Molecular Pharmacognosy: an Explanatory Model

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Increased interest in the study of natural products as potential drugs and rapidly changing research strategies are driving us to reassess the role of pharmacognosy in the wider context of pharmaceutical research. The authors propose a new definition and an explanatory model of modern pharmacognosy that can be used as a theoretical foundation for future development of this classical branch of the life sciences.

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In the latest edition (1996) of the classical textbook *Trease and Evans' Pharmacognosy,*[1] pharmacognosy is defined as 'the study of those natural substances. principally plants, that find use in medicine'. In the 1992 edition of *Drugs of Natural Origin* by Gunnar Samuelsson[2], we lern that pharmacognosy deals with ' natural products used as drugs for the preparation of drugs'. Samuelsson also describes pharmacognosy as a multidesciplinary subject that comprises parts of botany, organic chemistry, biochemistry and pharmacology.

With increasing specialization through out the pharmaceutical sciences, such broad description, however true, are not specific enough to convay an understanding of the principals underling modern research and teaching in pharmacognosy. We would like to present here our view of modern pharmacognosy and a model that we have successfully used to explain the subject to scientist of other disciplines, as well as undergraduate in pharmacy and medicine. In addition, we would like to emphasize the continuing usefulness of the pharmacignostic approach in the drug discovery process.

Research in Pharmacognosy

The appearence of biologically active metabolites in nature is determined by ecological needs and biosynthetic possibilities. The co-evolution of plants, insects, microorganism and mammals leads to the systhesis of secondary metabolites with defensive, attractive and other functions[3]. Classical pharmacognosy dealt with the taxonomy and morphology of plant drugs, and neglected their chemistry. In the past 40 years, however, much research has forcussed on the isolation and structure elucidation of active constitutes, but the bioactivity itself has often been neglected. Modern pharmacognosy cannot ignor this fundamental feature of plant-derived drugs, specially because the development of new technologies offers novel research possibilities.

By forcussing on biological activity, modern pharmacognostic research has a very good chance of finding new drug leads. In his resent book on drug prototypes, W. Sneader enumerates 244 prototype molecules, most of which are derived from natural products. He also shows that academic research has been a great source of such discoveries[4]. pharmacognostic research can point the way to new descoveries not only by identifing new structures, but also by pointing to 'new' biological activities. A classical example here is the extensive research on hallucinogenic plant drugs, including the discovery of agents such as mescaline, psylocybin and tetrahydrocanabinol, which has had a profound impact on neuropharmacology. Another birilient example is the work on poisonous frogs and their toxins (Batrachotoxin, Epibatidine and Histrionicotoxin)[5]. An excellent review on natural products as leads for new pharmaceuticals has recently been published[6].

Three-point model

Our model to explain modern pharmacognosy is presented in Figure 1. An organism displaying some kind of biological activity is selected and studied to find a molecular explanation of the activity. Usually, bioassay-directed separation from the complex biomass is undertaken in order to isolate and identify the active constituent(s). This can result in the isolation of both novel and known chemical stuctures with specific bioactivity (see below). This basic knowledge can then be applied in the drug discovery process for development of new drugs, models for synthesis, precursors for semisynthesis and pharmacological tools.This model has helped us to look upon our discipline with new eyes. The task of explaining modern pharmacognosy is made a lot easier, and the role of pharmacognosy among the other pharmaceutical sciences is made more clear.

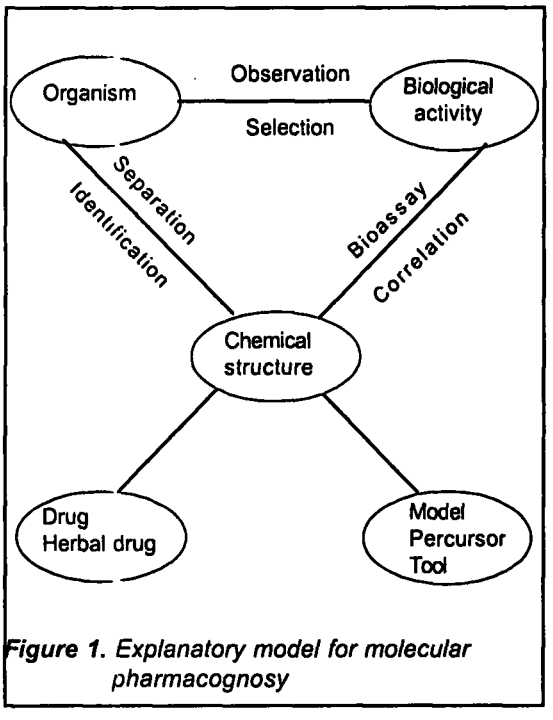

Figure 1. Explanatory model for molecular pharmacognosy

Activity and structure

The search for bioactive compounds of specific organism can result in any one of four different possibilities, some of which are more rewarding than others (Figure 2).

