Antimicrobial compounds as side products from the agricultural processing industry

Sumthong, P.

Chapter 8

Future perspectives in biodiversity exploration

Pattarawadee Sumthong and Robert Verpoorte

Division of Pharmacognosy, Section of Metabolomics,
Institute of Biology, Leiden University,
Einsteinweg 55, P.O. Box 9502, 2300 RA Leiden, The Netherlands

Thailand has extensive forestry, agri/horticulture and processing industries that produce raw materials and waste materials. It is interesting to add value to resources like sawdust, fruit peels, seeds, and bagasse of sugarcane. There are many possible applications of natural components from such resources, as medicines, food supplements, functional food, food additives, feed additives in animal farms, crop protectants in agri/horticulture, antibiotics in fish and prawn tanks, wood impregnation and household products. Table 8.1 summarizes possible uses of plant products and some of their associated risks. For applications in agri/horticulture and livestock, a mixture of active compounds or the crude extracts can be used to reduce costs. On the other hand, for applications in medicine this is not a possibility because they require extensive studies of toxicity and quality control, including tests for contamination with e.g. pesticides or heavy metals to assure safety in human use.

Table 8.1 The use of plant products and the consideration of contamination and toxicity in some applications.

Type of applications	Possible material used			Risks	
	Plant materials	Crude extracts	Pure compounds	Toxicity	Pesticide and heavy metal contamination
1. Medicines	+	+	+++	+++	+++
2. Food	++	++	++	+++	+++
3. Feed additives in animal farms	+++	++	+	+++	++
4. Fish and prawn tanks	+	+++	+	+	+
5. Agri/horticulture	+	+++	+	+	+
6. Wood impregnation and household products	-	++	+	+	+

+++, very important; ++, medium importance; +, important; –, not important.

The plant sources discussed above represent economically efficient sources to search for new bioactive compounds with various applications, for example, finding active compounds from fruit peels with the ability to inhibit pathogenic microorganisms in fish and prawn tanks, could increase profits without needing to increase fruit production. Selection of these plant materials may be based on local knowledge, chemotaxonomy or previous research. The activity should be proven by bioassays such as antifungal, antibacterial, antiviral or anti-nematode activity. TLC, HPLC, GC, IR, MS and NMR spectrometry can be used to identify and elucidate active compounds To know more about the interaction between compounds and living cells, investigations on their mechanism of action by advanced screening assays can be used to measure specific effects on cell walls and membranes, nucleic acids or enzymes. The safety of natural products is also important. Thus, the toxicity and possible contamination with pesticides and heavy metals should be evaluated in products having interesting activities. In the case of applications in fish and prawn tanks or feed additive in animal farms, residues such as toxic compounds, pesticides or heavy metals accumulating in animal tissues, can be harmful to the consumers. But, due to the dilution of the material in this sort of applications, risks are smaller than in cases of plant material directly used for human consumption.

Finding novel products from existing crops leads to increased economic value for these crops. From the point of view of environmental conservation there may be additional advantages, such as creating a sustainable production of materials and the use of biodegradation products. The transformation of wastes from agriculture and horticulture into new products may also solve the environmental problems of disposing of these wastes.

After screening for activities, identification of active compounds and their mode of action, including checking toxicity and monitoring for pesticide and heavy metal contamination, the next step is to optimize the extraction method in order to get a high yield of the bioactive compounds. For example the use of water, EtOH, MeOH, dichloromethane, ethyl acetate or supercritical CO_2, has to be considered in terms of yields and costs. Costs include the effect on the environment, and consequently, use of nontoxic solvents is preferred. Possibilities to improve the efficiency of extraction are method of grinding the material, use of ultrasonicator or microwaves, increasing the temperature or including additive solvents. The optimized extraction system should be suitable for large scale production. Air drying or freeze drying can be used to dry the crude extract. Metabolic profiling (with TLC, GC, HPLC or NMR) can be used to evaluate the extraction products. In the case of pure compounds, an efficient low-cost separation and isolation of those compounds should be developed, for example using a large-scale CPC (Centrifugal Partition Chromatography) apparatus.

For quality control of antimicrobial active/crude extracts, autobiography assays, paper disc diffusion assays (for non-polar fractions), well diffusion assays (for polar fractions) or broth dilution assays can be used. The end product can be developed into different formulations, for example a dry powder or a liquid with hydrophobic or hydrophilic character.

