

Allelopathic Compounds as Naturally Occurring Herbicides

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Abstract

Allelopathy is the result of biochemical interactions between plants. It is caused by toxic chemicals which are released by the plant through volatilization, leaching, and root exudation or produced during the decomposition of plant residues in soil. Phytotoxins, including fatty acids, phenolics, flavonoids, terpenoids and alkaloids have been found in plants and soils from different habitats around the world. Since 1972, we have reported that allelopathic metabolites are released from woody plants, such as *Phyllostachys edulis*, *Leucaena leucocephala*, *Vitex negundo*, and *Delonix regia*, to suppress the growth of understory species, causing a significant reduction of biomass and a low diversity of understory plants. The development of dominance of the tree species mentioned above is due primarily to the phytotoxic effects of metabolites released from plant parts, such as leaves, flowers, and twigs, and the decomposition of fallen litter. A pasture grass-forest intercropping system was established by introducing kikuyu grass into a deforested region where coniferous trees or hardwood plants were allowed to regenerate. Because of the allelopathic potential of the kikuyu grass, weeds grown in the deforested land were suppressed by the grass six months after the grass was planted; the growth of regenerated forest plants was not suppressed, however, but was stimulated. This example clearly demonstrated that allelopathic compounds can be used as natural herbicides to control weed growth in agricultural practice in order to reduce the use of synthetic herbicides, avoid agrochemical runoff and reduce high labour costs.

Key words: allelopathy, phytotoxin, phenolics, flavonoid, alkaloids, sustainable agriculture, allelochemicals

Introduction

Secondary plant metabolites include a variety of compounds which, when released from plants into the environment often attract or repel, nourish or poison other organisms. Yang and Tang (1988) made an extensive review of plants used for pest control as described in Shengnoon Ben Tsao Jing in 25-220 A. D. in China. They described 267 plants containing pesticidal activity, many of them also exhibiting an allelopathic potential. In 1832, De Candolle, a Swiss botanist, suggested that the soil sickness problem in agriculture might be due to exudates of crop plants (Rice, 1984). Whittaker and Feeny (1971) clarified that "chemical agents are of major significance in adaptation of species and organization of communities". Since then, secondary metabolites have no longer been considered as metabolic waste, but play an important role in plant adaptation and plant-insect coevolution (Chou and Waller, 1989; Ehrlich and Raven, 1965; Harborne, 1977). Allelopathy, a detrimental biochemical interaction between plants, plays a direct or indirect role in the mechanism of plant dominance, succession, and climax, and in the regulation of crop productivity, genetic diversity, and ecosystem stability (Muller, 1966; Rice, 1984; Chou and Waller, 1989; Chou, 1993). For example, the compounds produced from California chaparral vegetation, such as *Salvia leucophylla*, and *Artostaphylos glandulosa* var. *zacaensis* (Muller, 1966; Muller and Chou, 1972), suppress the growth of the understory or nearby plants. The compounds are released into the environment of four ecological processes: volatilization, leaching, decomposition of plant residues in soil, and root exudation.

In the past decades, agricultural practices drastically changed by the application of an increased amount of fertilizers and agrochemicals, such as herbicides, fungicides, and pesticides into fields. The continuous use of agrochemicals in farms may cause severe environmental problems, such as loss of efficiency of nitrogen fixation in soil, disruption of the balance of soil microorganisms, residual effect of agrochemicals, eutrophication of water reservoirs, or water pollution. As a result of long-term application of agrochemicals into the fields, certain dominant soil microorganisms occur, resulting in a pathogenic effect on crop growth (Patrick, 1971; Wu *et al.*, 1976 a and 1976 b). The concept of sustainable agriculture, which involves the use of organic, regenerative, biodynamic, intensive, low-input systems and resource-conservation has been much emphasized since 1980 (Francis and Sahs, 1986). Alternate agricultural practices, such as organic farming, biological control, and crop rotation were recommended to solve the problem. In biological control, many natural plant growth regulators, such as agrostemin can be used for weed control (Gajic and Nikocecic, 1973). Other allelochemicals were also used as fungicides, insecticides, and nematocides (Rice, 1984; Waller, 1987; Rizvi and Rizvi, 1992), and compared with man-made agrochemicals, damage to the global ecosystems was reduced. Efficient use of farm resources relies on a minimum purchase of inputs, and minimizes the influence of agricultural practices beyond the farm boundaries (Chang, 1992; Gliessman, 1986). Putnam and Duke (1974) selected for allelopathic activity when breeding weed-controlling cultivars of cucumbers. Since 1972, Chou and co-workers have conducted experiments to elucidate the role of allelopathy in agroecosystems in Taiwan and they identified many potential allelochemicals useful for agricultural practices (Chou, 1992). This paper is thus focused on allelopathic compounds isolated from local vegetation with a herbicide potential.

