INFORMATION GATHERING AND DATA BASES THAT ARE PERTINENT TO THE DEVELOPMENT OF PLANT-DERIVED DRUGS

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Abstract

An important element of drug development using plants is the accumulation and analysis of pertinent research data and purported ethnomedical (folkloric) uses for plants. Due to the quantity of information frequently available in the literature and its appearance over many years from worldwide sources, this information is best stored and analyzed by computers, There are 10 automated data bases that provide data on plant sciences, agriculture, and the chemistry and pharmacology of natural products, One of these data bases, the Natural Products Alert (NAPRALERT) file, affords both bibliographic retrieval and textual and numeric data relevant to drug development,

Status of Interest in Drug Development From Plants

A careful analysis of the National Prescription Audit from 1952 through 1973 indicates that, contrary to general belief, the higher (flowering) plants continue to provide useful, if not essential, drugs for people of the United States and elsewhere (23). The National Prescription Audit (NPA) collects data on a monthly basis from a statistically significant number of community pharmacies throughout the United States, These data include the number of prescriptions dispensed, the trade mark or generic name of the product prescribed, and the price to the patient, Using these data and information on the origin of active principles (i.e., plant-derived, microbial metabolizes, animal products, mineral and/or pure synthetic), it is possible to determine which of the thousands of different prescription items included in the NPA contain active principles extracted from higher plants.

From 1959-73, new and refilled prescriptions containing plant products represented 25 percent of all prescriptions dispensed from community pharmacies in the United States. Over the survey period, this percentage was never lower than 24 or higher than 26 percent in any year. Incomplete analysis of data for 1980 shows that plant-derived drugs still represent approximately 25 percent of the total prescriptions dispensed.

Since the total number and average price of prescriptions sold annually at community pharmacies are known, the value, at the consumer level, for those prescriptions containing active principles from higher plants can be calculated. However, it is difficult to make the same calculation on the increasing number of prescriptions and quantity of drugs dispensed from out- patient hospital pharmacies, extended care facilities, government hospitals, and other legitimate, but off-market, outlets because data from these sources are not conveniently available. Nevertheless, in consultation with academicians and others knowledgeable in pharmaceutical administration and economics, it was determined that U.S. noncommunity pharmacy prescription sales represent a dollar value almost equivalent to that of drugs dispensed from community pharmacies. According to NPA data, 1.532 billion new and refilled prescriptions were dispensed from community pharmacies in the United States in 1973. At an average cost to the consumer of $4.13 per prescription, the dollar value for the prescription market in 1973 was $6.327 billion. Since 25.2 percent of the prescriptions contained active principles of higher plant origin, the dollar cost to the consumer in 1973 was $1.594 billion. Adding almost an equal amount to this figure for plant-derived drugs dispensed from other outlets, an estimated total cost to the consumer was $3 billion in 1973.

Extrapolating from the 1959-73 data, and looking at high-volume prescription drugs dispensed from community pharmacies in 1980, the current annual cost to the consumer is estimated to be in excess of $8.116 billion,

NPA data for 1980 show that 41 species of higher plants yield all of the plant-derived prescription drugs. An additional 62 species of plants entered the prescription market in the form of crude extracts with active principles. Less than 50 pure compounds from plants were represented in the prescriptions analyzed. The pure compounds, the plants from which they are derived, and other plants used in extract form in prescriptions are listed in table 1.

Virtually all of the approximately 50 pure compounds used as prescription drugs (table 1) still are produced commercially by extraction of plant material or by chemical modification of extracted plant compounds. Although most of these drugs have

*Presented at OTA workshop by N.R. Farnsworth,

Table 1.—Flowering Plants That Currently (1980) Are Sources of Useful Drug in the United States

Plant Names	Family	Type of Drug
Ammi majus	Umbelliferae	Xanthotoxin[a]
Ananas comosus	Bromeliaceae	Bromelain
Atropa belladonna	Solanaceae	Belladonna Extract
Avena sativa	Gramineae	Oatmeal Concentrate
Capsicum species (4)	Solanaceae	Capsicum Oleoresin
Carica papaya	Caricaceae	Papain
Cassia acutifolia	Leguminosae	Sennosides A + B
Cassia angustifolia	Leguminosae	Sennosides A + B
Catharanthus roseus	Apocynaceae	Leurocristine (vincristine)
		Vincaleukoblastine (vinblastine)
Cinchona species (3)	Rubiaceae	Quinine
		Quinine
Citrus limon	Rutaceae	Pectin
Colchicum autumnale	Liliacae	Colchicine
Digitalis lanata	Scrophulariaceae	Digoxin
		Lanatoside C
		Acetyligitoxin
Digitalis purpurea	Scrophulariaceae	Digitoxin
		Digitalis whole leaf
Dioscorea species (several)	Dioscoreaceae	Diosgenin
Duboisia myoporoides	Solanaceae	Atropine
		Hyoscyamine
		Scopolamine
Ephedra sinica	Ephedraceae	•Ephedrine
		•Pseudoephedrine
Glycine max	Leguminosae	Sitosterols
Papaver somniferum	Papaveraceae	Opium
		Codeine
		Morphine
		Noscapine
		•Papaverine
Physostigma venenosum	Leguminosae	Physostigmine (Eserine)
Pilocarpus jaborandi	Rutaceae	Pilocarpine
Plantago species (3)	Plantaginaceae	Psyllium husks
Podophyllum peltatum	Berberidaceae	Podophyllin
Prunus domestica	Rosaceae	Prune Concentrate
Rauvolfia serpentine	Apocynaceae	Reserpine
		Alseroxylon Fraction
		Powdered whole root Rauwolfia
Rauvolfia vomitoria	Apocynaceae	Deserpidine
		Reserpine
		Rescinnamine
Rhamnus purshiana	Rhamnacee	Cascara Bark
		Casanthranol
Rheum species (4)	Polygonaceae	Rhubarb Root
Ricinus communis	Euphorbiaceae	Castor Oil
		Ricinoleic Acid
Veratrum viride	Liliaceae	Veratrum viride Extract
		Cryptennamine

aI.Underlined names indicate single chemical compounds of known structure.
● Also produced by synthesis

been synthesized, synthesis is not commercially feasible, with the exception of a few drugs such as ephedrine, pseudoephedrine, and papaverine.

