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Prospectives of botanical and microbial products as pesticides of tomorrow

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This article deals with some botanical and microbial products which have demonstrated their efficacy in the management of agricultural pests. Their merits over the existing synthetic pesticides have been discussed.

IN SPITE of the use of all available means of plant protection, about one-third of the yearly harvest of the world is destroyed by pests. Tropical countries, because of their temperature and their particular environment suffer severe losses from pests. Until 1945, the measures used against the pests were mainly based on metallic salts (frequently copper salts) and these were efficient in protecting cultivated crops only from some pest invasions. The discovery and use of DDT in 1940 and then BHC and subsequent development of the chlorinated cyclodienes marked a major advance in the field of crop protection. These chemicals have made great contributions to plant protection but have also raised a number of ecological and medical problems. The environmental problems caused by these xenobiotics have been recognized from time to time as these chemicals lace the food with residues^{1,2}. It has been estimated that hardly 0.1% of the agrochemicals used in crop protection reach the target pest leaving the remaining 99.9% to enter the environment to cause hazards to non-target organisms including humans³.

Effective pest control is no longer a matter of heavy application of pesticides, partly because of rising cost of petroleum-derived products but largely because excessive use of pesticide promotes faster evolution of resistant forms of pests, destroys natural enemies, turns formerly innocuous species into pests, harms other non-

target species and contaminates food^{2,4}. The number of fumigants for insect control are decreasing drastically as problems of insect resistance have intensified and mounting social pressures against the use of toxic chemicals have limited the introduction of new compounds. At present only two fumigants are in common use: methyl bromide and phosphine. Methyl bromide has been identified as a major contributor to ozone depletion, which casts a doubt on its future use in insect control. There have been repeated indications that certain insects have developed resistance to phosphine which is widely used today. It has been recently reported that at least 447 species of insects and mites and 200 species of plant pathogens and 48 species of weeds are now resistant to chemical pesticides. A serious re-evaluation and radical revision of the basic underlying tenets of pest control philosophy has, therefore, taken place in the past 5-10 years and the concept of pest management advocating suppression of pest populations below levels capable of causing economic injury rather than total eradication has emerged⁵⁻⁷.

The presence of organochlorine in the human body and the associated danger of long-term toxicity has resulted in national and international legislation to limit and even to ban the use of some compounds of this group. There is thus an urgent need for control agents which are less toxic to man and more readily degradable.

Basic research for over more than forty years in biology and biochemistry has made it possible to envisage not only how new pesticides may be synthesized but also a completely new approach for the protection of plants using secondary plant products which may be toxic to a specific pest yet harmless to man. There has been a re-

newed interest in botanical pesticides because of several distinct advantages: (i) Pesticidal plants are generally much safer than conventionally used synthetic pesticides. Pesticidal plants have been in nature as its component for millions of years without any ill or adverse effect on the ecosystem. (ii) Plant-based pesticides will be renewal in nature and would be cheaper. (iii) Some plants have more than one chemical as an active principle responsible for their biological properties. These may be either for one particular biological effect or may have diverse biological effects. The chances of developing quick resistance to different chemicals are highly unlikely⁸.

Some of the secondary metabolites are merely the end products of aberrant biosynthetic pathways and other excretory products. Plants synthesize a dazzling array of biologically active products. In the case of insects, various plant products affect nerve axons and synapsis [e.g. pyrethrins, nicotine, picrotoxinin, muscles (e.g. ryanodine), respiration (e.g. rotenone, mamein), hormonal balance (e.g. juvenile and molting hormone analogues and antagonists), reproduction (e.g. β asarone) and behaviour (e.g. attractants, repellents, antifeedants)].