Secondary metabolites as potential naturally occurring herbicides

Many secondary plant metabolites, such as phenolics, flavonoids, alkaloids, terpenoids, and cyanogenic glycosides, have often motivated scientists, particularly organic chemists, to study their structure, biosynthesis, and natural distribution, but not their function until Frenkel (1959) and Whittaker and Feeny (1971) pointed out the important functions of the secondary metabolites in plants and ecosystems. Waller and Nowacki (1975) note that plants produced high levels of alkaloids which are toxic to many organisms when the plants grew in soil with a low nitrogen content. In addition, Koeppel *et al.* (1976) found that significantly larger amounts of allelopathic substances were also produced when plants grew in phosphorus-deficient soil. There is evidence that the secondary plant metabolites are often stored in vacuoles or in intercellular spaces when they are not used. The compounds are released into the environment by the four ecological processes mentioned previously (Chou, 1989). Based on the nature of allelopathic compounds isolated from each specific plant, the compounds are grouped into phenolics, flavonoids, and alkaloids as described below.

Phenolic compounds

A unique pattern of weed exclusion by bamboo vegetation is often found on many hillsides of mountains at elevations below 1500 m in Taiwan. Fourteen bamboo species were selected for evaluation of allelopathic potential. Among 14 bamboo species, *Sinocalamus latiflorus* showed the highest phytotoxicity. and *Bambusa oldhami*, *B. pachinensis*, *B. ventricosa*, *Phyllostachys edulis*, and *P. makinoi* revealed a significant phytotoxicity (Chou and Hou, 1981; Chou and Yang, 1982). The allelopathic substances isolated from the bamboo leaves and soil were identified as phenolic acids, namely, *o*-hydroxyphenylacetic, *P*-coumaric, *P*-hydroxybenzoic, ferulic, vanillic, and syringic acids (Table 1 and Fig. 1). Most of these phytotoxic phenolics are also distributed in 12 subtropical grasses (Table 1) (Chou and Young, 1975), and in *Miscanthus* species (Chou and Chung, 1974; Chou and Lee, 1991).

Moreover, Chou and Chen (1976) also evaluated the allelopathic potential of 25 woody species commonly occurring in northern Taiwan and 5 major compounds similar to those mentioned were present in leaves and litter of the plants.

In leguminous plantations of *Leucaena leucocephala*, there is an absence of understory growth other than itself, due primarily to the phytotoxins, including eight phenolic acids, namely gallic, protocathechuic,

p-hydroxybenzoic, *p*-hydroxyphenylacetic, vanillic, caffeic, *p*-hydroxycinnamic and ferulic acids released from its leaves and litter (Table 1 and Fig. 1). The compounds can suppress the growth of many weeds and forest species, such as *Acacia confusa*, *Ageratum conyzoides*, *Liquidambar formosana*, *Casuarina glauca*, *Mimosa pudica* and *Alnus formosana* (Chou and Kuo, 1987).