In large-scale production, the prospective fields of application and the type of plant product used (plant materials, crude extracts or pure compounds) are important parameters for choosing the processing methods. For crude extracts or pure compounds as end product, the (dry) plant materials need to be ground to a fine powder for an efficient extraction. Concerning quality control, the purity of the isolated compounds can be detected by TLC, HPLC, GC, MS or NMR spectrometry. In the case of crude extracts, a reproducible chemical profile needs to be established. Contamination with pesticides, mycotoxins and heavy metals also needs to be controlled. For crude extracts the active compounds and the ratios at which they are active, must be identified to be able to standardize the final formulation. For pure compounds the standardization of the final product is much easier. In Figure 8.1 a general flow scheme is presented for laboratory scale development of products based on the research presented in this thesis. Industrial processes are described in Figure 8.2.

Figure 8.1 Standard laboratory protocol for product development of bioactive compounds

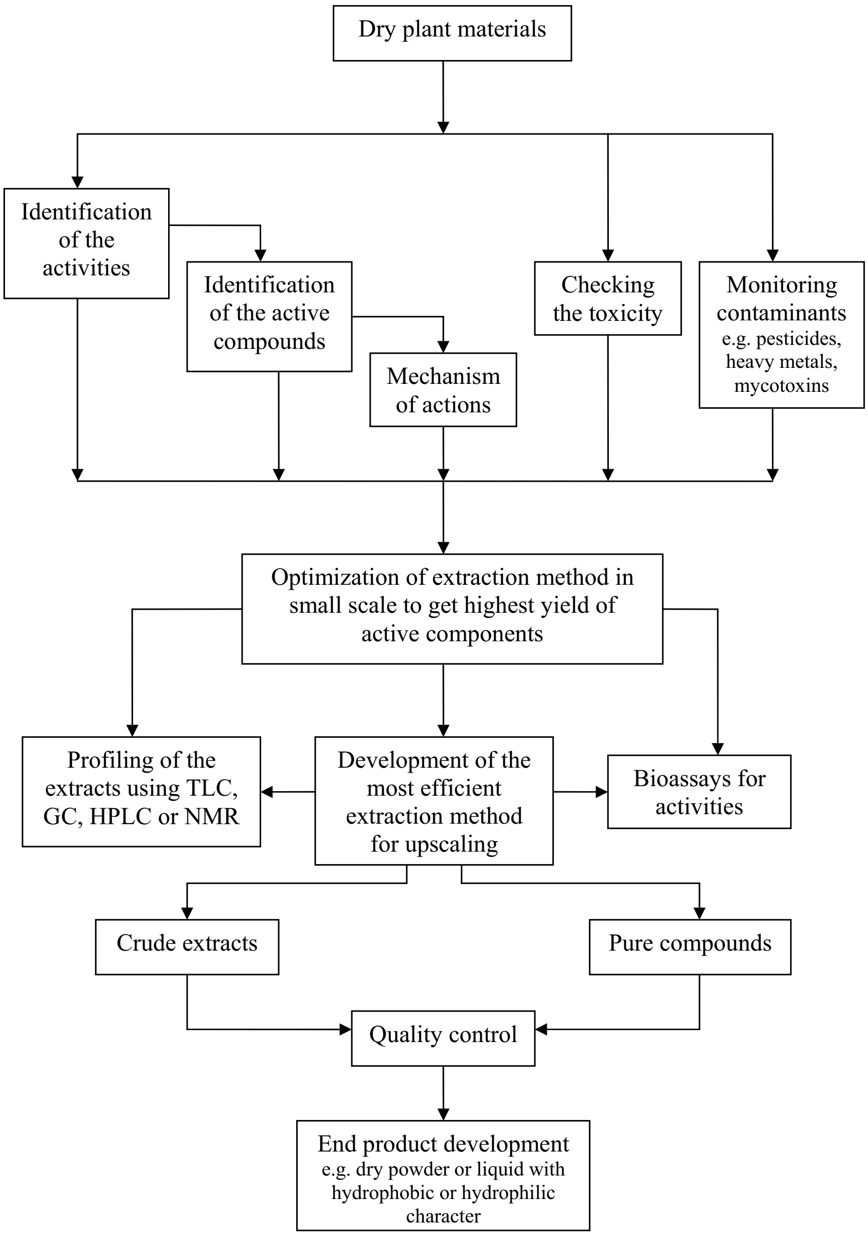

Figure 8.2 Large scale production of bioactive products