In addition to the prescription drugs, the sale of herbal teas in the United States is estimated at least $200 million annually and the sale of over-the-counter (OTC) drugs obtained from plants is probably over $1 billion. For example, about one-half of the OTC laxative preparations sold annually in the United States are of plant origin. In 1980, exclusive of prescription sales, the laxative market was about $331 million (an increase of 10.3 percent over 1979). Data such as these are unavailable from other countries, but a similar incidence of use is likely in most of the developed countries.

One might presume that with a current market value at the consumer level in excess of $8 billion annually in the United States, most pharmaceutical firms would have major research programs designed to discover new drugs from plants. This is not the case. In 1974 the American pharmaceutical industry's total R&D budget for pharmaceuticals for human use was $722.7 million. Only one firm was involved in direct research to explore plants for new drugs. At that time a conservative estimate suggests that the annual research budget for that single program was less than $150,000. Although much has been written in scientific journals of this country and abroad that should spark industrial interest to invest in this area of research, * interest in this country has declined. Today there are no U.S. pharmaceutical manufacturers involved in a research program designed to discover new drugs from higher plants.

Other developed countries generally have followed the United States pattern, with two major exceptions: West Germany and Japan. Exploration for new plant-derived drugs in West Germany, however, has not captured the imagination of large pharmaceutical firms. Instead, these studies are supported by many small pharmaceutical manufacturers.

The Japanese situation is nothing short of phenomenal. Not only do Japanese academicians receive government and industrial support for research on plant drugs, but Japanese pharmaceutical firms also have major research departments dedicated to the same goal. Scientific papers from Japanese research laboratories relating to drug-plant development generally are of high quality and outnumber those from the United States by at least tenfold. In addition, Japanese patents for plant-derived biologically active substances outnumber those from the United States by a factor of at least **50.**

*If none have been tested in humans yet.

Factors Responsible for Decline of Interest in Drug Plant Discovery

Important decisionmakers connected with drug development apparently are unaware of the extent to which plants contribute to our source of drugs. During the past three or four decades, research administrators have been scientists trained and experienced primarily in synthetic organic chemistry, biochemistry, molecular biology, microbiology, medicine, or related fields. Plant sciences have not had a voice in corporate or high-level governmental decisionmaking, except in the area of basic agriculture. Further, science has advanced to a point where interest has not been on the intact organism—whether it be plant, animal, or microbe—but rather on the cell, cell contents, and cell biochemistry. Typical industrial drug development increasingly has involved synthesis of molecules based on structure-activity relationships, and natural product drug development has been restricted to antibiotic production by micro-organisms.

If drug development programs now are based on synthesis and involve structure-activity relationships, what scientific process is available to identify plant-derived drugs? Most useful drugs derived from higher plants have been "discovered" through scientific inquiry into alleged folkloric claims of therapeutic effects. The only other approach used in recent years, although unsuccessful, has been the random collection of plants followed by pharmacologic evaluation. This approach has been modified in several ways. For example, the Smith, Kline and French Pharmaceutical Co. (SKF) had an extensive program for several years. SKF sent workers into the field to carry out "spot tests" for alkaloids on any plant that was reasonably abundant. Plants giving positive tests were collected in 1-to 5-kg amounts and sent to the parent research laboratories in Philadelphia. The total alkaloid fraction was separated from each species and then subjected to a series of pharmacological tests. Nothing has ever been published to provide an understanding of the rationale for the selection of pharmacological "screens" used in this program. The total number of plants bioassayed was also unreported, although they probably numbered several hundred species. Many scientific reports came out of the SKF studies, but they lacked pharmacologic information.

A major program of the National Cancer Institute involving the "screening" of plants randomly collected worldwide ended in late 1981. This program, which was started in about 1956, tested some 35,000 species of plants for antitumor activity in laboratory animals. Although a large number of highly active antitumor agents were discovered by the NC I program, none have been approved for use against human cancer by the Food and Drug Administration (FDA). A few remain to be studied in humans. These* plant-derived antitumor agents that appear promising and presumably will undergo eventual testing in humans are tripdiolide, indicine-N-oxide, baccharin, isobaccharin, taxol, 10B-hydroxy with anolide, bruceantin, and homoharringtonine. The results of this program have been reviewed through early 1982 (8,12,60,61).

One could argue that this approach of "blind" screening any and all plants available in sufficient quantity for testing is entirely unscientific and too costly to be continued. Perhaps these are reasons for the NCI's canceling its program on antitumor plants. Surely a program that fails to produce a useful drug after 25 years of testing more than 35,000 species of plants must be examined to determine what went wrong!