Flavonoids

Vitex negundo a dominant coastal vegetation, is widely distributed in the southern parts of Taiwan. Chou and Yao (1983) found that the biomass and density of its associated understorey were reduced compared with those in an adjacent pasture. Field results showed that, compared to rainwater control, the leachate of *V. negundo* significantly retarded the growth of *Digitaria decumbens* but stimulated the growth of *Andropogon nodosus*. The growth of *D. decumbens* in pots under greenhouse conditions was significantly retarded by watering with a 1% aqueous extract of *V. negundo*, whereas the growth of *Andropogon nodosus* and *M. pudica* was stimulated. The aqueous extract was phytotoxic to lettuce and ryegrass seeds. The aqueous effluents from a polyamide column chromatograph were bioassayed; some fractions inhibited the growth of lettuce and rice seedling radicles, whereas other fractions stimulated growth. The

Table 1 Presence (+) of phytotoxic phenolic compounds as naturally occurring herbicides in grasses and woody plants in Taiwan

| Compounds | Grasses | | | | | Woody plants | | | | | |
|------------------------------------|-------------------|----|----|----|----|--------------|----|----|----|----|----|
| | Dd ⁽¹⁾ | Mf | Mt | Pc | Pe | Af | Ca | Cl | Dr | Ll | Zf |
| Caffeic acid | | | + | | | | + | | | + | |
| Cholorogenic acid | | | | | | | + | | + | | |
| Cinnamic acid | + | | | + | | + | | | | | + |
| <i>o</i> -Coumaric acid | | | | | | | | | | | + |
| <i>m</i> -Coumaric acid | | | | + | | | | + | | | |
| <i>p</i> -Coumaric acid | + | + | | + | + | | + | | | + | |
| 3, 4-Dihydroxybenzaldehyde | | | | | | | | | + | | |
| 3, 4-Dihydroxybenzoic acid | | | | | | | | | + | | |
| 3, 4-Dihydroxycinnamic acid | | | | | | | | | + | | |
| 3, 5-Dinitrobenzoic acid | | | | | | | | | + | | |
| Ferulic acid | + | + | + | + | + | | | + | | + | + |
| Gallic acid | | | + | + | | | | + | + | + | |
| <i>p</i> -Hydroxybenzoic acid | + | + | + | + | + | | + | + | + | + | |
| <i>p</i> -Hydroxyphenylacetic acid | | | | | | | | | | + | |
| <i>o</i> -Hydroxyphenylacetic acid | + | + | + | | + | + | | | | | |
| Phloridzin | | | + | | | | | | | | |
| Protocatechuic acid | | | | + | | | | + | | + | + |
| α -Resorcylic acid | | | | | | + | | | | | |
| β -Resorcylic acid | | | | | | | | | | | |
| γ -Resorcylic acid | | | | | | + | | | | | + |
| Scopoletin | | | | | | | + | | | | |
| Syringic acid | + | + | | + | + | | + | + | | | |
| Vanillic acid | + | + | + | + | + | + | + | + | | + | + |
| 4-Hydroxycoumarin | | | + | | | | | | | | |

(1) Abbreviations of each plant name are: Dd: *Digitaria decumbens*

Mf: *Miscanthus floridulus*, Mt: *M. transmorrisonensis*

Pc: *Pennisetum clandestinum*, Pe: *Phyllostachys edulis*

Af: *Alnus formosana*, Ca: *Coffea arabica*, Cl: *Cunninghamia lanceolata*, Dr: *Delonix regia*

Ll: *Leucaena leucocephala*, Zf: *Zelkova formosana*.

