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THE RHIZOSPHERE MICROBIOLOGY  
OF ROOTED AQUATIC PLANTS

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by

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<p>The rhizosphere includes the root systems of higher plants and the assemblages of microorganisms associated with them. Plant-microbe interactions in the rhizosphere have not been extensively examined in freshwater plants. However, information based on studies conducted in other environments indicates that these interactions may be important in freshwater systems. An understanding of the microbial processes occurring in the rhizosphere is needed to better elucidate nutritional relationships affecting aquatic plant productivity and distribution.</p> <p>This report, based on a review of the literature, examines several aspects of the rhizosphere of rooted aquatic plants. These include: the nature and properties of the rhizosphere; the manner in which plants influence the rhizosphere microflora; the microbial transformations of nutrients in the rhizosphere; and the overall role of the</p> <p style="text-align: right;">(Continued)</p>					
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Decomposition	Nitrogen fixation	Plant growth regulator
Denitrification	Organic acid	Rhizoplane
Fungi	Phenol	Rhizosphere microflora

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rhizosphere microflora in affecting plant growth regulation through influences on nutrient availability, sediment oxidation-reduction status, and gas formation.

Sediment is a complex chemical and microbiological environment, even in the absence of plant roots. The presence of anaerobic conditions, accompanied by a low oxidation-reduction potential and often by toxic constituents, places stresses on plants using anaerobic sediments as a rooting medium. Plant roots have developed strategies for coping with life in anaerobic sediments; certain of these strategies directly influence the surrounding sediments and the rhizosphere microflora.

Some authors have suggested that the rhizosphere microflora-plant root interrelationship may be obligatory for nutrition of some aquatic plants. Certain rhizosphere microorganisms are dependent upon the host plant as a source of necessary organic substances. The plant, in turn, may derive one or more benefits; examples include nitrogen fixation, solubilization of phosphate, oxidation or degradation of toxic substances, and possible elaboration of plant growth factors. Colonization of host plant roots by beneficial microorganisms may also serve a protective function by preventing colonization by pathogenic microorganisms.

Very little is known about the rhizosphere microbiology of freshwater plants. However, based on the literature examined, there are several areas of major importance that appear to warrant investigation. These areas include: the role of rhizosphere microflora in aquatic plant nutrition; production of plant growth-regulating substances by rhizosphere microflora; the differences in rhizosphere microflora between different rooted aquatic plants occurring in the same sediment and between the same rooted plant in different sediments; the role of the rhizosphere microflora in aquatic plant senescence; the role of the plant growth-promoting rhizosphere bacteria in plant growth, development, and productivity, and in inhibition of plant pathogens; and the role of the rhizosphere microflora in the formation and stabilization or destabilization of organic complexes.



## Preface

The study reported herein was sponsored by the Department of the Army, Office of the Chief of Engineers (OCE), Directorate of Civil Works (DAEN-CW), through the US Army Corps of Engineers Aquatic Plant Control Research Program (APCRP). Funds were provided by DAEN-CW under Department of the Army Appropriation No. 96X3122 Construction General. The APCRP is managed by the US Army Engineer Waterways Experiment Station (WES), Vicksburg, Miss. Technical Monitor for OCE was Mr. E. Carl Brown. Principal investigators for this study were Drs. Douglas Gunnison and John W. Barko, Environmental Laboratory (EL), WES. The report was prepared by Dr. Gunnison. Reviews of this report were provided by Ms. Judith C. Pennington, Mr. William D. Taylor, and Dr. Thomas L. Hart of the EL. Technical assistance was provided by Ms. Valerie Warner of the EL. The report was edited by Ms. Jessica S. Ruff of the WES Information Technology Laboratory.

This investigation was performed under the general supervision of Dr. John Harrison, Chief, EL, and Mr. Donald L. Robey, Chief, Ecosystem Research and Simulation Division, and under the direct supervision of Dr. Hart, Chief, Aquatic Processes and Effects Group. Manager of the APCRP was Mr. J. Lewis Decell.