Speaking in defense of the NCI screening program, Suffness and Douros (59) stated:

> It is unfortunate that there has been a mutual skepticism between theoretical researchers, who are interested in a "rational" approach of discovering the biological differences between neoplastic and normal cells in order to develop drugs, and the approach of the empiricists, with the former group feeling that the latter are using a hit or miss unscientific approach and the latter feeling the former are divorced from the practical aspects of the problems. One hopes that this situation will progress in such a way that the information gained by the empiricists can be used to help the theorists develop new experimental methodologies and that the theoretical information produced by the rationalists can help the empiricists in their research. Structural analysis of new compounds developed through an empirical approach has given us the basis to develop analogs of a compound which might be beneficial and better than the parent active compound.

With the amount of data now available from the NCI antitumor plant program, it would be interesting and useful to conduct a scientific retrospective analysis on data of all 35,000 species of plants tested in this program,

A program at the Eli Lilly Co. * discovered two useful antitumor drugs that still are widely used today. This program identified about 440 species of plants for inclusion in a broad biological screening program, based on literature reports and recommendations of Dr. Gordon Svoboda. *Catharanthyus roseus,* the Madagascar periwinkle, was the 40th species bioassayed within the program and yielded the useful antitumor agents leurocristine and vincaleukoblastine. From the discovery of the antitumor activity of a crude extract of this plant, through the isolation, biological testing, toxicity, and clinical trials of vincaleukoblastine, less than 4 years were required for FDA approval and the marketing of this drug for the treatment of human cancer. The second useful alkaloid, leurocristine, was marketed 2 years later.

Nothwithstanding the work on *Catharantus,* industry's development of useful drugs from higher plants during the past 30 years has not been more productive than government efforts. Since 1950, pharmaceutical companies have produced only three plant-derived drugs that have reached the U.S. prescription market: reserpine (and the related deserpidine and rescinnamine), vincaleukoblastine, and leurocristine. The total number of industry research dollars required to discover these drugs has been miniscule compared with the cost of placing synthetic drugs on the market. If industry invests next to nothing in drug-plant development, what can it expect in the way of marketable compounds?

Perhaps a major deterrent to industrial interest in plant drug development is that much of the unexplored flora of the world currently is found in tropical and/or semitropical areas, especially in developing countries. Because many of the countries' governments are considered to be unstable, an uninterrupted supply of raw material for manufacture of a new drug developed from a plant indigenous to such areas may not always be assured. This seems a weak argument to justify the lack of plant drug development, since rarely is a plant found only in one country. In addition, alternate sources of supply, including cultivation, always should be developed when a useful drug is discovered.

Some companies feel that patent protection is less secure with natural products than with synthetics. We have not been able to document examples supporting this contention. To the contrary, there is no evidence that the Eli Lilly Co, experienced difficulties with patents assigned to them for the anti-

*(5), (11), (14), (15), (16), (17), (18), (19), (20), (23), (24), (54), (55).

*See paper by Svoboda in this volume.

tumor alkaloids vinceleukoblastine and leurocristine, whose annual worldwide sales are $50 million at the manufacturer's level.

Information That Should Be Included in Data Base Useful for Drug Development From Plants

A data base that would be useful in a program of drug development from plants should include the following information:

The Plant

Information on the plant should include: its most current Latin binomial and all recently employed botanical synonyms, along with the authority, varieties, subspecies and lower taxonomic groups, the family name of the plant, and vernacular (common) names; the geographic area where the plant was collected and all known areas where the plant occurs naturally or under cultivation; whether or not a voucher specimen representing the investigated material was prepared and stored; and the part(s) of the plant studied.

Ethanomedical Uses

Documentation of the crude plant material (fresh, dried, pulverized) or type of extract employed in preparing the ethnomedical dosage form, route of administration, dose employed, dosage regimen, side effects, and beneficial effects claimed should be retrievable from the database. The ethnomedical claim(s) should also be computerized to allow for retrieval of information on analysis of related effects. For example, a plant extract maybe alleged to be useful as an ethnomedical preparation for arthritis and/or rheumatism. Experimental data may have been reported elsewhere indicating that an extract of the same plant has shown anti-inflammatory activity in vivo and in vitro. It may also have been reported to have analgesic activity and to inhibit prostaglandin synthetase in vitro. These experimental pharmacologic effects suggest that the extract could be beneficial in treating arthritis and/or rheumatism. Ideally, a data base useful for drug development would have the capability of relating general ethnomedical information with experimental data to predict useful drug effects.

Chemical Constituents of Plants

It is important to be able to retrieve data on the chemical constituents of plants, the part of the plant in which the constituent is reported to be present, the range of percentage yield of the chemical, and any available data on seasonal or other variations in chemical quality or quantity.

If a chemical compound is known to produce a pharmacologic effect, the presence of such a compound in the plant may explain either the experimental pharmacology of an extract of the plant or an ethnomedical claim relating to the effect of the known chemical constituent. An appropriate computer analysis relating ethnomedical claims, experimental pharmacology of plant extracts, occurrence of chemical constituents, and pharmacological effects of naturally occurring chemical constituents may indicate that all aspects of a given problem have been studied, even though the answer is scattered in several publications from different parts of the world. Such computer analysis can prevent costly and time-consuming redundant research. Sources of all data should be coded and specifically linked for subsequent retrieval and reevaluation or reinterpretation as may be necessary.

Additional valuable information in a data base is classification of various naturally occurring chemical compounds by general class, sub-structure, and functional groups on the molecule. Computer analysis of a series of plant-derived chemical constituents with regard to chemical structure biological effects could be useful in the advanced stages of drug development.

Experimental Biological Testing

For Plant Extracts. It is important to be able to identify the type of solvent used to prepare an extract of a plant tested for biological activity, since, for example, an extract using petroleum ether would be expected to react differently than one prepared with ethanol. Further, the type of biological test—i.e., in vitro, in vivo, or human study—is important, since the credibility of data with respect to its ultimate value as a drug would be of the order in vitro < in vivo < human study.

