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Effect of Pesticides on Some Common Blue Green Algae

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ABSTRACT

Once known as blue-green algae, cyanobacteria are the most diverse photosynthetic bacteria. The gram negative bacteria have chlorophyll a and photosystems I and II that allow them to perform oxygenic photosynthesis. Unlike most bacteria, cyanobacteria lack α-ketoglutarate dehydrogenase and therefore do not use the citric acid cycle for carbohydrate metabolism, but the pentose phosphate pathway. Cyanobacteria, a group of prokaryotic, oxygen-evolving, photosynthetic Gram-negative bacteria, survive in a wide variety of extreme environmental conditions; they are exposed to various types of natural stresses, such as nutrient limitation, pesticides, pollution, drought, salinity, temperature, pH, light intensity and quality, etc. A Protein in the cyanobacterial thylakoid membranes was identified as a sensitive protein to environmental stress conditions: under various unfavorable conditions like drought, nutrition deficiency, heat, chemical stress, ozone fumigation as well as UV-B and visible light stresses can influence the turnover of protein. Many species are capable of not only surviving, but thriving in conditions previously thought to be inhabitable, tolerating desiccation, high temperatures, extreme pH, high salinity and pesticides illustrating their capacity to acclimate to extreme environments. The major abiotic factor affecting the distribution of algae in soils is solar radiation, moisture, temperature, nutrients, and pH, organic matter content and soil texture are less important. Generally, the higher the soil moisture, soil temperature, and sunlight penetration to the soil surface, the greater the population and activities of algae. The impacts of pesticide microflora in wetland soils depends on their persistence, the concentrations attained in the environment, and synergistic/antagonistic effects among pesticides and between pesticides and fertilizers. In rice fields, pesticides can be sprayed, applied in the floodwater, incorporated into the soil, or used for dipping rice seedling at transplanting.

1. Introduction

Blue-green algae (Cyanobacteria) is convenient for talking about organisms in water that make their own food. Cyanobacteria are relatives to bacteria, not eukaryotes, and it is only the chloroplast in eukaryotic algae to which cyanobacteria are related. Some cyanobacteria are aquatic and photosynthetic, that is, they live in water, and can manufacture their own food (Adhikari, 1989). They are quite small and usually unicellular, though they often grow in colonies large enough to see (Anderson, J.R. 1978). In fact, it may surprise you then to know that the cyanobacteria are still around; they are one of the largest and most important groups of bacteria on earth (Batterton, et.al 1971). The great contribution of cyanobacteria is the origin of plants chloroplast with which plants make food for themselves is actually a cyanobacterium living within the plant's cells. Sometime in the late Proterozoic or in the early Cambrian, cyanobacteria began to take up residence within certain eukaryote cells, making food for the eukaryote host in return for a home. Cyanobacteria have an impressive ability to colonise infertile substrates such as volcanic ash, desert sand and rocks (Butler, 1977). They are extraordinary excavators, boring hollows into limestone and special types of sandstone. Another remarkable feature is their ability to survive extremely high and low temperatures. Cyanobacteria are inhabitants of hot springs, mountain streams, Arctic and Antarctic lakes and snow and ice(Chinnaswamy and Patel. 1983). The cyanobacteria also include species that run through the entire range of water types, from polysaprobic zones to katharobic water. Bergey's Manual has divided the organism into five subsections. The classical taxonomy of cyanobacteria divides these organisms into five 'subsections' or orders, three for non-heterocystous types and two for heterocystous types. The non-heterocystous cyanobacteria comprise Subsection I (Chroococcales), which are unicellular cyanobacteria that reproduce by binary fission; Subsection II (Pleurocapsales) are unicellular cyanobacteria that produce daughter cells smaller than the parent; and Subsection III (Oscillatoriales) consists of cyanobacteria that produce filaments of cells known as trichomes. All three subsections have N2-fixing representatives. Heterocyst formation is an important aspect to nitrogen fixation. The filamentous cells differentiate into heterocysts when the cells are deprived of dissolved inorganic nitrogen (Das, and Singh,1979). A heterocyst consists of a thick cell wall and only contains photosystem I for ATP production. Photosystem II is degraded to prevent O2 production. O2 inhibits nitrogenase, the enzyme responsible for N2-fixation (Da Silva, et.al. 1975).