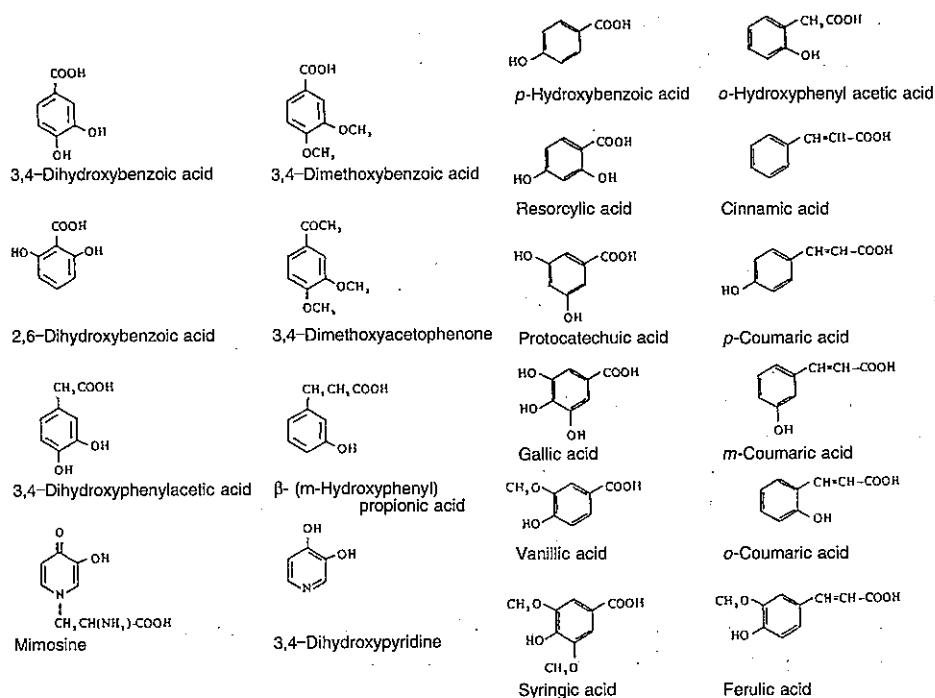


Fig. 1 Chemical structure of potential allelopathic phenolics isolated from plant parts mentioned in the text

responsible substances, isolated and identified, consisted of several phenolic acids and ten flavonoids, including 3'-hydroxyvitexin and its derivative (Fig. 2) (Chou and Yao, 1983). These metabolites could be used as herbicides.

In connection with a chemosystematic investigation of *Agastache pallidiflora*, Chou *et al.* (1979) identified 6 flavonoids, namely kaempferol, quercetin 3-rhamnoside, kaempferol 3-galatoside, diosmetin 7-glucoside, and acacetin 7-glucoside. The alycone moiety of flavonoids may exhibit allelopathic nature, although the biological activity has not been fully confirmed.

Alkaloids

The mature leaves of *Leucaena* contain about 5% dry weight of mimosine (Fig. 1), the amount varying with varieties. The seed germination and radicle growth of lettuce, rice, and ryegrass were significantly inhibited by aqueous mimosine at a concentration of 20 ppm, while that of the forest species mentioned was suppressed by the mimosine solution at 50 ppm or above. However, the growth of *Miscanthus floridulus* was not suppressed by a mimosine solution at 200 ppm (Chou and ; Kuo, 1986). Of eighty-four *M. pudica* seedlings tested, only two seedlings survived, showing that mimosine can be of practical use in the control of field weeds.

Recently, Chou and Leu (1992) have reported that another plant, *Delonix regia*, allelopathically excludes understorey species. The aqueous extracts of the leaves, flowers, and twigs of *D. regia* exhibited a significant phytotoxicity (over 70%) against tested species such as *Isachne nipponensis* and *Centella asiatica*. The responsible allelopathic substances are 3, 4-dihydroxybenzaldehyde and the acids: 4-hydroxybenzoic; chlorogenic; gallic; 3, 4-dihydroxybenzoic; 3, 4-dihydroxycinnamic; 3,5-dinitrobenzoic (Table 1. Figure 1); and L-azetidine-2-carboxylic (Fig. 3). In addition, some unidentified flavonoids are present in the plant.

Furthermore, regarding the allelopathic compounds isolated from *Coffea arabica*, there are 7 phenolic compounds and 4 alkaloids, namely caffeine, theobromine, theophylline, and paraxanthine (Fig. 3). At the concentration of 100 ppm in aqueous solution, the alkaloids exhibited a significant phytotoxicity to test plants (Chou and Waller, 1980). Waller *et al.* (1989) further studied the action model of these alkaloids