A system for identifying similar and related biological effects would be required if computer analysis of the data is anticipated. Thus, central nervous system, autonomic, hematologic, chemotherapeutic, and/or hormonal effects, etc. should be organized so they can be retrieved and analyzed as a group or for a specific effect within each group. Test species should be identifiable, since data using rodents, for example, may not be as significant as data derived from primate studies. The route of administration is important, as is the dose, dosing scheduled, and qualitative and/or quantitative

result. The disease states of animal models or the type of experimental model also should be documented.

For Pure Compounds. All of the above parameters should be documented for test results on plant chemical constituents, with the obvious exception that the type extract would not apply.

Other Important Information

It would be of interest in any plant-derived drug development program to be able to retrieve references to naturally occurring chemical compounds that have been partially or completely synthesized. Similarly, data relating studies on the cultivation of useful drug plants and the effects of cultivation conditions, processing, and storage on the yield of useful active constituents would be useful in a data base for drug-development purposes.

Eventually plant-derived drugs probably will be produced by cell culture, although the necessary technology is not so advanced that commercial applications of tissue culture for this purpose will be available soon. Little work is being supported either by industry or government in the United States to develop plant tissue culture for this practical application.

Similarly, genetic engineering some day may be applied to producing plant-derived drugs. However, the production of useful drugs by plants is always a multistage process. Transfer of several sets of genes to microbes would be required to produce secondary metabolizes that are useful drugs. We do not know of any current efforts in this area. Most likely a detailed examination of the biosynthesis of the drug that would be a candidate for production by genetic engineering would have to be carried out in each case, Again, basic studies that would provide this type of information are not being supported to any extent in this country,

Accessibility of Data Pertinent to Plant-Derived Development

Data of value in organizing and implementing an effective plant-derived drug development program are difficult to obtain through currently available on-line data bases. Further, much of the useful pharmacologic data on plant extracts was published prior to the earliest data covered by all of the available online data bases. Also, the format in which data can be retrieved from most available systems is inadequate for practical use in a major program.

It has been our experience that the pharmaceutical companies are reluctant to share data derived from testing plant extracts, even if the data are negative and are of no commercial interest to the firms, In the United States alone, some pharmacological data on perhaps several thousand species of plants have been documented by industrial firms, but remain unpublished. Negative data are as important as positive data in a broad plant-based drug development program, and it is unfortunate that some mechanism cannot be found to persuade industrial firms to make their unpublished in-house pharmacologic data on plants available to the general scientific community.

The National Cancer Institute published negative data regularly at the beginning of the plant screening program. This apparently became an expensive exercise because most of the data now remain unpublished. If one requests information on a few species of plants, NC I will provide the information from its files, but probably would not supply large amounts of data.

If one examines the format for the initial negative data published by NCI on testing plants for antitumor activity, it is obvious that much of the space-consuming information was superfluous-i. e., one line of data for each dose level tested. Since all the data are computerized, NCI should be encouraged to produce several volumes of negative data, allowing only one line per species with similar testing conditions. An entry of data would then appear as follows: **Genus-Species-Authority-Family-Country Where Collected-Part-Tumor System.** Such a list of negative data would be of enormous interest to scientists throughout the world and would help them in selecting new plants for study without duplicating past work.

A similar problem exists with obtaining large amounts of useful data from published reports. For example, there are several hundred literature reports in which large numbers of plants have been screened for one to at least thirty biological effects. The names of plants in these multiple-entry reports rarely are obtainable from the subject indices of *Chemical Abstracts or Biological Abstracts,* and presumably from data bases using these sources.

Steps Involved in Gathering Information and Implementing a Plant Drug Development Progam

Diczfalusy (11) has discussed the general steps in developing drugs of plant origin. After the initial decision is made concerning how candidate plants

are to be selected for investigation, procedures to be followed are reasonably straightforward (see table 2).

Information Gathering

Suffness and Douros (59) have given their views on the relative advantages of using folkloric information, botanical relationship, and random collection approaches to identifying plants for inclusion in the NCI screening program (see table 3). If the plants to be screened are not chosen randomly, they should be chosen on the basis of available information.

Information-gathering to identify candidate plants for study will vary considerably, but most likely will necessitate searching for data in the following areas.

Ethnomedical data. Ethnomedical (folkloric) information on plants is not indexed or abstracted in any organized way. Sources include books on medical botany, review articles, floras of various geographic areas, pharmacopoeias of different countries, anthropological writings, folkloric writings, and popular books on uses of plants. No one can be reasonably certain that all data collected for a given plant or for a biological effect of interest have been acquired from ethnomedical reports. To the best of our knowledge, only the USDA data base* and our NAPRALERT system (to be described sub-

*See paper by Duke in this volume.

Table 2.—Selection of Approachs for Drug Development From Plants[a]

	Method of Selection	Advantages	Disadvantages
L	Folklore Use	High ratio of activity	Role of psychology in folk medicine
		Lower screening costs	Secrecy of primitive cultures
			Difficulty of botanical identification
			Use of complex plant mixtures
			Use of rare plants
			High procurement costs
			Leads may be missed
			Limited amount of screening possible
			Lack of novel active compounds
2.	Botanical Relationship	High ratio of activity and Discovery of useful analogs	Reisolation of known compounds
3.	Random Collection	Best chance to find novel active compounds	Low percentage of active leads
			Large bioassay capacity needed
		Plants are more readily available	High cost per lead

[a]Modified from Suffness and Douros (1979).