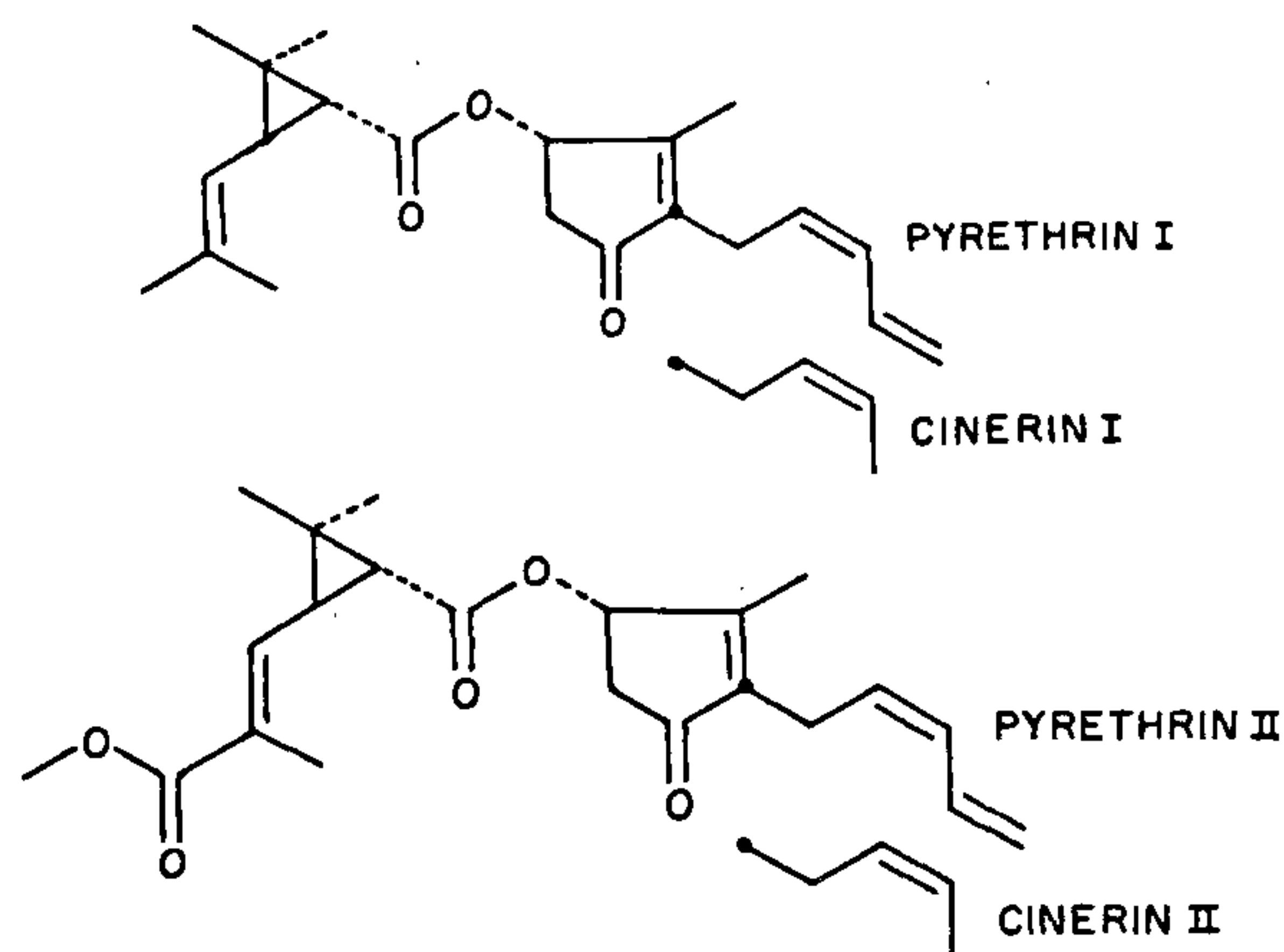
In recent years, considerable attention has been directed to the research and application of insect growth regulators, juvenile hormone analogues, antijuvenile compounds (precocenes) and phytoecdysones in plant protection. These substances have been used successfully on a large scale in plant protection. The present article deals with some botanical and microbial products which have demonstrated their efficacy in the management of agricultural pests as alternative pest management technology.

Botanical products as insecticides and fungicides

The plant world comprises a rich storehouse of biochemicals that could be tapped for use as pesticides. The toxic constituents present in the plant represent the secondary metabolites and have only an insignificant role in primary physiological processes in plants that synthesize them⁹. Plants are the richest source of renewable bioactive organic chemicals. The total number of plant chemicals may exceed 4,00,000: of these 10,000 are secondary metabolites whose major role in the plants is reportedly defensive¹⁰. Numerous defensive chemicals belonging to various categories (terpenoids, alkaloids, glycosides, phenols, tannins, etc.) which cause behavioural and physiological effects on pests have already been identified. Plants are thus not helpless when confronted with insects and other pests.