2. Materials and Methods

Methodological aspects of the study of pesticide impacts on soil and water microflora methods used to study pesticide impacts on rice field microflora include tests on cultures of microorganisms isolated from rice soils, small-scale experiments on soil in test tubes and beakers, and pot and field experiments(Gangawane, L.V. and Kulkarni. 1979). In Vitro Experiments Many studies of the effects of pesticides on soil microflora are laboratory experiments conducted with cultures of microorganism (Hutber, et.al. 1979). Experiments with cultures of microorganisms can give an index of the sensitivity of the strains to pesticides, but it is difficult to draw general conclusions from such data because microorganisms of a same taxon often show different responses to the same pesticide and toxicity in vitro depends on the culture conditions, the nutrient concentration, and the initial size of the inoculum. This is especially well demonstrated with cyanobacteria. Marked differences in response of Nostocaceae (cyanobacteria) The effect of culture conditions was demonstrated with pesticides, which was more toxic to Nostoc muscorum under conditions reflecting or causing a low growth of the cyanobacteria than under conditions favorable for its growth (Kumar, 1988). The initial size of the inoculum had a significant role on the tolerance of Anabaenopsis ruciborskii, Anabaena aphanizomonoides, A. spiroides, and Microcystis jlos-aquae to 2,4-D and BHC. In addition toxicity tests were often performed in a way that hardly permits comparisons and extrapolations. Most in vitro experiments only indicate the concentration of pesticide used in the culture medium and not the recommended level for field application (RLFA). Assuming a floodwater depth of 5 to 10 cm and a homogeneous dissolution of pesticides-which is indeed quite far from the reality-the application of 1 kg of active ingredient (ai) per hectare would correspond to 1 to 2 ppm ai in water. Assuming a puddled layer of 15 cm with a bulk density of 0.5, the application of 1 kg ai/ha would correspond to about 1.33 ppm on soil dry weight basis (Shivaram, and Shetty 1988).

3. Results and Discussion

As in the field most pesticides are usually applied at dosages lower than 3 kg ai/ha, concentrations of 10 to several hundred ppm often tested in flask cultures appears to be used more to estimate a lethal level than to reflect field situation (Singh, and Singh. 1988). They are of little value for drawing conclusions on impacts, except when no significant effect was recorded. Results of in vitro trials can hardly be extrapolated to field conditions for several reasons summarized thereafter:

(i)Toxicity is likely to be higher in flask cultures than in the field. In soil, many factors interact with pesticides and modify their effect as compared with flask culture of a single organism and enhance pesticide degradation. These factors include biological degradation of the pesticide by the soil microflora, nonbiological degradation, photodecomposition, leaching, volatilization, and adsorption to the soil particles.

(ii) Toxicity depends on the initial microbial population and its nutrient status. These conditions are likely to markedly differ in vitro and in situ. In the field, toxicity depends on the method of pesticide application and water management. In vitro experiments are of limited practical value and should be limited to toxicity tests under standardized conditions in order to allow comparisons. A possible standardization could be the determination of the concentrations that would reduce by 50 percent the growth of reference organisms in exponential phase and the concentration that would totally inhibit their growth.

Data on cyanobacteria and microalgae collected in the database were primarily percentages of inhibition estimated by various measurements on cultures (dry weight, fresh weight, total nitrogen, chlorophyll content, etc.). They were tabulated for each pesticide in a especially designed Hyper Card stack, using a grid combining a geometric scale of pesticide concentration and four levels of inhibition: none, ~50 percent, 50 percent, >50 percent, and total inhibition. This tabulation allowed to identify results obtained at concentrations higher than the RLFA (Singh et.al. 1986). Data for other microorganisms were bacterial counts and activity measurements, often performed at several time intervals after pesticide application. Inhibition, no effect, and enhancement were reported. For tabulating such data, each experiment was attributed a score of one within a five-case scale:

l All negative: the treatment was statistically lower than the control for all measurements;

l Negative trend: various effects were recorded, the balance was negative; Cards are connected to the bibliographic database. The upper field provides general information on the pesticide and a summary of the effects recorded by the authors listed in the bibliographic database. The lower field shows the recommended level for application (black dots) and the recorded effects. Numbers in boxes are the number of algal strains presenting a given inhibition at a given concentration of pesticide. “s” refers to a field or pot experiment (soil). “A” refers to symbiotic cyanobacteria (Azolla). The vertical bold line indicates an estimate of the upper limit of field concentration calculated on the basis of the RLFA and 1 kg ai/ha-’ = 2 ppm

l No effect: no statistically significant difference between treatment and control;

l Positive trend: various effects were recorded, the balance was positive; and

l All positive: for all measurements the treatment was statistically higher than the control.