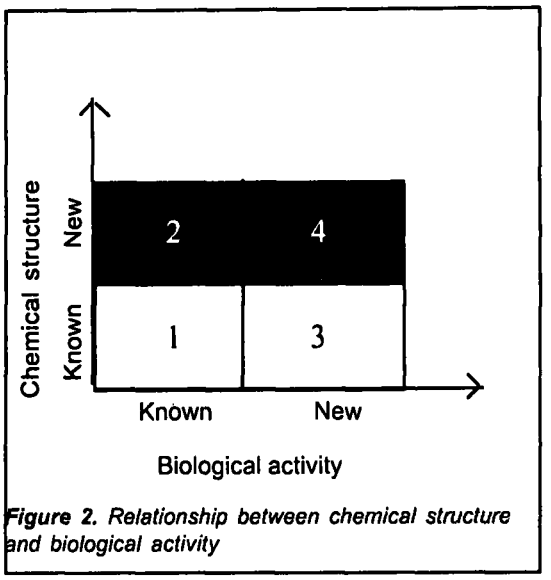

Figure 2. Relationship between chemical structure and biological activity

1: Known activity/known structure. Too often, considerable effort is expended in the isolation of previously established chemical structures with known bioactivity; for example, ubiquitous plant phenolics showing antimicrobial activity. The extract of *Ipomoea pes-caprae* used in Thailand as an antidote against skin damages caused by jelly fish toxins exhibited significant anti-inflamatory activity in experimental models. Bioassay guided fractionation led to the isolation of eugenol, a known anti-inflammatory compound[7]. However such results are important for an understanding of the activity of a specific traditional medicine and are necessary in order to be able to standardize drugs to be used in phytotherapy. As lead structures in modern drug discovery, however, such results are of minimal interest.

2: Known activity/new structure. The isolation of new chemical structures but with known activity is typical of modern research in pursuit of a specific biological activity. This can be demonstrated with the isolation of new triterpene saponins with muscle-relaxants activity, called zizyphoisides, from *Alphitonia zizyphoides*[8]. Many compounds are reported as new but are in fact modifications of already known skeletons, exemplified by our isolation of new anti-inflamatory biflavanoids from *Sarcophyte piree*[9]. In the search for new members of known mechanistic classes, for example finding a novel ligand to a certain receptor, single-specific assays can be used. However, this approach

can hardly be expected to reveal entirely new targets for drug action or new mechanisms of action.

3: New activity/known structure. A third possibility, of greater interest, would be to find a new activity coupled to a known structure. Disappointing as this may seem from the purely chemical point of view, it is a most interesting lead, and many related. Such research has long been pursued in the drug industry, where most companies routinely run all available substances through new systems. In our laboratory, a known plant constituent, a sulphoquinovosyl diacylglycerol (SQDG), was isolated from the fern *Polypodium decumanum* - used in psoriasis. The compound inhibited the exocytosis induced by platelet activating factor (PAF) in human neutrophils in vitro[10]. Further in vitro studies demonstrated that SQDG causes dose dependent displacement of [^{3}H]PAF from its receptor. This finding was quite unexpected, but shows the strength of the bioactivity-guided isolation procedure. Such findings direct attention to substance classes that may have been pharmacologically overlooked because they are widely considered to have been fully characterized.

4: New activity/new structure. The final, and most challenging, question is whether we can detect novel substances with novel mechanisms of action. If we adhere to the already established bioassays, the answer will in most cases be 'no'. The assays have been established to detect already known activities. In such circumstances, the prospects of finding new structures as models for future drugs, or as competitors or successors of existing drugs, are limited. In classical pharmacology and pharmacognosy, the effects of drug were studied in whole animals and in human experiments. These models, however basic, were able to detect new activities. During pharmacological screening in mice of the African Strychnos species, one extract induced neurological effect such as abduction of hind legs, tail erection and slight tremor of the head, in addition to its muscle-relaxant effect. This combination of effects was surprising and previously unknown. A new alkaloid, strychnocarpine, was isolated and found to stimulate 5-HT receptor, thus resembling LSD in action[11].

A careful analysis of the pharmacological pattern of natural product mixtures undergoing high-throughput screening will probably also identify new pharmacological profiles and eventually lead to novel structures.

Further research can of course uncover completely new mechanisms of activity that are no revealed in the first screening. This can be exemplified with the discovery of camptothecin with a novel structure but with the known antitumour activity to prolong life of mice treated with L1210 leukaemia cells. Futher research showed that camptothecin, by a unique mechanism, inhibits the enzyme topisomerase 1 (Ref.12). A further discussion of bioassay methods used in natural product research aimed at drug lead discovery is presented elsewhere[13].