Over the past 50 years, more than 2,000 plant species belonging to different families and genera have been reported to contain toxic principles which are effective

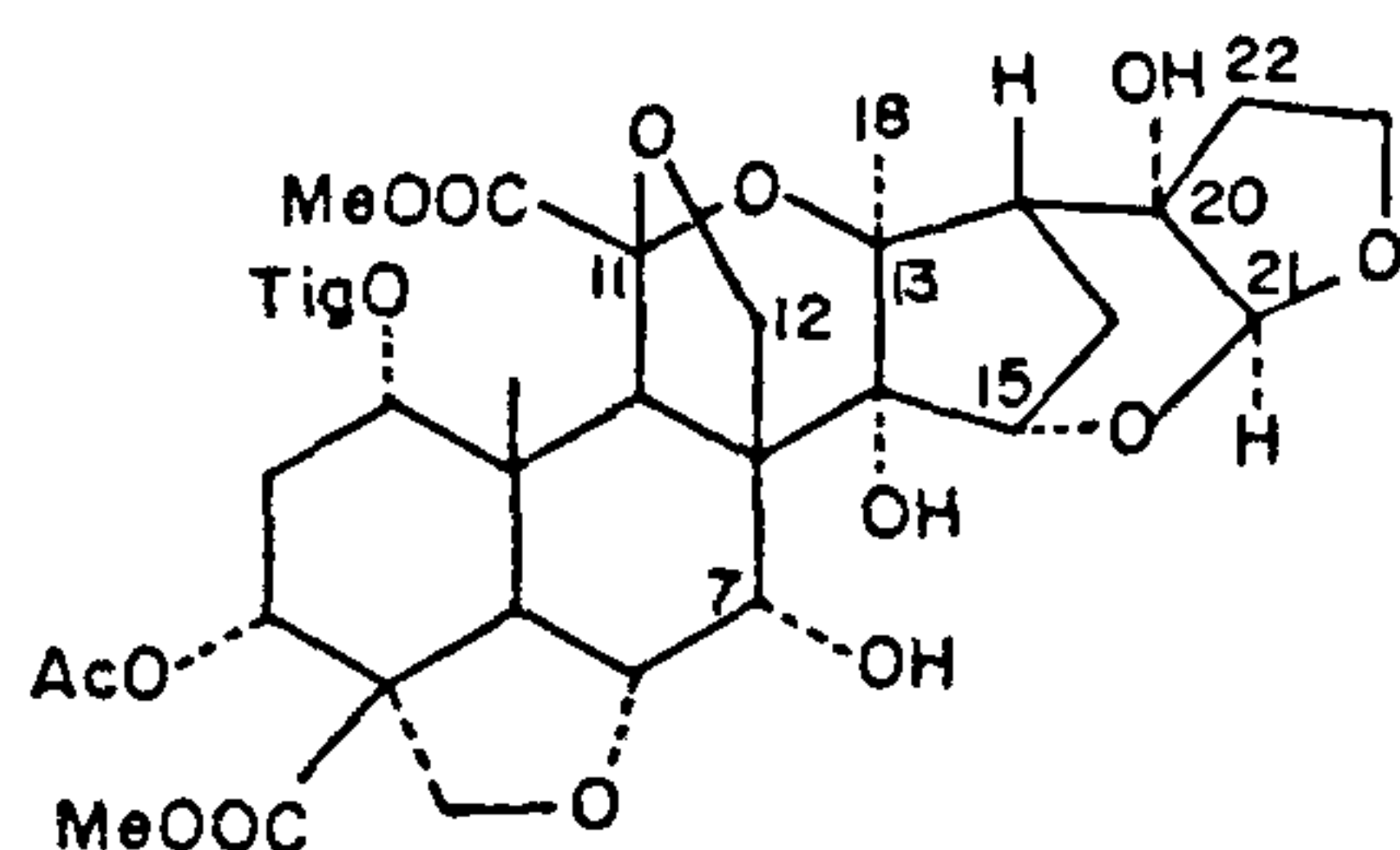
against insects. Among the well represented plant pesticides is 'Pyrethrums' obtained from *Chrysanthemum cinerariaefolium*, which is mainly used as a domestic insecticide because it is non-toxic to man and warm-blooded animals and is highly sensitive to light. There are four main principal ingredients in *Chrysanthemum*, viz. pyrethrum I and II and cinerin I and II. Pyrethrums do not leave persistent residues and have a long record of safe use¹¹.



Some of the natural pyrethroids have been used against lice and fleas in domestic as well as public buildings. It has also been successfully used against mosquitoes, house flies and other insects that spread diseases in animals and human beings. The first synthetic pyrethroid named allethrin (active moiety chrysanthemic acid) was made in 1949 but it possessed weaker knock-down effect than natural pyrethrins. Presently these are three main synthetic pyrethroids, viz. cypermethrin, deltamethrin and fenvalerate which are commonly used in agriculture in India. The presence of α -cyano group in cypermethrin makes it more light stable. Presently Kenya is the key producer of pyrethrum followed by Tasmania, Uganda, Congo and Japan. In India, the synthetic pyrethroids were first introduced in 1980 when five tonnes of cypermethrin were imported. The annual consumption of pyrethroids is more than 1,000 tonnes¹². In India, it is grown in Kashmir and Nilgiri Hills (Tamil Nadu). Some essential oils have been reported as synergists in enhancing the insecticidal activity of pyrethrum. Combination of *Blumea lacera* and pyrethrum has been compared with pipernyl butoxide and a pronounced variation in synergistic coefficient has been recorded¹³.

