Table II. R_f Values of Heme Compounds Derived from Irradiated and Nonirradiated Myoglobin Preparations

Sample Treatment	R_f Values[a] Experiment 1	Experiment 2
Nonirradiated meat extract	0.77	0.65
Hemin chloride	0.80	0.66
Irradiated meat extract	0.82	0.70
	0.62	0.47
Nonirradiated meat extract plus irradiated meat extract	0.79	0.70
	0.60	0.47
Hemin chloride plus irradiated meat extract	0.84	0.74
	0.64	0.53

[a] Two duplicate series of experiments. Separation of altered heme porphyrin was achieved in both experiments.

the chromatogram under a Mineralite lamp of 3600 A. wave length.

Results

It is apparent from Table I that the method outlined is successful for the production and detection of the altered heme compound in the irradiated extracts. The R_f values presented in Table II show that two heme compounds can be detected in the irradiated extracts, and the presence of the altered heme compound can be demonstrated in the presence of added nonirradiated control samples or hemin chloride. One of the compounds has an R_f corresponding to protoporphyrin IX derived from unreacted myoglobin and the second compound has a lower R_f. On the basis of earlier studies with this chromatographic system (*1*, *4*), the new heme compound (derived from the green pigment) probably has four carboxyl groups, as compared to two for unchanged hemin.

These techniques have permitted the detection and separation of the altered heme compound arising from myoglobin during irradiation. Studies to produce larger quantities of the isolated heme compounds for further chemical characterization are now in progress. The exact quantity of green pigment produced cannot be precisely calculated, until the compound has been isolated and the extinction coefficient determined. On the basis of the quantity of unaltered myoglobin present in the irradiated extracts as compared to nonirradiated extracts, the maximal quantity of green pigment produced would approximate 20% of the total pigment concentration.

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PESTICIDES LITERATURE

A Multi-Indexed Machine-Sorted, Punch Card System for Pesticide Metabolism Data

G. CONGDON WOOD and ISAAC D. WELT

Chemical-Biological Coordination Center, National Academy of Sciences-National Research Council, Washington 6, D. C.

IBM punch card methods have been perfected by the Chemical-Biological Coordination Center for coding information concerning the biochemical transformations undergone by pesticides in the course of their metabolism. A code based upon a comprehensive classification of enzymes permits the recording of the effects of pesticides upon these all-important biochemical catalysts. This information can be retrieved by searching for the type of reaction, the organ, the species, the names of the pesticides, and their products.

RESEARCH IN THE PESTICIDE FIELD during the past decade has resulted in the accumulation of an enormous body of information, and there is no indication that activity in this area of chemical and biological endeavor will level off in the near future. With the ever-increasing number of publications in this field, the investigator is faced with the almost insoluble problem of keeping up with the literature.

The situation is aggravated by the fact that pesticide research cuts across traditional lines of academic disciplines with abandon. It is not unusual to find pertinent information concerning a chemical compound, of interest as a pesticide, in journals devoted to organic chemistry, entomology, biochemistry, pharmacology and toxicology, industrial medicine, and public health. For example, the *Biochemical Journal*, for July 1955, contains two excellent papers on insecticides (*1*, *7*), and the December

1954 issue of the *British Journal of Pharmacology and Chemotherapy* contains a paper on the effects of anticholinesterases, almost all of which are widely used as pesticides (*4*).

The Chemical-Biological Coordination Center of the National Academy of Sciences-National Research Council, as an outgrowth of the Insect Control Committee of the wartime Office of Scientific Research and Development, has been attempting to solve the difficult problems of literature searching and render useful information to the pesticide chemist or biologist.

In dealing with the enormous body of information available in the literature and in unpublished reports concerning the effects of chemical compounds upon biological systems, the center has had recourse to punch card techniques. A code for the recording of chemical structures on IBM punch cards was developed and used successfully for the coding of some 63,000 compounds on the basis of functional groups, ring systems, and so forth (*2*). These compounds include a number of pesticides and potential pesticides. On the biology side, a comprehensive coding system designed to cover the vast field of biology in all its extensive ramifications has been developed (*3*).

