. A study of the impact by toxic agents on the processes of pregnancy in women. (Summary). Gigiena i sanitariya, 1971, 5, 117 (in Russian).

Sidorenko G. I., Krasovsky G. N., Pinigin M. A. Some trends in deriving hygienic standards for carcinogenic substances in the environment. Gigiena i sanitariya, 1974, 12, 75–77 (in Russian).

Sidorenko G. I., Pinigin M. A. "Concentration-time" relationship in different inhalation regimes of some organic substances. Proc. 1st Summing-Up USSR-USA Symp. Environ. Hygiene. December 9–14, 1974. Moscow, 1975, 15–21 (in Russian).

Sidorenko G. I., Pinigin M. A. Hygienic criteria for the complex action of chemical environmental pollutants. Gigiena i sanitariya, 1976, 6, 77—80 (in Russian).

Sidorov K. K. Some methods for quantitative evaluation of the cumulative effect. In: Toxicology of New Industrial Toxic Agents. Leningrad, 1967, 9, 19–27 (in Russian).

Sidorov K. K. Calculation of safety factor to validate MAC values for harmful substances in the air of the working area. Gigiena i sanitariya, 1971, 6, 54–57 (in Russian).

Sidorov K. K. Evaluation of cumulative effects from chemical substances for different modes of inhalational exposure. Gigiena i sanitariya, 1972, 5, 93—95 (in Russian).

Sidorov K. K., Shapiro A. B. A method to calculate TSELs of chemical compounds for different modes of inhalational exposure. Gigiena i sanitariya, 1972, 5, 93–95 (in Russian).

Sidorov K. K., Shapiro A. B. Validation of tentative safe levels of exposure to the chemical substances present in the air of the working area. Vrachebnoye delo, 1977, 3, 131—133 (in Russian).

Slobodkin V. I. Toxicologic-hygienic profiles on some mixtures of organophosphorus pesticides with a synergist. Kiev, 1975, 24 (in Russian).

Sós J. In the book: Pathogenesis of Civilization Diseases. Budapest, 1976, 9–39 (in Russian).

Spynu E. I. Toxicology of the organochlorine pesticides of diene synthesis and work hygiene during their use. Kiev, 1975 (in Russian).

Spynu E. I., Sova R. E., Bolotny A. V. Complex action of organophosphorus pesticides. Gigiena i sanitariya, 1978, 12, 15—17 (in Russian).

Stepansky G. A. Human adaptation to modern chemical environment. In: Problems of Toxicology. Adaptation and compensation during chemical exposures. Moscow, 1973, 5, 5–41 (in Russian).

Stenberg A. I. (ed.) Residual pesticide concentrations in food. Moscow, 1973, 200 (in Russian).

Stenberg A. I., Orlova N. V., Torchinsky A. M. The impact of pesticides with different chemical structures on gonads and embryogenesis in experimental animals. Gigiena i sanitariya, 1973, 8, 16—20 (in Russian).

Stenberg A. I., Torchinsky A. M. Some aspects in hygienic investigation of teratogenic effects of substances foreign to the organism that are contained in food. Gigiena i sanitariya, 1974, 37 (in Russian).

Svetly S. S. Comparative data on hydrogen sulfide concentrations in the air inhaled by laboratory animals after TMTD and fenthiuram administration to them. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1969, 299—303 (in Russian).

Svetly S. S. Toxicologic-hygienic investigation of the combined fungicide fenthiuram. Kiev, 1970, 21 (in Russian).

Svetly S. S. Some distinctive patterns of conversion in the body of the dithiocarbamates that form part of combined pesticides. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1976, 92—95 (in Russian).

Svetly S. S. Principal approaches to and current questions of studies into the combined action of pesticides. In: Current Questions of Pesticide Use Hygiene for Different Climato-Geographic Zones. Yerevan, 1976, 89—92 (in Russian).

Terekhov N. T., Tarakhovsky M. L. Pathogenetic and symptomatic therapy of acute intoxications. In: Treatment of Acute Poisonings. Kiev, 1973, 29—51 (in Russian).

Tiunov L. A. Enzymes and poisons. In: Questions of General Industrial Toxicology. Leningrad, 1963, 80—85 (in Russian).