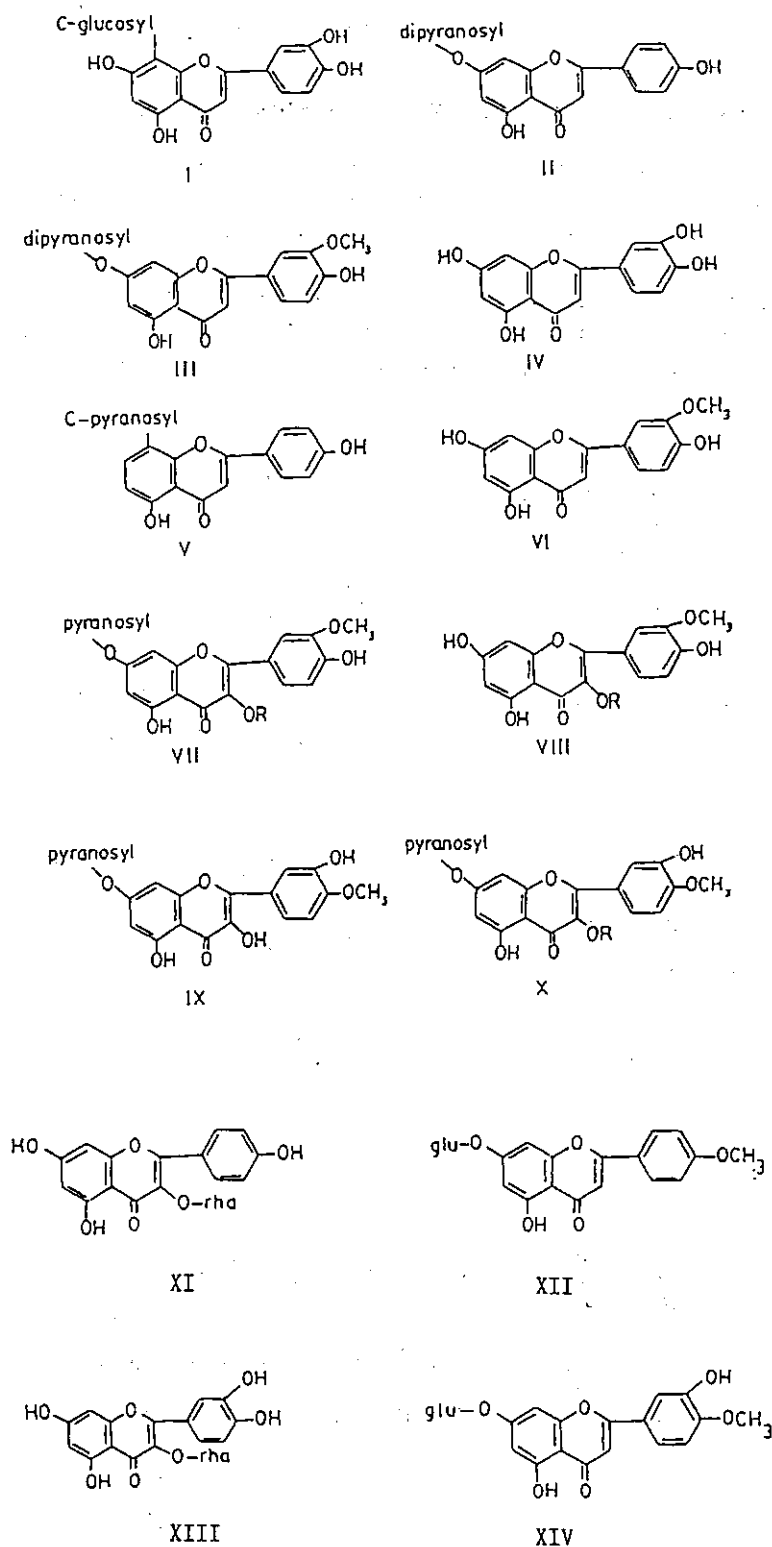


Fig. 2 Chemical structure of potential allelopathic flavonoids (I - X) isolated from *Vitex negunda* and flavonoids (X I - X IV) isolated from *Agastache pallidiflora*

Compounds X I to X IV are X I = Kaempferol 3-rhamnoside

X II = acacetin 7-glucoside

X III = quercetin 3-rhamnoside

X IV = diosmetin 7-glucoside

and interesting findings were associated with the biosynthesis of protein and nucleic acids.