The pesticidal plant receiving global attention for the last two decades, is the wonder tree of Indian origin, neem (*Azadirachta indica*). Its seeds are a rich store-

house of over 100 tetranortriterpenoids and diverse non isoprenoids¹⁴. The most touted biologically active constituent of neem has been the highly oxygenated azadirachtin and some of its natural analogues and derivatives. Neem-based products have different mode of action and are medium to broad spectrum insecticides. In addition, the neem products are harmless to humans and other mammals. The aqueous neem leaf extracts have shown inhibition of DNA polymerase enzyme of hepatitis B virus. Multinational firms from Japan, Germany and UK are trying to extract an enzyme from neem which inhibits division of AIDS infected cell.



AZADIRACHTIN

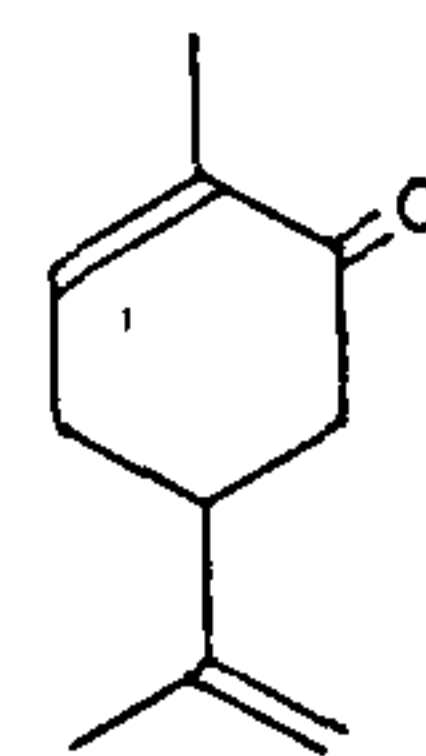
Several antifungal compounds occur in plants and it is of interest to note that leaves of certain species were used for treating fungal infections in man long before the presence of an antifungal component was suspected. *Artemisia capillaris* used for many years by paddy field workers to control fungal infections of the feet, has been shown to contain potent fungicide capillin¹⁵.

African plants used in folk medicine and in traditional pest management practices continue to yield interesting and potential useful research. The leaves of the wild shrub *Ocimum suave* and the flower buds (cloves) of *Eugenia aromatica* are traditionally used as effective stored grain protectants. Eugenol, a common constituent of the plants, was found to be a repellent to the maize weevil, *Sitophilus zeamais*, Hildecarpan, a pterocarpan from *Tephrosia* is an antifeedant against the legume pod borer *Maruca testulalis*, and the rotenoids and rotenone are very potent antifeedants against a number of lepidopteran¹⁶.

The fumigant activity of some essential oils has been evaluated against a number of stored product insects. The essential oils of *Pogostemon heyneanus*, *Ocimum basilicum* and *Eucalyptus* have shown insecticidal activity against *Sitophilus oryzae*, *Stegobium paniceum*, *Tribolium castaneum* and *Callosobrunchus chinensis*^{17,18}. Fumigant activity and reproductive inhibition induced by a number of essential oils and their monoterpenoids have also been evaluated against the bean weevil *Acanthoscelides obtectus* and the moth *Sitotroga cerealella*^{19,20}. In India, the first report on fumigant activity of essential oils was reported by Dubey *et al.*²¹

who demonstrated the efficacy of essential oils of *Ocimum canum* and *Citrus medica* as volatile fungitoxicant in protection of some spices against their post-harvest fungal deterioration. The essential oil of *Cymbopogon citratus* has shown its *in vitro* fumigant activity in the management of storage fungi and insects of some cereals without exhibiting mammalian toxicity on albino rats²². The essential oil of *Caesulia axillaris* has been found to have a strong volatile activity (fumigant activity) in the protection of wheat samples from fungal deterioration caused by *Aspergillus flavus* as well as insect deterioration caused by *Sitophilus oryzae* and *Tribolium castaneum* during *in vivo* trials. The pharmacological analysis of the oil has been done and it has not shown any mammalian toxicity to albino rats²³. After a large-scale trial, some of the oils may be recommended as substitutes for the synthetic fumigants. Unlike the prevalent fumigants, the problem due to development of resistant strains of fungi and insects may be solved by the use of essential oils as fumigant in management of stored insects pests because of the synergism between different components of the oil.