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Introduction

1. Rooted aquatic plants play an important role in aquatic systems by producing food and providing habitat diversity (Pennak 1971, Wiley et al. 1984). Despite these positive considerations, many aquatic plants have caused concerns in areas where they occur at nuisance levels. The current understanding of the effects of environment on the growth and distribution of rooted aquatic plants is limited, but improving (cf. Barko, Adams, and Clesceri 1986). A large amount of information on the physiology and ecology of rooted aquatic plants has been developed in recent years. However, only abiotic factors, including light, temperature, nutrients, and sediment composition, have been effectively considered. The influence of biotic factors, including plant-microbial interactions, on rooted aquatic plants is poorly understood, and thus requires increased investigative attention. This lack of understanding extends to the plant root-microorganism association, known as the rhizosphere. The rhizosphere consists of the root system of higher plants and its associated assemblages of microorganisms.

2. Information obtained to date on the rhizosphere of agronomically important terrestrial plants indicates that rhizosphere microorganisms are important in nutrition, and therefore affect plant growth and survival. Sediment, however, differs from soils in fundamental ways, providing a unique chemical and microbiological environment. Sediments are typically anaerobic, thus requiring specific adaptations by rooted plants to cope with physiological stresses. Certain of these adaptations also influence the rhizosphere microflora. Evidence obtained to date indicates that the rhizosphere microflora can play a critical role in the nutrition of rooted aquatic plants (Durako and Moffler 1987). However, the extent to which microbial mineralization processes refurbish nutrient supplies in sediments, depleted due to uptake by rooted aquatic plants, needs to be determined (Barko et al., in preparation).

3. Better information on the interactions among aquatic plants, sediments, and rhizosphere microflora will be of value in improving our understanding of the nutritional ecology of rooted aquatic plants, ultimately leading to the development of innovative, ecologically oriented plant

management techniques. This report reviews the current state of understanding of rhizosphere microbiology in freshwater habitats. Although the primary focus of the report is on freshwater plants, a great deal of reliance is placed on review of literature concerning rhizosphere processes in terrestrial and marine plants.

### Nature and Properties of the Rhizosphere

#### Sediment environment

4. A great deal of research has been conducted on the rhizosphere of terrestrial plants, while comparatively little has been done on the rhizosphere of aquatic plants. An important part of our understanding of what occurs in plant rhizospheres in aquatic sediments is inferred from what has been learned about terrestrial plant rhizospheres. It is important to understand the differences between the soil and sediment environments; these differences have a bearing on the environmental adaptations required by the plants and their respective microfloras. A brief summary of the differences between these environments is presented in Table 1. A general comparison of the characteristics of each environment reveals that these three substrates are quite different from one another. Differences between terrestrial soils and aquatic sediments are the most distinctive, while flooded soils are variable and transitional between the other two. Flooded soils and sediments are characterized by high moisture levels and the presence of varying degrees of reducing conditions.

5. Plants exert an important influence on the physical and chemical nature of bottom sediments. For example, several studies have demonstrated that seagrasses can modify the textural properties of sediments. Once established, these plants decrease mean sediment particle size (Burrell and Schubel 1977; Kenworthy, Zieman, and Thayer 1982). Kenworthy, Zieman, and Thayer (1982) presented evidence that sediments underlying seagrasses have substantially higher levels of fine-textured particles, organic matter, and both total and intermediate pools of nitrogen. A number of factors account for the effects of seagrasses on sediment properties. The leaves of seagrasses decrease flow velocity through obstruction of currents (Gregg and Rose 1982) and, along with a dense array of roots and rhizomes, establish depositional conditions within the seagrass meadow (Ginsburg and Lowenstam 1958; Kenworthy,

Table 1  
Comparison of Terrestrial Soils, Flooded Soils, and Aquatic Sediments\*