It has been a difficult, but a most rewarding task to coordinate such a large variety of scientific disciplines into a single unit. This biology code, by virtue of its all-encompassing nature, makes it possible to disregard the artificial boundaries between various fields of biological science, and to collect, abstract, and code information on the effects of chemical compounds on biological systems without much concern as to its source among the many subdivisions of biology. The effects of DDT upon such dissimilar organisms as the mosquito and the laboratory rat, as well as its metabolism in both of the species, can be incorporated into this punch card file and made readily available, no matter in which journals the information was published. Thus, as a result of the use of punch cards, specific information can easily be retrieved, regardless of its original sources. Abstracts of the information sought are available on these code sheets, which also cite the original literature references.

The organization, techniques, and objectives of the Chemical-Biological Coordination Center are described in a booklet, which is available to all interested persons (*6*). Several other detailed pamphlets concerning the center's activities may be obtained on request (*8–13*).

The staff members, who are all qualified scientists and represent a good cross section of the biological sciences, read the literature in their own fields carefully and assign codable data to qualified abstractors and coders. A special type of abstract, suitable for coding, is prepared on a code sheet form. The information is broken down into coding fields, such as taxonomy, organ, host, specific effect, general effect, and dose level. After the abstracted and coded data have been rigorously checked, punch cards are prepared. These, as well as the code sheet abstracts, are filed and provide the raw material for the retrieval of information concerning the effects of chemicals on biological systems.

In the past few years, as pesticide research has emerged from the strictly empirical stage, and publications have become available on the effects of these compounds on enzyme systems in a variety of organisms, as well as on their metabolic transformations in these organisms, it has become necessary to provide techniques for the recording of such information in our punch card system.

Two years ago, a new enzyme code was organized, utilizing a four-column field. The classification of enzymes is a difficult task, especially since enzymologists tend to disagree among themselves. However, several hundred enzymes have been classified. Table I illustrates a small part of this scheme.

Table II illustrates a portion of the alphabetical listing of enzymes and is designed to be used by the coder.

As a result of this classification, it is possible to obtain information on the effects of a compound on an enzyme, irrespective of its source—the source is, of course, indicated in the taxonomy section of the punch card and code sheet. It is also possible to pull out one enzyme, such as true cholinesterase; a family of enzymes, such as the cholinesterases; or a major class, such as the esterases.

If a compound inhibits a particular enzyme in a particular species only, it is a good lead in the search for a selective pesticide, as well as an indication of its posssible mechanism of action. The toxic effects of pesticides in mammals are well represented in the files, and papers dealing with the reasons for these toxicological manifestations often yield data concerning effects on enzymes or enzyme systems.

The fundamental unity of all life is perhaps best illustrated in the ubiquitous occurrence of enzymes and the many parallel observations recorded by both plant and animal biochemists, entomologists, and pharmacologists.

In order to handle data concerned with the many and varied complex alterations undergone by pesticides in the course of their metabolism, a classified list of known biochemical transformations has been developed and assigned code symbols. Table III illustrates a small portion of this list.

Table I. Enzyme Classification

[CBCC enzyme code (in part)]

Kinases
Mutases
Oxidases
A. Iron enzymes
B. Copper enzymes
C. Others
Dehydrogenases
A. Flavoprotein enzymes
B. TPN-linked enzymes
C. DPN-linked enzymes
D. Miscellaneous

Table II. Alphabetical Listing of Enzymes

(Excerpts from CBCC enzyme code)

Enzyme	Code
Choline phosphokinase	784
Cholinesterases	7S4
Cholinesterase, true	7S41
Cholinesterase, pseudo	7S42
Chondrosulfatase	7S6

As a result of the use of these symbols, it is possible to compare the biochemical techniques employed by organisms for the metabolic alteration of pesticides, and often to discover common precursors and degradation products of a substance. Lack of suitable metabolic pathways in some organisms—i.e., the inability of the Dalmation coach hound to degrade uric acid—is of interest both theoretically and practically. Demonstration of this phenomenon may lead to the discovery of new compounds suitable as pesticides for some species, yet innocuous to others. This concept of selective toxicity is amply illustrated among the center's files. The correlation of metabolic alterations of a particular compound occurring in plants, insects, microorganisms, and animals is thus greatly facilitated by the coding procedures.