Recently carvone, a monoterpene, standardized from essential oil of *Carum carvi* has been recently introduced with the trade name of 'TALENT' in The Netherlands, which has the ability to inhibit the sprouting of potatoes during storage and it also exhibited fungicidal activity in protecting the potato tubers from rotting²⁴ without exhibiting mammalian toxicity. This is an interesting approach in exploiting the plant products in enhancing the shelf life of stored fruits and vegetables from their microbial deterioration without altering the taste and quality of the treated commodities. Some of the products may be quite economical in their exploitation.



CARVONE

Some of the plant products have been reported to cause reduction in the disease incidence when used in fruit and seed treatments. The essential oil of *Chenopodium ambrosioides* has been found to control the damping-off disease of some vegetable seedlings significantly during pot trials²⁵. Sinha and Saxena²⁶ reported that tomato fruit treated with aqueous and ethanolic extracts of neem leaves before and after inoculation with the *Aspergillus flavus* remained free from rotting up to 5 days as against 2 days in untreated control. The aqueous bark extract of *Acacia nilotica* has been found to provide complete protection to oranges

from blue mold rot infection. The oranges treated with aqueous *Acacia nilotica* have been extract, before and after inoculation with *Penicillium italicum* remained free for 10 days as against 3 days in untreated control²⁷. Twigs of *Acacia nilotica* have been used by Indian villagers as tooth brushes since long and there would be no possibility of mammalian toxicity, if *A. nilotica* is exploited as pesticidal plant.

It has been estimated that only 5 to 15% of the 2,50,000 to 7,50,000 existing species of plants have been surveyed for biologically active compounds and even this is an over estimate, as the investigated plants have been partially screened for a single or at best, few types of activity. Due to rapid deforestation, phytologists may have only few decades left for surveying and cataloguing the fast extinguishing flora²⁸.

Antifeedants

Antifeedants are substances which when tasted can result in cessation of feeding, either temporarily or permanently, depending upon the potency. Absinthin, a dimeric sesquiterpene, obtained from *Artemisia absinthium* has been reported to exhibit antifeedant activity against a number of insects.

Azadirachtin isolated from *Azadirachta indica* and *Melia azadarach* induces pronounced morphological changes in *Antestiopsis* (coffee bug) and desert locust *Schistocera gregaria*²⁹. It is also a systemic growth disruptor³⁰. Ajugarins are the moderately effective antifeedants isolated from the leaves of *Ajuga remota* (Labiatae) which are not attacked by African army worms. They are reported as insect antifeedants. Two antifeedants against the larva of African army worm (*Spodoptera exempta*), have been isolated from the petroleum ether extract of *Clausena anisata* (Rutaceae). The compounds have been identified as the coumarins imperatorin and xanthoxyletin³¹. Withanolides, extracted from solanaceous plants belonging to the genera *Withania*, *Acnistus*, *Physalis*, *Jaborosal* and *Datura*, are reported as antifeedants³¹.

Antifeedants should have the quality of systemicity otherwise as the plant grows, the insect will soon find a place (area) on which the chemical has not being applied. This would also destroy the internal feeders and may provide a chemical resistance to the plant after application. The azadirachtin has the systemic property as it also protects the newly growing leaves of the crop plant from feeding damage³³.

Repellent and attractant chemicals

For a long time various folk remedies have been used to repel pests with extracts from plant. The most effective group of repellents includes amine and pyridines which are derivative of quaternary ammonium bases like cyclic

amides and rosins. The oils of Citronella, an extract of *Andropogon* sps. and *Cymbopogon vardus* have been claimed to be particularly effective as mosquito repellent³⁴. Attractants may be used to lure the insects to a place where they can be destroyed by physical means, heavy dosage of chemicals or attracting them to a false host plant. The essential oil of *Pogostemon parviflora* and its isolated components showed varied degree of repellancy/attractancy against *Sitophilus oryzae* and *Bruchu schinensis*. The difference in the activity of oil and its component at particular concentration may be because of the masking of the activity of essential oil active component by other components present in the oil.

Moreover, the plants containing the compounds which can attract the insects may be used as trap tree operation for control of insect invasion in forest trees. For this, one or two trees are selectively felled in an area and are cut into two or three meter long logs. The sap oozing out from the end of the bark may attract insects from the surrounding trees. The attracted insects may be killed in different ways.