Constituent	Terrestrial Soils	Flooded Soils	Aquatic Sediments
Color	Variable	Variable	Dark, except for sands and gravels
Moisture	Intermittently wet and dry	Intermittently saturated	Permanently saturated
Dissolved oxygen	Oxygen present in gaseous form, in dissolved form in soil water; anoxic conditions occur at depth	Soil system becomes devoid of dissolved oxygen within a few days after flooding	Sediment anoxic, except for a few millimeters or centimeters at the surface
Oxidation state	Oxidized	Intermittently oxidized and reduced, reduced with degree of reduction increas- ing with increased duration of flooding	Permanently reduced, except for oxidized surface layer
Organic components	Labile organics are rapidly con- sumed, and humic materials are accumulated	In dry periods, labile organics accumulate; in flooded periods, a temporary buildup of reduced organic com- pounds occurs	Significant accumu- lation of reduced organic compounds and partially degraded plant and animal matter
Inorganic compounds	Inorganic com- pounds primarily in oxidized state; mobile forms move downward with movement of ground water	Inorganic compounds primarily in oxi- dized state during dry periods; in wet periods, a temporary accumulation of reduced forms occurs**	Inorganic compounds are primarily in reduced form,** except for oxidized surface layer, where oxidation of diffusing species occurs

\* Based on Gunnison, Engler, and Patrick (1985).

\*\* Reduced species include ammonium-nitrogen, ferrous iron, manganous manganese, sulfide, and methane.

Zieman, and Thayer 1982). The plants themselves contribute large amounts of organic matter through root and rhizome biomass production and leaf deposition. Organic matter and fine-textured sediments provide a large surface area for adsorption-desorption phenomena, including sorption of organic molecules and inorganic ions, as well as attachment of microorganisms (see Muller 1977; Rosenfeld 1979; Kenworthy, Zieman, and Thayer 1982).

6. Nutrient uptake from sediments by freshwater macrophytes has been demonstrated to dramatically decrease concentrations of nitrogen and phosphorus in the root zone (Prentki 1979; Carignan 1985; Barko et al., in preparation). Mechanisms, including microbial, whereby these nutrients are replenished are essentially unknown. The balance between nutrient uptake by rooted plants and nutrient replenishment may underlie major differences in the productivity and successional development of aquatic macrophyte communities (Barko et al., in preparation).

#### Adaptations of plant roots to life in anaerobic sediments

7. In addition to difficulties associated with rooting in unstable aquatic substrates, rooted aquatic plants have also had to adapt to physiological problems related to conditions of poor aeration and/or anoxia present in most aquatic sediments (Sculthorpe 1967). Anoxic conditions in sediments favor formation and accumulation of reduced forms of manganese, iron, sulfur, and carbon (Ponnamperuma 1972). Some of these substances can be toxic to plants, if present in high levels (Sanderson and Armstrong 1980). The roots and rhizomes of aquatic plants occurring in these substrates must either be able to tolerate such conditions, while simultaneously absorbing nutrients and synthesizing cellular constituents (Sculthorpe 1967, Drew and Lynch 1980), or else have some strategy for supplying oxygen to root cells.

8. A description of some of the mechanisms used by aquatic plants to meet life requirements under these strenuous conditions is presented in Table 2, where an attempt has been made to separate adaptive mechanisms into several distinct groups. It should be noted, however, that several plants listed in Table 2 fall into more than one group. Some plants are capable of supplying their roots with sufficient oxygen to provide root cells with oxygen levels in excess of their needs. As a result, the sediments immediately surrounding these roots are oxidized. In other cases, oxygen may be present within the roots in excess of respiratory requirements; however, a