Tolokontsev N. A. Relationship between the amount of toxic exposure and its effect. In: Golubev A. A., Liublina E. I., Filov V. A., Tolokontsev N. A. Quantitative Toxicology. Leningrad, 1973, 23—46 (in Russian).

Tolokontsev N. A., Filov V. A. (Ed.) Fundamentals of General Industrial Toxicology. Leningrad, 1976, 303 (in Russian).

Tolokontsev N. A., Rusin V. Y. A toxic effect from combined action of chemical and physical factors in working environment. In: Fundamentals of General Industrial Toxicology. Leningrad, 1976, 124—126 (in Russian).

Trakhtenberg I. M. (Ed.) Toxicologic evaluation of volatile substances released from synthetic materials. Kiev, 1968, 195 (in Russian).

Trakhtenberg I. M. Chronic mercury exposure and its effect on the organism (a current perspective on the problem of mercurialism and its prevention). Kiev, 1969, 392 (in Russian).

Trakhtenberg I. M. Problems of the cardiovascular diseases of chemical origin. Moscow, 1978, 51—78 (in Russian).

Trakhtenberg I. M., Tychinin V. A. General biological concepts concerning adaptation and compensation in modern preventive toxicology. Gigiena i sanitariya, 1977, 1, 40—41 (in Russian).

Trakhtenberg I. M., Sova R. E., Sheftel V. O., Onikienko F. A. Normal indices of normalcy in laboratory animals in toxicologic experiment. Moscow, 1978, 175 (in Russian).

Tsybulkin E. K., Markova I. V. Diagnosis and treatment of acute intoxications. In: Poisonings at the Juvenile Age. Moscow, 1977, 19—47 (in Russian).

Turkevich N. M., Samudzhaya E. M. Hormones and carcinogenesis. Kiev, 1975, 168 (in Russian).

Turusov V. S. Voprosi onkologii. Leningrad, 1975, XXI, 12, 127 (in Russian).

Turusov V. S. Methods of identifying the blastomogenic effect of chemical agents. Toksikologiya. Moscow, 1978, 9, 59—88 (in Russian).

Ulanova I. P. Dependence of blastomogenic effect on chemical structure for different classes of haloid-containing hydrocarbons. (A contribution to hazard evaluation of industrial toxic agents). Moscow, 1971, p. 53 (in Russian).

Ulanova I. P., Avilova G. G., Bazarova L. A., Maltseva N. M., Migukina N. V., Khalepo A. I., Eitingon A. I. Experimental profiles on adaptation to poisons under their different modes of action. In: Pharmacology. Chemotherapeutic Drugs Toxicology. Moscow, 1973, 64—75 (in Russian).

Ulanova I. P., Pinigin M. A. Toxicity and hazard evaluation of chemical substances for hygienic standardization of their concentrations in environmental media. J. Nat. Mendeleev Chem. Soc., 1974, XIX, 2, 130—135 (in Russian).

Varshavskaya S. P. Present concepts concerning long-term effects of chemical environmental pollutants. A scientific review. In: Long-Term Effects from Biological Action of Some Chemical Environmental Pollutants. 1975, 8—15 (in Russian).

Van der Waerden B. L. Arch. exp. Pathol. & Pharmac., 1940, 195, 379.

Varshavskaya S. P., Dinerman A. A. Methodologic approaches to studying long-term effects of chemical environmental pollutants. A scientific review. In: Long-Term Effects from Biological Action of Some Chemical Environmental Pollutants. 1975, 17—32 (in Russian).

Vashakidze V. I. A contribution to the study of long-term effects of pesticides. In: Current Questions of Pesticide Use Hygiene for Different Climate-Geographic Zones. Yerevan, 1976, 87—89 (in Russian).

Vievskaya G. A. The pattern of psychic disorders associated with identified carriage of organochlorine pesticides in chronic alcoholics who are organochlorine-compound carriers, and with chemical allergoses. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1973, 373—375 (in Russian).

Vievskaya G. A. About neural-psychic conditions in persons subject to protracted occupational exposure to organophosphorus pesticides. Vrachebnoye delo, 1973, 136—138 (in Russian).

Voitenko G. A. Toxicologic and hygienic characteristics of the new insecticide chlorten. Gigiena truda i professionalnye zabolevaniya, 1957, 4, 51—53 (in Russian).