Table 3.—Steps Required for Drug Development From Plants

1. Decision on method to select plants
2. Preparation of extracts for bioassay.
3. Initial collection and identification of plants
4. Bioassay of plants
5. Recollection of active plants for confirmation of activity
6. Fractionation of active plants following bioassay.
7. Isolation of active compound(s) in sufficient quantity for bioassay and identification or structure elucidation
8. Determination of pharmacologic profile for active compound(s)
9. Acute and chronic toxicity studies
10. Large scale preparation of the active compound(s)
    a. Large scale procurement of active plant
    b. Pilot plant scale development of efficient isolation procedures
11. Initiate studies designed to prepare the active compound by complete synthesis and to prepare active analogs
12. Formulation studies
13. Pharmacokinetic and metabolism studies
14. File an Investigational New Drug Application (INDA) with the Food and Drug Administration
15. Phase I human studies
16. Phase 11 human studies
17. Phase HI-IV human studies
18. File a New Drug Application (NDA) with the “Fred and Drug Administration
19. Market the new drug

sequently) contain retrievable ethnomedical information. Some people discount the value of folkloric reports of medical efficacy of plants, but in doing so refute the historical documentation of how such information was used in the discovery of such major drugs as digitoxin, reserpine, tubocurarine quinine, codeine, morphine, and others, The degree to which one uses ethnomedical information will vary according to the background, training, and capabilities of scientists involved in a drug-development program.

Experimental Data on Plants. The subject indices of several abstracting services usually identify plants tested for biological effects or plants from which chemical compounds have been obtained. We routinely use those abstracting services listed in table 4, which identify the period and type of literature covered in each case.

A word of caution has to be given, however. We have found less information on plants in decennial indices of **Chemical Abstracts** when compared to annual indices. We have also found that a great deal

Table 4.-Secondary Literature Indices Used in the NAPRALERT Program

Title	Period Covered
INDEX CATALOG OF THE SURGEON GENERAL	*1880-1961*
INDEX MEDICUS	*1897- 1927; 1960 to present*
CHEMICAL ABSTRACTS	*1907 to present*
QUARTERLY CUMULATIVE INDEX MEDICUS	*1916-1956*
BIOLOGICAL ABSTRACTS	*1926 to present*
CURRENT LIST OF MEDICAL Literature	1941-1959
UNITED STATES ARMED FORCES MEDICAL JOURNAL	*1950-1960*
NATIONAL LIBRARY OF MEDICINE CATALOG	*1956-1965*
NATIONAL LIBRARY OF MEDICINE CURRENT CATALOG, CUMULATIVE LISTING	*1966 to present*
CURRENT CONTENTS, LIFE SCIENCES	*1967* to *present*

of useful information, especially concerning pharmacological effects of plant extracts, is found in earlier abstracting services (i.e., those indicated in table 4 that cover periods prior to 1907). We also routinely check the bibliographies of pertinent articles, especially review articles, on subjects of concern to the area of the chemistry and pharmacology of specific plants and/or specific pharmacologic effects and find a significant number of useful papers that were not identified through systematic literature searches of the services indicated in table 4.

If current work indicates that one or more research groups throughout the world are publishing extensively in an area of interest, correspondence with those groups usually uncovers additional literature that is not usually available through indexing services.

Finally, the use of one or more of the online computer data bases to acquire citations and/or abstracts on a particular subject may be advisable.

In our laboratory, experimental work on a plant does not commence until we have acquired all of the ethnomedical and scientific literature available through the aforementioned means, including data on taxa closely related to the plant of interest.

Plant Collection. It has been our experience that although botanists are essential to the success of plant-derived drug development programs, they are the most unappreciated of the scientists involved in such a program. The botanist can not only clarify nomenclatural problems but has the credentials to contact other botanists in countries where the plants of interest grow. The botanist can provide sources of background information that would be unknown to the chemist and/or pharmacologist. Often this information is critical to a project's success. Through field work, the botanist can clarify vague points found in the literature on which several varieties of a species should be collected and can gather additional information from users of plants in the area where collections are to be made. These data may be useful to the pharmacologist on a drug development team in designing an animal study to assess the biological effects of a plant.

A number of government regulations pertaining to plant importation must be considered. For ex-

ample, depending on the method used, fumigation of a plant entering the United States could produce chemical changes in the plant, rendering the chemical constituents inactive. The botanist can maintain communication with government botanists at points where plants enter the country and often can prevent such problems.

Bioassay Procedures. A critical stage in drug development from plants is the selection and implementation of bioassay procedures and interpretation of their results. Few pharmacologists are experienced in evaluating crude plant extracts or are interested in developing bioassay systems outside their own area of interest, In a large program, where several pharmacologists can become involved, "specialization" may be possible. However, even though a program is designed to identify only one or two types of biological activity, it would be cost ineffective not to carry out as many bioassays as possible, since the expense of acquiring the plant may be quite high.

Because there is inherent variation in laboratory animal experimentation, drug development programs involving plants should include a series of so-called in vitro "pre-screens. " Plants should be selected on the basis of having predictive activity in animals and/or humans. Examples of some in vitro bioassays used for plant extracts are presented in table 5. If initial in vitro activity seems promising, in vivo activity can be confirmed in laboratory animals. Alternatively, isolation of the compound responsible for the activity can be accomplished using bioassay-directed fractionation with simple, rapid, and less expensive in vitro tests. The pure compound then can be tested in laboratory animals.