Table 2

Adaptations of Aquatic and Wetland Plants to Life in Anaerobic Sediments

Adaptation Strategy	Examples*	References**
Plants transporting oxygen to roots, often in excess of plant root requirements	<i>Egeria densa</i>	Sorrell and Dromgoole 1987
	<i>Eriophorum angustifolium</i>	Armstrong 1967
	<i>Isoetes lacustris</i>	Tessenow and Baynes 1978; Sand-Jensen, Prah1, and Stockholm 1982
	<i>Juncus effusus</i> +	Armstrong 1967
	<i>Lobelia dortmanna</i>	Sand-Jensen and Prah1 1982, Sand-Jensen, Prah1, and Stockholm 1982
	<i>Littorella uniflora</i>	Sand-Jensen, Prah1, and Stockholm 1982
	<i>Menyanthes trifolata</i> +	Armstrong 1967
	<i>Molina coerulea</i> +	Armstrong 1967; Smith, Dennison, and Alberte 1984
	<i>Myriophyllum verticillatum</i>	Carpenter, Elser, and Olsen 1983
	<i>Narthecium ossifragum</i>	Armstrong 1967
	<i>Nuphar luteum</i>	Dacey 1980
	<i>Oryza sativa</i> +	Armstrong 1967
	<i>Potamogeton crispus</i>	Sand-Jensen, Prah1, and Stockholm 1982
	<i>P. friesii</i>	Sand-Jensen, Prah1, and Stockholm 1982
	<i>Schoenus nigricans</i> +	Armstrong 1967
	<i>Sparanium simplex</i>	Sand-Jensen, Prah1, and Stockholm 1982
	<i>Spartina alterniflora</i> +	Teal and Kanwisher 1966
	<i>Spartina townsendii</i> +	Armstrong 1967
	<i>Zostera marina</i>	Iizumi, Hattori, and McRoy 1980
		Laing 1940
Plants having structural and physiological mechanisms to acquire maximum available O <sub>2</sub> or able to respire anaerobically and tolerate organic by-products	<i>Acorus</i> , <i>Asclepias</i> , <i>Nuphar</i> , <i>Nymphaea</i> , <i>Peltandra</i> , <i>Pontederia</i> , <i>Saggitaria</i> , <i>Scirpus</i> , <i>Sparangium</i> , <i>Typha</i> , <i>Zostera marina</i>	

(Continued)

\* Plants denoted with "+" are emergent species having shoots in the air. Remaining plants are submersed species.

\*\* Based on information and references given in Sculthorpe (1967). References followed by "+" were cited by Sculthorpe. Others are included in the References section of this paper.



Table 2 (Concluded)

Adaptation Strategy	Examples	References
Plants having physiological mechanisms to tolerate periods of anaerobiosis	<i>Equisetum fluviatile</i> <i>Iris pseudacorus</i>  <i>Zostera marina</i> 1962	Barber 1961++ Boulter et al. 1963++ Henshaw et al. 1961++, 1962++ Penhale and Wetzel 1983
Plants having compensation mechanisms to decrease rate of $O_2$ consumption with $CO_2$ accumulation in tissues	<i>Menyanthes trifoliata</i>	Coult 1964

nutrient-rich sediment harboring high levels of metabolically active microorganisms may remain anoxic in the presence of these roots. The rate of oxygen release from the roots under such circumstances is greatly exceeded by the rate of oxygen consumption in the surrounding sediment.

#### Characteristics of rhizosphere microorganisms

9. Although limited in the past to plants used for agricultural purposes, information on the rhizosphere microbiology of terrestrial plants has increased rapidly over the past few years and has expanded to encompass many uncultivated species. Most recently, the rhizosphere of plants in extreme terrestrial environments has been examined, including areas as diverse as arctic and tropical desert regions (see Lifshitz et al. 1986, Reinhold et al. 1986). The emphasis of research on terrestrial rhizosphere microorganisms has also changed over the years. The initial interest was in the identity of microorganisms associated with nitrogen fixation in crop plants. Recently, the research emphasis has shifted to intensive biochemical and genetic investigations of the plants and rhizosphere microorganisms themselves and the formation of attachment structures (see, for example, studies by Gardes, Bousquet, and Lalonde 1987; Sargent et al. 1987; Vesper 1987).

10. Microorganisms living in the rhizosphere communities of terrestrial and aquatic plants occur in the soil or sediment surrounding the plant and on the surfaces of roots and root hairs. The rhizosphere consists of (a) the

inner rhizosphere, i.e., the immediate root surface, and (b) the outer rhizosphere, i.e., the immediately adjacent soil. The term rhizoplane refers to the root surface and its adhering soil. For terrestrial plants, the most active zones of microbial activity are generally those areas slightly behind the tip of actively growing roots and on senescent roots and/or sloughed off root hairs (Russell 1968).