Role of academic research

The objective for natural product research aimed at drug lead discovery is to find unique chemical structures with a new mode of action. This is a very challanging area for academic reearch, but also one of new possibilities. For the drug industry, it is most important to generate leads efficiently-rapidly and cheaply, and on the basis of a high degree of chemical diversity. It is obvious that academic research cannot proceed in this way, even if the goal is the same[14].

Academic research in the field of natural products should therefore focus on defining scientific problems for which the solution will yield fundamentally new data and insights. Our goal is to establish successful strategies in the search for specific, potentially useful natural products with biological or other activities and preferably, novel sructures.

We have therefore, reorganized our own research, on the basis of the proposed model, into two major projects: 'natural products with anti-inflammatory activity, and in the second the chemical structure. Important aspects of this research include the following:

- Development of fractionation protocols and separation method that permit isolation of minute quantities of bioactive substances from plant biomass with complex chemical composition;
- Development of selection strategies for plant materials based on biodiversity, ethnomedicine and chemotaxaonomy;
- Structure determination of isolated substances; and
- Characterization of biological (Pharmacological properties of isolated substances on organ (guinea pig ileum), cell (human neutrophil) and enzyme (COX-1, COX -2, elastase and neuropeptidases) levels.

Research activities are also focused on how to dereplicate -how to avoid the repetitive isolation of common and well known structures with known biological activity and how to prevent the emergence of artefact and interference with the bioassay applied. Other groups are formulating similar research strategies15.

Molecular pharmacognosy

Our didatic model illustrates that pharmacognosy is now a molecular science (cf. Ref. 16). Therefore, we propose the following definition based on the above:

> Pharmacognosy is a molecular science that explores naturally occurring structure-activity relationships with a drug potential.

Pharmacognosy has generally been pursuded for utilitarian ends. and this is reflected in the increased interest in herbal drugs and in natural products-based drug discovery in the industry17,18. This is augmented by the interest in the application of natural products as pharmacological tools, synthones and models in the drug development process. In our model, these to major applications form two branches of applied research (see Figure 1).

As pointed out by G.M. Cragg and others, no chemist could ever dream up a structure like paclitaxel. Learning from the paclitaxel story, the National Cancer Institute (Bethesda, MD,USA) and other research organizations now sense the necessity to develop new screens and to look upon nature as the largest and most diverse combinationatorial library available[19].

The ability of modern pharmacognosists (a) to select organisms and activities of potential interest, and (b) to avoid the pitfalls of re-isolating substances already well known pharmacologically, will determine the success or failure of the present interest in dug discovery from natural sources.

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References

1 Evans, W.C. (1996) Trease and Evans' Pharmacognosy (14th edn), W.B. Saunders.
2 Samuelsson, G. (1992) Drugs of Natural Origin (3rd edn), Swedish Pharmaceutical Press.
3 Rhodes, M.J.C. (1994) Plant Mol Biol 24, 1-20.
4 Sneader, W. (1996) Drug Prototype and their Expfoita tion, John Wiley & Sons.
5 Daly, J.W., Garraffo, M. and Spande, T.F (1993) in The alkaloids. Chemistry and Pharmacology (Vol. 43) (Cordell, G.A. ed), pp. 185-288, Academic Press.
6 Buss, A.D. and Waigh, R.D. (1995) in Burger's Medicinal Chemistry and Drug Discovery (Vol. 1 5th edn) (Wolff, M.E. ed.), pp. 983-033. John Wiley & Sons.
7 Pongprayoon, P. et. al. (1991) Planta Med 57. 515-518
8 Perera, P. et al. (1993) Magn. Res. Chem. 31, 472-480
9 Ogundaini, A et al. (1996) J. Nat Prod. 59, 587-590
10 Vasange, M., Rolfsen, W. and Bohlin, L (1997), J. Pharm Pharmacol 49, 562-566
11 Rolfsen, W., PhD Thesis. Uppsala University, 1980.
12 Wall, M.E., and Wani, M.C., (1996) Ethnopharmacol. 51, 239-254
13 Claeson, P. and Bohlin, L. Trends biotechnol. (In press)
14 Cordell, G.A. (1995), Phytochemistry. 40. 1585-1612.
15 Williamson, E.M., Okpako, D.T. and Evans, F.J. (1996), Pharmacological Methods in Phytotherapy Research Selection, Preparation and Pharmacological evaluation of Plant Material (Vol 1), John Wiley & Sons.
16 Bohlin, L. (1993), Ethnopharmacol. 38, 225-231
17 Editorial (1994), Lancet. 343, 1513-1515.
18 Press, J.B. (1996), Chemtracts-Organic Chemistry. 9, 286-298.
19 Cragg, G.M., Newman, D.J. and Snader, K.M. (1997), J.Nat. Prod. 60, 52-60.