Many ubiquitous chemical compounds found in plants elicit biological effects but have no practical use as drugs, It would be important to identify such compounds, carry out a simple chemical test to indicate the presence of the compound in plants to be bioassayed, then modify experiments to rule out the effect of the undesired ubiquitous compound.

Isolation and Identification of Biologically Active Principles From Plants. Presuming that the goal of a drug development program is to develop pharmaceuticals, it is first necessary to establish a biological effect for a plant, then isolate the compound(s) responsible for the effect by bioassay-directed fractionation. Some biological activity may not be detected by means of a simple and rapid in vitro bioassay, thus presenting a major problem. If, however, the desired biological activity can be demonstrated in a laboratory animal after one or two doses, the in vivo bioassay can be used as an aid to fractionation. It is most useful to have bioassay capability in-house rather than to rely on outside chemical work.

Routine techniques are available for isolation and purification of the active principle(s) after one has fractionated a plant so that biological activity has been enriched severalfold. Column chromatography, ion-exchange, droplet counter-current chromatography, and high performance liquid chromatography are some well-established techniques. However, if the active compound is to be developed into a marketable item, a practical means for isolating it from the plant must be found, Few compounds exist that could be separated economically from plants by chromatographic techniques and every effort should be made to use simplified procedures.

Once the active principle has been obtained in pure form, identification is routine if it is a known substance. In cases of novel compounds, methods are available to determine the structures in a relatively short period of time, providing that modern instrumentation is available, including pmr, cmr, ms, and X-ray crystallography equipment.

Clinical Evaluation and Marketing. Any drug development program, whether it is industrial, academic, or governmental, must at some period involve the partnership of an industrial firm. Academia and Government usually do not have the capability to market a drug, but industry has the experience and know-how. Clinical evaluation is an expensive and complicated process, and is possible in the United States only after filing an investigational New Drug Application (INDA) with the Food and Drug Administration (FDA). If approved, human experimentation within certain guidelines is allowed. Prior to marketing a drug, a New Drug Application (NDA) must be filed with FDA, providing all data from the results of Phase I-IV human studies allowed by the INDA, The drug, if approved, may then be marketed. The estimated cost for developing a drug varies depending on the clinical applications, but would not be less than $5 million and could be as high as $35 million per new drug.

Data Bases Pertinent to Drug Plant Development

There are about 1,000 online data bases—i.e., those searchable by the user from various types of terminals (10,27,66). Ten data bases seem to have information useful to a plant-derived drug develop-

Table 5.—Examples of in vitro Bioassays That Are predictive for Useful Drug Effects or Improvement of Health

Type Assay	Type System	Implied Useful Effect	Reference
Adenosine deaminase inhibition	in vitro	Enhancement of drug efficacy	Kalckar, 1947
Angiotensin-converting enzyme Inhibition	in vitro	Antihypertensive	Yun, Chung & Han, 1981
Antibacterial activity	bacterial culture	Anti infective	Farnsworth, et al., 1966 Mitscher, et al., 1972
Antifungal activity	fungal culture	Antiinfective	Farnsworth, et al., 1966 Mitscher, et al., 1972
Antimitotic activity	cell culture	Anticancer	Suffness & Douros, 1982 Abbott, 1980 bade, et al., 1981
Antimutagenic activity	cell culture or bacterial culture	Anticancer	Calle & Sullivan, 1982 Katak, et Q., 1980 Stich, Rosen & Bryson, 1982 Wood, et al., 1982
Antiviral	cell culture	Anti Infective, anticancer	Farnsworth, et al., 1966 Markkanen, et al., 1981
ATPase inhibition	in vitro	Cardiotonic	Barnett, 1970; Chen, et Q., 1982 Duggan & Nell, 1965
Benzpyrene hydroxylase (AHH) inhibition	in vitro	Carcinogenesis inhibition	Pezzuto, et al., 1978
Cell transformation	cell culture	Carcinogenicity detection	Merriman & Bertram, 1979 Nescow & Heidelberger, 1976
Cytotoxicity	cell culture	Anticancer	Suffness & Douros, 1982 Germ, et al.,
Free radicals	in vitro; cell culture	Anticancer	Sarriff, White & DiVito, 1979 Tappel; 1972
HMG-COA reductase inhibition	in vitro	Antihypercholesterolemic, antiatherosclerotic	Stacpoole, Varnado & Island, 1982
Human stem cell assay	cell culture	Anticancer	Salmon, et al., 1978 VonHoff, et al., 1981

a β-Hydroxy-β-methylglutaryl CoA reductase

Type Assay	Type System	Implied Useful Effect	Reference
Insect antifeedant	in vitro	Prevent crop damage and insect-borne diseases	Meisner, et al., 1981 Munakata & Wada, 1981
Insecticide	in vitro	Prevent crop damage and insect-borne diseases	Su, Horvat & Jilani, 1982 Jacobson, et al., 1950
Molluscicide	in vitro	Lower incidence of snail-borne diseases, .e. schistosomiasis	Anon., 1965
Monoamine oxidase (MAO) inhibition	in vitro	Antihypertensive	Robinson, et al., 1968
utagenicity	cell culture or bacterial culture	Carcinogenicity detection	Ames, McCann & Yamasaki, 19' O'Neill, et al., 1977 Ungsurungsie, et al., 1982 Pezzuto, Moore & Hecht, 1981
Nucleic acid biosynthesis	in vitro; cell culture		Robinson, et al., 1968 Kuchler, 1977
hosphod inhibition	in vitro		Nikaido, et al., 1981
Piscicide	in vitro	Predictive for molluscicide effect	Bade, et al., 1981
Platelet aggregation inhibition	in vitro	Cardiovascular problems	Makheja, Vanderhoek & Bailey, 1979
g an synthetase inhibition	in vitro	Antiinflammatory	Ohuchi, et al., 1981 Saeed, et al., 1981
Protease inhibition	in vitro	an plant protection	Lewasz, Rys & Reda, 1981 Norioka, Omichi & Ikenaka, 1982 Ohkoshi, 1981
Protein biosynthesis	in vitro; cell culture	Antibiotic; anticancer	Pezzuto & Hecht, 1980 Woodward, Ivey & Herbert, 1974
Sister chromatid exchange	cell culture	Carcinogenicity detection	Perry & Evans, 1975
Tyrosine hydroxylase inhibition	in vitro	Antihypertensive	Waymire, Bjur & Weiner, 1971
Unscheduled DNA synthesis	cell culture	Carcinogenicity detection	Freedman & San, 1980 Sirica, et al., 1980