Insect growth regulator

The insect growth regulators (IGRs) are generally selective in their toxicity, slow in their killing action, and safe to both the environment and the natural enemies of insect pests. This is the outcome of biorational approaches to exploit physiological and biochemical processes unique to insects or arthropods with the aim of providing biochemical leads for the discovery of unique new chemical control agents.

In agriculture and forestry, IGRs are generally compatible with the agroecosystem and possess great potential in the integrated pest management programme. The rational application of these materials may lead to new strategies for the prevention and delay of occurrence of insect resistance problem.

Plants offer a rich source of novel compounds with IGR properties. The ample potential in using phytoecdysones as IGRs cannot be overlooked. It has been reported that there are more than 1,000 species of plants (belonging to more than 200 families, 800 genera) containing these bioactive substances. Isolation of Ajugarins from *Ajuga remota* is a good example³⁵. Research is now underway in China to formulate enriched products from plants with high IGR properties for practical application in the field. The chemistry and mechanism of toxic action of these plant products to insects are also being studied.

Juvenile hormone

If insects undergoing metamorphosis are treated with juvenile hormone (JH), they moult into intermediates

which are half immature and half adult, which die without gaining reproductive competence.

JH and analogues are also present in some plants. One of the first observations was made by Carol Williams³⁶ who noted that the water of Rio Negro, was rich in suspended organic materials from wood decay and free from mosquito larvae as do other rivers of the Amazon basin. JH analogues have been found in various plants and it may interfere to some extent with the control of several species of insects.

There have been numerous reports on the occurrence of plant metabolites having JH activity, i.e. phytojuvencoids such as juvabione, farnesol, juvocimene I and II, sesamin and sesamol, thujic acid, sterculic acid, tagetone, ostruthin, echinolone, baku-chiol, juvadezene. The first JH analogue was discovered by chance from paper and was termed as paper factor (third generation pesticides). The compounds isolated from paper made from Balsam fir (*Abies balsamea*) were identified as juvabione and dehydrojuvabione^{37,38}.

Precocenes

These are also known as antijvenile hormones. A number of chromone derivative precocenes were isolated from *Ageratum haustonianum*^{39,40}. They are anti allatotrophic, i.e. they prevent JH synthesis. This accelerates the development of insects, producing dwarf sterile animals which are unable to survive. The open chain analogue of precocone, β -asarone which is a constituent of the rhizome of sweet flag (*Acorus calamus*) showed antigonadal activity in insects. The vapours of the oils have exhibited complete inhibition of ovarian development when given to a number of stored grain insects. Such oils may be utilized as chemosterilants in plant products and the problem of development of physiological races of the pests may be minimized. This would be a noteworthy merit of the biorational chemicals over synthetic pesticides.

Microbial exploitation as pesticides

After 1950, bacteria were used to control insect pest population. Over 90 species of bacteria have the ability to kill insect pests. Bacterial toxins have been shown to be effective weapons against many insects. But only toxins of *Bacillus thuringiensis* Berling var. *kurstak*. have given positive results in the field against lepidoptera. Much of the success lies in the ways in which it can be considered as similar to chemical insecticides. *B. thuringiensis*, a gram-positive bacteria, produces a proteinaceous parasporal crystalline inclusion during sporulation. This crystal structure is non toxic to insects. Upon ingestion by insect, this prototoxin, due to high pH in the stomach of the insects, cleaves the protein into smaller subunits called δ endotoxins. The activated

toxins interact with the larva midgut epithelium causing a disruption in membrane integrity and ultimately leading to instant death⁴¹. Insects that do not have a high pH in the gut are incapable of this reaction. Likewise mammals which have an acidic pH in the stomach, are not capable of breaking the prototoxin down into smaller units.

The most predominant *B. thuringiensis* product is BTK and is sold under several trade names (i.e. Dipel, Thuricide, Bactospeine, Biobit, etc.). There are currently 410 registered formulations of *B. thuringiensis* and 6 registered formulations of *B. lentimorbus* and *B. popillae* approved for use against insect pests.

The spores are partially dominant BTK formulations. But once inside the gut in the proper environment, they germinate and pass through the gut wall. This also contributes to the eventual death of the insects, over the next two or three days.