An excellent up-to-date example of the kind of information which can be handled is furnished by Casida (*1*). He studied the effects of acetyl cholinesterase enzymes derived from 15 insect species representing eight distinct insect orders,

Table III. Classification and Coding of Specific Biochemical Reactions

(Excerpts from CBCC biology code)

Conjugation (undefined)	FE4
With glucuronic acid	FE41
With glycine	FE42
With sulfate	FE43
Formation of mercapturic acid	FE44
With glutamine	FE45
Acylation	FE5
Acetylation	FE51

upon the compounds acetylcholine, benzoylcholine, acetyl-β-methylcholine, triacetin, and *o*-nitrophenyl acetate, as well as the effects of these enzymes from a variety of organs of *Periplaneta americana*, the American cockroach. Inhibitors such as choline, eserine, and tetraethylpyrophosphate were also used in order to clarify enzymatic mechanisms.

This wealth of information is readily abstracted and coded—the task being greatly facilitated by the excellent tables in the paper. As a result, the information may be retrieved and made available in a variety of ways.

1. Information concerning the metabolism of each of the five compounds in each of the insect species and in the organs of the cockroach.
2. Enzyme activities of acetylcholinesterases in each of the insect species and in the organs of the cockroach.
3. Effects of each of the three acetylcholinesterase inhibitors upon each of the insects in the paper.

These data, correlated with the effects of the same compounds in other insect species in the same or in different orders, upon the mammalian nervous or cardiovascular systems, yield a well-rounded picture of the effects of the chemicals upon biological systems as a whole and provide insight into their mechanism or mechanisms of action. Information which may be present in an author's scientific paper but not necessarily stressed as evidence supporting his major thesis, becomes available as correlative data along with pertinent facts gleaned from numerous other published or unpublished contributions.

Smith (7) studied the metabolism of nine phenols in locusts (*Locusta migratoria*, *Schistocerca gregaria*, and *Nomadacris septemfasciata*) and isolated and chemically characterized their metabolites. In addition, the excretion of ethereal sulfates and glucosides by 15 other insect species after the administration of *m*-aminophenol, 8-quinolinol, and 7-hydroxycoumarin was determined and conclusions were drawn concerning the extent of conjugation of these compounds with one or both of the conjugating agents. As a result, the phenyl sulfuric acids were found to be less toxic to locusts than the phenols. By learning how each species handles a group of representative chemical compounds, the search for better, more effective insecticides will be greatly facilitated. The emergence of resistant strains can be explained on a biochemical basis only when the metabolic systems employed by such species and strains have been unraveled and the information collected by many experimenters working in diverse fields and with diverse experimental approaches and procedures is reduced to a standardized form and disseminated more widely among investigators.

Many entomologists may not read the *Biochemical Journal*, and would therefore benefit from a service which makes such information readily available to them. The bringing together of such research data, obtained by the entomologist, the biochemist, or the pharmacologist, and published in the journals devoted to their respective fields, is one of the most important tasks of the Chemical-Biological Coordination Center.

These coding techniques also make it feasible to answer certain questions and to furnish useful information which would otherwise not be available.

Consider, for example, the search for a hypothetical insecticide of the organophosphorus type which must have an LD_{50} of the order of 10 γ per gram for locusts but an oral LD_{50} in humans not exceeding 200 mg. per kg. If such a compound were to exist and had been added to the files, the answer to this question could be made available as a result of machine searching. Existing abstracting and indexing services are not set up to handle this type of information retrieval.

One of the major goals of the general pharmacologist, and also workers in the pesticide field, is the correlation of chemical structure with biological activity. Taken to its logical conclusion, this would permit the synthesis of compounds to be tailor-made for a specific biological effect. In certain restricted fields, such as plant growth regulators, this approach has proved to be most fruitful. As more and more information is collected, brought together, and made available in suitable form, chemical-biological correlations will be greatly facilitated. In fact, it might be possible to use the information gained in one field of biological research to solve vexing problems in another. For example, correlations between structure and activity of anticholinesterases active in mammals could shed light on the problems of the development of new insecticides.

The program of the Chemical-Biological Coordination Center takes full cognizance of these facts. The assembly of coded data, both chemical and biological, concerning these interrelationships is the raw material from which useful generalizations may be derived.

Summary

Despite its short history and the many difficulties besetting such an ambitious venture, the Chemical-Biological Coordination Center has succeeded in setting up a repository of coded information concerned with the effects of chemicals upon biological systems. This information is easily retrievable by the use of punch cards and machine methods of searching, and is available to all interested scientists.

The assembly and codification of the metabolism of pesticides in living organisms, including their absorption, distribution, and metabolic alterations such as detoxification and excretion, are proceeding successfully with the use of the techniques described. This information is taking its proper place among the nearly 200,000 items of biological data in the files at the present time.

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