ment program. Nine of these are summarized in table 6, the tenth is developed at the USDA by Dr. James Duke.* We are unaware of data bases that contain information pertinent to plant-derived drug development in the People's Republic of China, Japan, or the U.S.S.R.

AGRICOLA (formerly CAIN). This database is produced by USDA but is available through commercial vendors such as BRS, DIALOG, and others. It covers the literature from 1970 to the present on agricultural chemistry and engineering, food and plant science, nutrition, and related agricultural fields. Approximately 6,500 journals are reviewed for input. The bibliographic file is composed of 90 percent journal articles and 10 percent monographs, published proceedings, and/or theses, Subject analysis by the user is carried out using controlled keywords-i. e., those provided in a prepared thesaurus or through enriched titles. Chemical identifiers include the use of nomenclature codes, fragmentation schemes, or the chemical name as it appears in the source of data.

BIOSIS PREVIEWS. This file has been produced by Biosciences Information Services and is also available through commercial vendors. Its subject matter includes life science data for microbiology, the plant and animal sciences, experimental medicine, agriculture, pharmacology, ecology, biochemistry, and biophysics. A search of the file provides bibliographic data for information obtained from approximately 94 percent journal articles, less than 1 percent government documents, and the remainder from monographs, published proceedings, or theses. Search methods include the use of enriched titles, uncontrolled keywords (i.e., keywords selected by the user and not from a prepared thesaurus), and subject headings that may appear in the title. Chemical identification uses the same type information described above (see AGRICOLA).

CAB SYSTEM. The Commonwealth Agricultural Bureau System of England produces this file, which is also available in the United States. The CAB System has bibliographic data and abstracts obtained from a review of approximately 8,000 journal titles covering the literature from 1973 on. It represents a compilation of 20 agricultural science databases, and includes subjects such as animal and plant breeding abstracts, plant pathology, nutrition, entomology, and related fields. Searching this database requires the use of uncontrolled keywords and subject titles.

*See paper by Duke in this volume.

FSTA. This is an acronym for Food Sciences and Technology Abstracts. Its subjects include agricultural chemistry and engineering, food science, home economics, and patents from approximately 1,300 journal titles. The resulting database is composed of 85 percent journal articles, 10 percent patent information, and 5 percent monograph, government document, or theses data. Search techniques use uncontrolled keywords and/or enriched titles. Chemical compounds again are identified using registry codes, fragmentation schemes, or the name which appears in the document processed. All data are bibliographic with abstract summaries for the years 1969 to the present,

CACON (Chemical Abstracts Condensate). CACON is one of the largest and most widely distributed bibliographic databases in the world. This file is a product of Chemical Abstract Services and covers most life sciences and physical sciences from 1968 on. Literature coverage includes input from approximately 14,000 journal titles and results in a computerized file made up of 72 percent journal articles; 2 percent government documents; 10 percent monographs, published proceedings, or theses; and 16 percent patent information. Subject retrieval is carried out using uncontrolled keywords, which may appear in the article title or its abstract, subject headings, and/or word strings. The latter are searched in the computerized abstract material.

MEDLINE. This file, produced by the National Library of Medicine (NLM), covers the literature from 1974 on. Depending on the literature period of interest, it is offered either on-line or off-line. Input data are obtained from approximately 3,000 journals titles comprising life sciences and/or medical information. These data are represented in the file by 99 percent journal articles, of which 65 percent are in the English language. The remaining 1 percent of data represents government documents. Retrieval provides bibliographic data with an abstract summary and is obtained by using keywords and/or subject headings.

IMEPLAM. This is the acronym for Instituto Mexicano Para el Estudio de las Plantas Medicinales (Mexican Institute for the Study of Medicinal Plants). Data contained in this data base have been derived from 41 literature citations primarily concerned with collections of natural history and folkloric claims for Mexican plants. These volumes, which have been computerized, date from 1552 to 1973, We have not been able to document whether current literature is being added to these data. Its primary value is with regard to ethnomedical data

Table &-Major Computerized Sources **of Scientific and Technical information on Natural Products**