Baculo viruses as pesticides

Baculo viruses constitute one of the largest and diverse groups of insect pathogenic viruses. Use of these viruses as pest management has been widely accepted. Baculo viruses are products of evolution and have been in contact with insects and the environment for a much longer time than the man-made pesticides. Such viruses are rod-shaped, enveloped, double-stranded DNA viruses. Naked non-occluded virions are treated very harshly in the environment but the protein crystal that surround the baculo viruses protects them from solar radiation, drying, changes in pH, etc. Nuclear polyhedrosis virus (NPV) plays a major role in pest control. NPV, a stomach poisoning agent is used against tobacco caterpillar and American bollworm. A variety of NPVs specially those of *Spodoptera*, *Trichoplusia* and *Heliothis* spp. of *Chilo infuscatellus*, codling moth, cabbage butterfly are widely used for control of vegetable and field crop pests in Central and South America, some countries in Africa, some states in India, Thailand, Malaysia, China, etc⁴².

Semiochemicals

The odour of specific chemicals that deliver behavioural message has been termed semiochemicals. Semiochemicals used for intra-specific communications between individuals of a single species are termed as pheromones⁴³⁻⁴⁵. The semiochemicals that mediate inter-specific interactions between the members of different species are termed allelochemicals.

Pheromones

Pheromones were initially called 'ectohormones' since like the hormones they exerted an effect at some dis-

tance from the point of release. They are exocrine secretions and are used by insects for a variety of purposes like finding mates (sex), aggregation, alarm, release, tracking or trail marking defence, etc. They are of different types as sex pheromones, alarm pheromones, aggregate pheromones or arrestants, territorial pheromones, epidectic pheromones.

In India research on insect pheromones started late. Most of the research work is done in the advanced countries especially USA, UK, FRG and Australia. As a result of this extensive work, about 670 chemicals with pheromone-like activity have been isolated. In light of recent advances, pheromones seem to fulfil their promise as components of integrated pest management programme.

The pheromones related with the plant protection are release pheromones, i.e. they elicit a behavioural response. Pheromones are smelt, i.e. they are perceived through chemoreception. Sex pheromones that initiate and control the mating behaviour can also be utilized in plant protection. Since these are species-specific and are secreted in minute quantities, their use in pest control will be highly selective, effective and least disruptive to the environment. Pink bollworm (*Pectinophora gossypiella*) is an important pest of cotton crop and causes heavy damage. As many as 15 to 20 insecticide sprays are resorted to check the pest nuisance. Use of sex pheromone, 'gossyplure' is found to be very effective and encouraging results have been obtained⁴⁶.

Sex pheromones are relatively simple molecules whose synthesis on a large scale seems feasible because of their efficacy at very small dose and species-specific action. They constitute a novel biorational measure in control of insect pests. Insect pheromones are non toxic to vertebrates and their practical application seems to be easier. Pheromonal communications amongst the insects can be exploited to manage insect pests.

Allelochemicals and herbicides

Allelochemicals that inhibit the germination and growth of weed species and are produced and released by growing crop plants or their residues, are receiving increased attention for use as weed controls and as herbicides. The exclusive presence of certain weed species in cultivated fields may be the result of selectively inhibitory substances produced by the crop plant. These chemicals are present in virtually all plant tissues and are released in a variety of ways such as volatilization, leaching, root exudation and decomposition of plant residues. Natural products identified as allelochemicals represent a myriad of compounds such as hydrocarbons, organic acids, terpenoids, steroids, unsaturated lactones, phenolics and polyphenolics. Some special terms such as phytonicide (an inhibitor produced by a higher plant

and effective against microorganisms), marasmin (a microbial secondary metabolite toxic to plants), koline (secondary metabolites of higher plants detrimental to higher plants) and antibiotic (microbial metabolite effective against microorganisms) are also referred with allelochemicals⁴⁷.

Several allelochemicals and microbial metabolites have the potential for use as herbicides and have provided structural models for herbicide development. Based on benzoic acid, a few halogenated derivatives have been also developed as herbicides⁴⁸. Cinmethylin, a cineole alcohol with the addition of substituted benzyl group represents a new class of herbicides which control grass and some broad leaf weeds. Shettel and Balke⁴⁹ reported that the common allelochemicals, viz. salicylic acid, *p*-hydroxy benzoic acid, hydroquinone and umbelliferone effectively suppressed the growth of several weeds when applied as spray. Unfortunately, they were not selective. Identification of the phytotoxins produced by weed pathogens could provide leads for the development of two strongly phytotoxic metabolites; prehelminthosporal and dihydro prehelminthosporal isolated from the culture filtrates of the fungus *Bipolaris* sp. showing herbicidal activity against Johnson grass (*Sorghum halepense*), one of the worst weeds in the tropical and subtropical areas of the world². Bialophos has been isolated from *Streptomyces viridochromogene* and has been marketed in Japan as a herbicide. The fact that microbial toxins may also have a broad spectrum of effects beyond plants demands that care must be taken in the utilization of such compounds.