Allelopathy in pasture-forest intercropping systems

Taiwan is an island, of which two-thirds consist of mountains, and its forests are extremely important for water conservation. The limited amount of agricultural land relegates farming activities to the hills and to higher elevations. A forest-pasture intercropping system is considered to enable to solve the problem and to increase livestock production. Recently we have conducted several experiments in the forest area of Hoshe Experiment Station of National Taiwan University, located at an elevation of about 1200 meters (Chou *et al.*, 1987). A one-hectare area was deforested. The leaf litter of the conifer tree *Cunninghamia lanceolata* was removed from one part, and the rest was left unchanged, as the control. Half of the

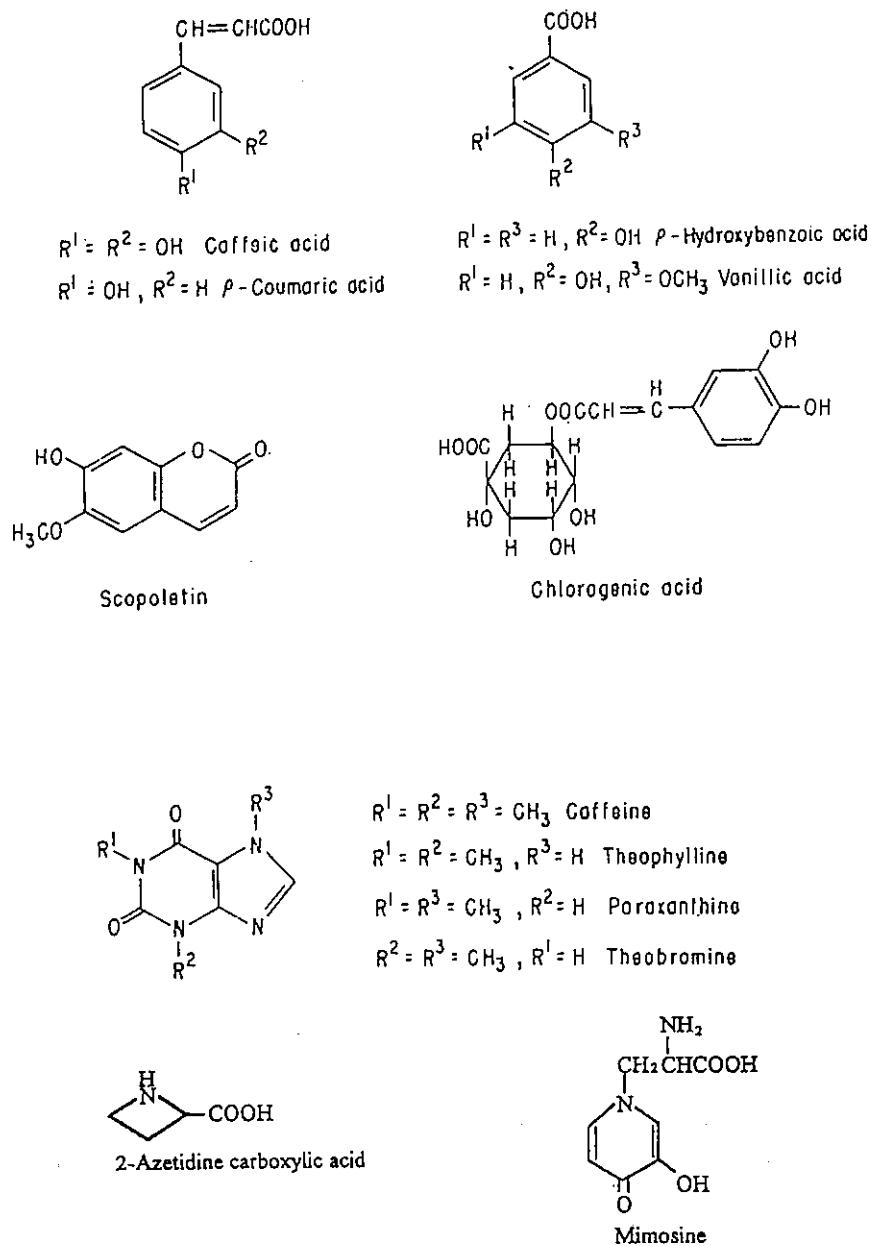


Fig. 3 Chemical structure of potential allelopathic phenolics and alkaloids from plant parts mentioned in the text