Voitenko G. A. Some questions of toxicodynamics in separate and combined exposure to tetramethylthiuramdisulfide, gamma-isomer of hexachlorcyclohexane and heptachlor. In the book: Industrial Toxicology and Clinical Features of Occupational Diseases of Chemical Etiology. Moscow, 1962, 197—200 (in Russian).

Voitenko G. A. Distinctive patterns of lindane toxicity by its complex (through the respiratory tracts and stomach) entry into the organism of albino rats. Gigiena i sanitariya, 1978, 3, 41 (in Russian).

Voitenko G. A., Larionov V. G. A study on phosphamide toxicity by simultaneous entry into the body through the gastrointestinal tract and respiratory organs. Farmakologiya i toksikologiya, 1976, 5, 622 (in Russian).

Voitenko G. A., Ershova E. A., Voronina V. M. et al. Experimental study into the toxic properties of new organophosphorus pesticides by the inhalational exposure.

Voronina V. M. Experimental data regarding the embryotoxic action of phthalophos. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Kiev, 1971, **9**, 254—257 (in Russian).

Voronina V. M., Pismennaya M. V. Distribution of phthalophos and its metabolites in rat organs and fetus. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1973, 10, 280—282 (in Russian).

Voronina V. M. Embryotoxic effect of phthalophos in rabbits. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1977, 119—122 (in Russian).

Vorotyntsev A. A., Luzhnikov E. A., Yaroslavsky A. A., Skachkov V. B., Barsukov Y. F., Kotov N. N. Comparative description of methods for rapid chlorophos elimination in experiment. Gigiena i sanitariya, 1977, 8, 76—78 (in Russian).

WHO Proceeds. 1975. Classification of pesticides.

Wilkinson C. F. (Ed.) Insecticide biochemistry and physiology. Plenum Press. New York — London. 1976, 753.

Zaeva G. N., Kulagina N. K. Rapid methods for deriving maximum allowable concentrations of harmful substances in the air of the working zone. Moscow, 1970, 24—33 (in Russian).

Zaeva G. N., Timofievskaya L. A., Fedorova V. I., Bruevich T. S., Shugeev B. B., Shumskaya N. I. Hygienic standardization of blastomogens, a current problem of industrial toxicology. In: Questions of Hygienic Standardization in the Study of Long-Term Effects of Industrial Chemical Exposure. Moscow, 1972, 130—141 (in Russian).

Zaugolnikov S. D., Loit A. O., Ivanitsky A. M. A contribution to classification of toxic substances. In: General Questions of Industrial Toxicology. Moscow, 1967, 46—49 (in Russian).

Zaugolnikov S. D., Loit A. O., Ivanitsky A. M. Toxicologic-Hygienic Classification of Harmful Substances. In: Allowable Concentrations of Harmful Substances in the Air of Work Rooms: Methods and Principles. Moscow, 1970, 76—83 (in Russian).

Zaugolnikov S. D., Kochanov M. M., Loit A. O., Stavchansky I. I. Rapid methods of determining toxicity and MACs and of hazard evaluation of chemical substances in the environment. Leningrad, 1975, 3, 75—83 (in Russian).

Zaugolnikov S. D., Kochanov M. M., Loit A. O., Stavchansky I. I. Rapid methods for determining toxicity and hazard of chemical substances. Moscow, 1978, 284 (in Russian).

Zilber Y. D. Neurotoxic action of poisons. In: Current Questions of Industrial Toxicology. 1970, 129—145 (in Russian).

Zlatev Z. D. Combined effect of some insectoacaricides on warm-blooded organisms. Kiev, 1976, 20 (in Russian).

Zlatev Z. D. Toxicity and combined effect of some mixtures of organophosphorus, organochlorine and dinitrophenol acaricides. In: Pesticide Use Hygiene, Toxicology and Clinical Features of Poisonings. Moscow, 1976, 11, 1, 118—120 (in Russian).

Zlatev Z. D., Sova R. E., Yakushko V. E. Orthogonal design of experiment to study combined pesticide exposure. Current Questions of Pesticide Use Hygiene for Different Climate-Geographic Zones. Yerevan, 1976, 96—97 (in Russian).

Zlatev Z. D., Kagan Y. S., Sova R. E., Svetly S. S. Methodologic recommendations for planning an experiment and estimating the effect of combined multiple exposure to chemical substances. Kiev, 1977, 14 (in Russian).

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