Nematicidal property

Some plants like *Tagetes*, garlic, lemon grass and *Crotalaria* sps. have been widely used in inter cropping and have shown promising nematicidal property. When lemon grass was planted in suburbs in Rio de Janeiro, human reinfection by nematodes of the Ancylostomadae in paddy fields occurred in only 12% of the population in one year and a half compared with 50% reinfection observed in similar period in an adjacent unplanted area. A serious study of the intercropping with the lemon grass, *Vetiver* or one of the mints in potato, tomato and other nematode susceptible cultivation would seem to be justified as an alternative to the use of synthetic nematicides. The intercrop plants would themselves be commercially exploitable⁵⁰. Neem cake applied at the rate of 1225 kg/ha is reported to be effective against *Meloidogyne incognita* in tomato and egg plants⁵¹. The treatment improved the yield without ill effect on quality. Besides cake, other parts of the neem tree have also been reported to affect nematodes. Incorporation of leaves of *Chrysanthemum* sps., *Melia azadirach*, *Tagetes patula*, *Datura fastuosa* and *Nerium*

indicum reduced the incidence of *Meloidogyne* sps. after six week of decomposition⁵².

Conclusion

Biologicals because of their natural origin, are biodegradable and they do not leave toxic residues or byproducts to contaminate the environment. Several plants have thousand years of history and the nontoxicity at least on oral level is proved. This safety feature is very important because it has an impact to a larger extent on the cost of development and registration of a new pesticide product. The research and development cost of botanical pesticides from discovery to marketing is much less compared to chemical pesticides. Most chemical pesticides have a long 7–10 year development period and registration time frame with registration costs ranging from \$20 to \$30 millions. This expense is in large part, due to the concern over possible high animal toxicities of such materials that necessitates long term toxicological testing on experimental animals. Biologicals because of their high target specificity typically require only short-term toxicological tests^{53,54}.

With the modern techniques now available and the attention being given to this area, we look forward to interesting developments on biological activity of botanical and microbial products so as to exploit them as pesticides in a world where one is in constant fear of a rupture of ecosystems and of undesirable consequences for human health from excessive use of non biodegradable pesticides in the protection of food. A proper study of the mode of action and structure–activity relationship will bring about a new class of interesting compounds for future pest control. Conservation of plants having biorational activity needs quick survey and screening before they are destroyed by deforestation. Plant cell cultures are now being used for the production of valuable secondary metabolites. Several secondary products of pesticidal interest such as alkaloids, pyrethrins, terpenoids and phytoalexins have been detected in cell cultures. Recently, the feasibility of generating transgenic crops by using *B. thuringiensis* insecticidal crystal protein has been demonstrated.

The naturally occurring pesticides thus appear to have a prominent role in the development of future commercial pesticides not only for agricultural crop productivity but also for the safety of the environment and public health⁴⁷. We in India are favourably placed with regards to the availability of different pesticidal plants for the manufacture of effective pesticidal products for use in agriculture. Some foreign firms are keen to produce botanical pesticides, but Indian firms should be encouraged in this direction by imposing some relaxations in registrations of such pesticidal products. It would be justified to mention here that although Indian agricultural scientists had indicated the powerful antifeedant

activity of neem kernels, the opportunity was, however, taken up in earnest by the West; culminating in the formation of azadirachtin.

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RESEARCH ARTICLE

Mean distance of closest approach of ions in NaCl (aq.) at 25°C calculated from degrees of association using Bjerrum's theory

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This paper demonstrates, for the first time, the applicability to 1:1 strong electrolytes of Bjerrum's pioneering theory, which relates the degree of ionic association with the mean distance a of closest approach of ions. It can be noted that only recently, strong electrolytes like NaCl were shown to be incompletely dissociated in aqueous solutions. The pre-

sent data on the degrees of dissociation of NaCl (aq.) at 25°C have been used to calculate the values of a for concentrations from zero to saturation, using Bjerrum's theory. It is found that both $Q(b)$, the Bjerrum's integral, and a are simple functions of the molal volume and concentration and that at saturation, a is less than q , the critical distance.

It was generally believed¹⁻³ that 1:1 strong electrolytes like NaCl dissociate completely into ions in aqueous solutions. However, recent X-ray diffraction and mo-

lecular dynamic simulation studies of saturated solutions^{4,5} and investigation of thermodynamic data at all concentrations^{6,7} have shown that even 1:1 strong elec-