test plot adjacent to the control plot was planted with kikuyu grass (*Pennisetum clandestinum*) and the other half left open. The experiment was designed to determine the reciprocal interaction of fir litter and kikuyu grass, and to evaluate the allelopathic influence of the two plants on weed growth. The biomass of kikuyu grass invading the cleaned plot was significantly higher than that invading the control plot (Chou *et al.*, 1987). In addition, the number of weeds that grew in the plot planted with kikuyu grass was smaller than that in the control plot, indicating that the kikuyu grass may compete with and suppress weeds. The seedlings of fir regenerating in the deforested area grew well and did not seem to be affected by the neighbouring newly planted kikuyu grass. The growth of kikuyu grass, however, was inhibited by the fir litter left on the unchanged plot in the first three months after deforestation. Bioassay of aqueous extracts showed that the fir litter extract exhibited a higher phytotoxicity than did the kikuyu grass. Nevertheless, four months after deforestation the growth of kikuyu grass in the field was luxuriant, indicating that the phytotoxicity of fir litter disappeared (Chou *et al.*, 1987).

In another experiment (Chou *et al.*, 1989), a split plot design of eight treatments was set up after deforestation of Chinese fir (*Cunninghamia lanceolata*): open ground without planting (control), planted with kikuyu grass, planted with kikuyu grass and *Alnus formosana*, planted with kikuyu grass and *Zelkova formosana*, planted with kikuyu grass and *Cinnamomum camphora*, planted with *A. formosana*, planted with *Z. formosana*, and planted with *C. camphora*. Field measurements showed that weeds grew luxuriantly six months after treatment in plots which had not been planted with kikuyu grass. The growth of weeds was significantly retarded, whereas that of woody plants was not affected when the plots were planted with kikuyu grass. Compared to the tap-water control, the aqueous leachate of kikuyu grass stimulated the seedling growth of *C. camphora* and *A. formosana*, while the extract stimulated the growth of *C. camphora* and inhibited that of *A. formosana*. The aqueous extracts of three hardwood plants exerted various levels of inhibition on the root initiation of kikuyu grass. The aforementioned extracts and leachates were bioassayed against seed germination and radicle growth of four test plants, including *Miscanthus floridulus*. The extract of *Z. formosana* revealed the highest phytotoxic effect on the test species while that of kikuyu grass showed the least effect. The phytotoxic phenolics (Table 1, Fig. 1) were identified by chromatography. The quantity of phytotoxins present was highest in the extract of *Z. formosana* and was lowest in that of kikuyu grass. The degree of phytotoxicity and quantity of phytotoxins were well correlated, indicating that a selective allelopathic effect was involved (Chou *et al.*, 1989).

Conclusion

Allelopathic chemicals could only temporarily suppress plant growth and regulate species diversity without leading to species extinction because naturally occurring compounds are rapidly degraded into non-toxic compounds and are associated with less residual effect. However, the synthetic agrochemicals often exhibit a greater toxicity and are not easily decomposed, leading to a longer residual effect on living organisms and causing environmental pollution. For sustainable development of agriculture, we have to be aware of the importance of biodiversity and balance of ecosystems, otherwise, the benefit of our economic success and agricultural production through advanced scientific technology would be offset by the feedback of environmental degradation.

Acknowledgement

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Discussion

Mukhopadhyay, S. (India): In India, in the course of our studies on allelopathy, we observed that eucalyptus trees possess two allelochemicals, citronellol and citronellal which suppress weed growth under eucalyptus trees. The leaf extract was able to suppress the germination of rice crop seeds also.

Answer: Such effect on weed suppression has been reported in eucalyptus trees growing in California.