4. Factors Affecting Pesticide Toxicity on blue green algae

- Soil Properties: Little information is available on soil properties that affect pesticide impacts on ricefield microorganisms. The few studies conducted with several soils report some differences in pesticide impacts. In particular the response of N,-fixing organisms to benomyl, carbofuran and gamma-BHC varied with the soil type.
- Water Management and Method of Pesticide Application: A faster pesticide dilution in wetland than in upland soils might be expected, with variations depending upon solubility and the surfactants used.

Bioconcentration of Pesticides in Microalgae and Cyanobacteria: Microalgae and cyanobacteria, the base constituents of the aquatic food web in wetland ricefields, have a high surface area/volume ratio, which give them a significant

potential for sorption of, and reaction with pesticides (Singh, 1973). They can concentrate pesticide many fold and, in general, are more resistant to their toxic effects than the food web's high members (Venkataraman and Rajyalakshmi, 1972). Little field data is available on bioconcentration of pesticide by phototrophic microorganisms in ricefields, but data from laboratory experiments and studies in microcosms and freshwater ecosystems, demonstrate that microalgae and cyanobacteria in ricefields may play an important role in accumulating pesticides that become available to bioconcentration through the food web. For example, cyanobacteria concentrated 100 to 250 times chlorinated pesticides introduced at a level of 1 mg/L in the culture medium. Maximum bioaccumulation ratio of fenitrothion ranged from 44 to 105 in living cells and from 100 to 1,810 in dead cells of Chlore Zla vulgaris, Nitschia closterium, and Anabaena JEos-aquae. Corresponding values for DDT were 420 to 82,000 and 1,000 to 210,000. This aspect is important when considering the ricefield ecosystem as a possible environment for aquaculture (rice-fish, rice-shrimp). As sensitivity to a given pesticide may vary between quite large limits among algal strains, pesticide application might cause shifts in dominant strains within the algal/cyanobacterial community rather than a decrease of the whole algal biomass (Wainright 2008).

Effects on Heterotrophic Biological N_2 Fixation: N,-fixing microflora and BNF were more affected by pesticides (no effect: 31 percent) than other populations and activities of the N cycle (no effect: 71 percent). The low percentage of nonsignificant effects on BNF was mostly due to a higher number of positive effects (45 percent), observed indiscriminately with fungicides, herbicides, and insecticides. Most often, a positive effect was observed, but a single pesticide could exhibit negative or positive effect depending on the soil type.

Effects on Nitrification- Denitrification: Nitrification was not affected by pesticides in about 60 percent of the cases. This value is similar to the average of the database. However, negative effects were much more frequent (34 percent of the cases) than positive effects (6 percent of the cases). Nitrification inhibition cannot be considered detrimental because it reduces losses from nitrogen fertilizer. In fact the identification of efficient and conomically feasible nitrification inhibitors has been an important objective of the research on the microbial management of ricefields. Denitrification was not affected by pesticides in 87 percent of the cases. This is probably because the denitrifying microflora, being complex and very versatile, is able to metabolize or to resist a wide range of substrates. As a result, high pesticide levels are needed to inhibit denitrification. Wainright, 1978 tested the effect of eight dithiocarbamate pesticides in a rice soil, found that 20 ppm Vapam (metham) or 100 ppm of the other pesticides was required to significantly decrease denitrification at two and five days after pesticide application. Such concentrations are higher than the RLFA.

5. Conclusion

A positive effect of insecticide gamma-BHC on N2 fixation and populations of anaerobic, phosphate-dissolving bacteria was reported by Raghu and MacRae (1967a, 1967b). A decrease in microbial population was reported after the application of insecticides diazinon, cytrolane, carbofuran, carbaryl + lindane, quinalphos, and Dursban at RLFA whereas fungal populations were not affected (Purushothaman, Venkataraman, and Kasirajan, 1976). The different methods can induce significant differences in pesticide behavior. However, because of the presence of floodwater and puddled soil, a faster dilution can be expected as compared with uplands, where pesticide remains at the soil surface until cultivation or watering incorporates them into the soil. Pesticide degradation in tropical rice fields is favored by

(1) temperatures and pH, which usually stabilize in a range favoring high microbial activity, and (2) reducing conditions caused by submersion and further accelerated by organic matter incorporation.

Therefore, pesticide degradation is often faster in flooded than in non flooded soils and other aerobic systems. As a result of a shorter persistence and faster dilution, pesticides should have less impact on soil microflora in wetland rice fields than in upland soils. Both positive and negative effects of pesticides have been observed. Significant effects of pesticides were less often recorded in situ than in vitro and they were more often negative than positive, whereas the same percentage of positive and negative effects (20 percent) was recorded with the whole dataset.

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