DATA BASE NAME (ACRONYM)	LITERATURE COVERAGE (from)	TYPE DATA RETRIEVABLE	NUMBER OF JOURNAL TITLES REVIEWED	SCOPE OF COVERAGE
AGRICOLA (formerly CAIN)	1970	Literature citations	6,500	Agricultural chemistry and engineering; food and plant science; nutritional data, etc.
BIOSIS Previews	1969	Literature citations	8,000	Life sciences; microbiology; plant and animal sciences; experimental medicine; agriculture; pharmacology; ecology; biochemistry; biophysics.
CAB System	1973	Literature citations + Abstr.	8,000	Agricultural science; plant and animal breeding; plant pathology, nutrition; entomology.
FSTA (Food Sci. & Tech. Abstr.)	1969	Literatue citations + Abstr.-summary	1,300	Agricultural chemistry and engineering; food science; economics; patents.
CA SEARCH	1978	Literature citations	14,000	Life sciences; physical sciences
MEDLINE	1974	Literature citations + Abstr. summary	3,000	Life sciences; medical information
IMEPLAM	(a)	Literature citations + Plant use data (b)	41 (c)	Clinical and folklore use; type pharmacological testing; Mexican plants only.
Chinese University of Hong Kong		Literature citations + Abstr. summary		Retrospect and current information on Chinese Traditional medical practices.
NAPRALERT (Natural Products Alert)	1975 (d)	Literature citations + tabular numerical and textual data from article	8,000	Plant, microbial and animal (primarily marine) chemistry and pharmacology; folkLore.

(a). Retrospect information obtained from reference materials and books covering periods from 1552 through 1973.

(b.) Categorical data only no numerical or textual information.

(c.) Fixed bibliography. No indication of current awareness data entry.

(d.) Also includes retrospect data on more than 500 genera of plants extending into the mid-1800's.

systematically presented for a significant number of Mexican plants. Pharmacological testing data in the file are not presented in detail.

Chinese University **of Hong Kong.** This data base is purported to contain both retrospect and current literature citations as well as brief abstracts of Chinese publications on traditional medical practices. The availability of this data base in the United States has not been established. Nevertheless, the systematic and well organized approach of traditional Chinese medicine makes this data base worth listing as an adjunct to any program for drug development from plants.

Although the literature coverage of these combined databases is quite comprehensive, the inability to compare, correlate, or analyze research data analytically—since they are bibliographic in nature—is a serious drawback of using the data bases in drug or new product development. These data bases provide only lists of citations within the specificity of the search terms—i. e., keyword and/or phrases. If one attempts to be too specific, many citations are overlooked because such specific terms are not used in titles or summary abstracts. On the other hand, broad descriptions within the search terms provide many citations that are not pertinent to new product development research.

Ideally, one would desire a nonbibliographic file that could list and, by computer, compare or correlate real data contained in pertinent citations, then present the data in a digestible tabular form and provide the citation sources. Such nonbibliograhic data files on natural products are not available on-line to the best of our knowledge.

Such a data base does exist off-line. In 1975, NAPRALERT, the acronym for Natural Products Alert, was conceived as a program to overcome the difficulties in using existing files for developing research projects associated with natural products and drug development.

NAPRALERT Content. NAPRALERT, representing a survey of the world literature on natural products, was started in 1975 through a systematic review of each issue of *Chemical Abstracts* and *Biological Abstracts*. Since that time, a large number of retrospect literature searches have been carried out on organisms or subjects of special interest. These retrospect searches cover the literature from the mid 1800's or early 1900's to the present. Approximately one-third of the citations now computerized in NAPRALERT represent these retrospect data. The NAPRALERT database contains information from about 65,000 research reports, books, reviews, patents, and dissertations.

a. **NAPRALERT Data Types.** In addition to bibliographic information, NAPRALERT records the chemical constituents and pharmacological bioassay information from plant, microbial, and animal (primarily marine) extracts. The accepted and synonymous nomenclature of the organism, the parts of the organism used in the study, and the geographic source of the material are also important elements of the data base. In addition to published data, many different indicator codes are used to alert the user that literature is available on the subject or to perform unique sorting capabilities. Another unique, but useful, parameter of the NAPRALERT file, is the recording of ethnomedical, or folkloric, notes as they are encountered in the literature. As a single bit of information, such claims may be of doubtful value, but when correlated with known pharmacologic data or chemical constituents, such data can provide interesting relationships to rationalize the plant's investigation.

b. NAPRALERT Application. Two especially important applications of this data base are related to identifying potential sources of useful new products. First, random screening of plants can be systematized by an organized computer search and analysis of data to provide a ranked list of the more probable candidates for study prior to any field efforts. Such a methodology was developed using NAPRALERT and was tested for a WHO Task Force assembled to identify indigenous plants potentially useful for human fertility regulation.

In this instance, approximately 4,500 plants were identified as purported to affect fertility, based on folkloric claims or laboratory experiments. To select plants randomly for laboratory investigation from a list this size would not be feasible or cost efficient. Thus it was decided to use the pharmacological data, folklore information, and geographic data contained in the NAPRALERT database as a means of identifying the most promising plants for initial study. After developing appropriate software and subsequently analyzing NAPRALERT-contained data, approximately 300 plants from the list of more than 4,500 were identified as the most promising for study. To date, 50 of these have received preliminary laboratory investigation and eight have confirmed activity using two different laboratory animal assays. Although successful development of a useful clinical drug has not been achieved yet, it seems that such a predictive analysis of appropriate computerized data can be a valuable adjunct to new drug development. In this instance, a list of plants with a predictable area of biological activity was generated by evaluation of

the non-bibliographic computerized research data rather than by a costly and time-consuming random field screening process. Certain aspects of the WHO-supported practical application of the NAPRALERT data base have been published (22,54).

A second important application of the information contained in the NAPRALERT file is the ability to present within one computerized report all published folklore information, biological activities—whether assayed in vitro, in animals, or in humans—toxicities, and chemical constituents. In this form, these data can be used effectively to assess potential dangers or suggest safeguards which should be employed when using certain plants. These same data may suggest areas for research on new uses or applications of a specified plant or group of plants. To envision such results only from a stack of publications can be a formidable and frustrating alternative approach.

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