11. In the terrestrial rhizosphere, several factors favor the growth and development of certain kinds of microorganisms. Among these factors are various combinations of substances released by roots coupled with physical changes in the soil caused by root activity. The exact processes by which microbial rhizosphere populations develop is not understood; however, it is known that the predominant microorganisms are those which, because of their physiological and metabolic traits, can most rapidly exploit the unique conditions in the rhizosphere. Thus, through specific biochemical compounds (carbohydrates, proteins, vitamins, etc.) excreted by the roots and products released by decomposition of root structural components, individual plants provide an environment favoring the growth of selected members of the native soil/sediment microflora. The microorganisms selected are those able to most effectively utilize this suite of compounds. In fact, many of these microorganisms may grow best only in the environmental circumstances provided by the rhizosphere. Microorganisms growing in the rhizosphere, in turn, produce metabolic products that may exert positive, neutral, or negative influences upon the plant (see Macura 1968; Alexander 1971, 1977; Lynch 1979).

12. Studies on the microbial composition of the rhizosphere of rooted aquatic plants are limited for aquatic environments in general and almost nonexistent for freshwater habitats. Of the microorganisms present in aquatic ecosystems, the bacteria inhabiting these communities have been examined most extensively (Table 3). Many studies have considered the occurrence of individual types of microorganisms in association with specific processes. The most complete freshwater study to date examined microbial populations associated with the rhizosphere of *Myriophyllum heterophyllum* and assessed the role of the rhizosphere microbial activity in essential nutrient transformations (Blotnick, Rho, and Gunner 1980). Rice, because of its importance as a crop plant, has been examined by several authors with respect to nitrogen fixation (Watanabe et al. 1977, 1979; Watanabe and Barraquio 1979; Barraquio, Ladha, and Watanake 1983).

Table 3  
Microorganisms Known to be Present in Aquatic Plant Rhizospheres

Plant	Processes and Responsible Microorganism	Reference(s)
<i>Myriophyllum</i>	No specific microorganisms. Did classification by activities carried out by microbes: ammonifiers, denitrifiers, acid production from glucose	Blotnick, Rho, and Gunner 1980
Rice	<i>Beggiatoa</i> sp. (organism oxidizes $H_2S$ to $H_2SO_4$ using $O_2$ released from plant roots) <sup>2</sup>	Joshi and Hollis 1977
Seagrass	<i>Beggiatoa</i> sp. (probably functions same as for rice)	M. J. Klug (personal communication with Penhale and Wetzel 1983)
<i>Halodule wrightii</i>	Nitrogen-fixing bacteria, including <i>Klebsiella pneumoniae</i>	Smith and Hayasaka 1982a
	Nitrogen fixation by a <i>Klebsiella</i> sp.	Schmidt and Hayasaka 1985
<i>Zostera</i>	Unspecified, but approximately one third of rhizosphere isolates were obligate aerobes; remainder were facultative anaerobes. Observed ammonification of amino acids, including some found on root surface	Smith, Hayasaka, and Thayer 1984
	Unspecified, but all were obligate aerobes. Bacteria solubilized calcium phosphate when grown on glucose, but not on amino acids. Acetic acid postulated as agent	Craven and Hayasaka 1982
<i>Thalassia testudinum</i>	Nitrogen fixation in rhizosphere and phyllosphere	Capone 1983

13. Fungi living in the rhizosphere are also prominent. Some of these organisms, the mycorrhizae, form mutually beneficial relationships with the host plant. In these mutualistic relationships, fungi make important contributions to the nutrition of the host plant, while obtaining selected nutrients from the plant (Alexander 1977, Pennington 1986). Pennington (1986) recently reviewed the literature on the use of mycorrhizal fungi for enhancement of marsh plant establishment on dredged sediments. Knowing which fungi are important for enhancement of plant growth in a marsh is difficult because most investigations have been conducted with fungi taken from terrestrial species. In general, the literature available concerning the mycorrhization of aquatic plants is extremely limited. Some aquatic plants lack mycorrhizae (see Khan 1974, Read et al. 1976), while others demonstrate slight to extensive mycorrhizal associations (Sondergaard and Laegaard 1977; Bagyaraj, Manjunath, and Patil 1979; Chaubal, Charma, and Mishra 1982; Clayton and Bagyaraj 1984). A listing of aquatic plants having mycorrhizal fungi (modified from information given by Pennington 1986) is presented below.

*Callitriche hamulata*

*Cyanotis cristata*

*Elatine gratioloides*

*Eichornia crassipes*

*Eleocharis palustris*

*Eleocharis pusilla*

*Elodea canadensis*

*Glossostigma elatinoides*

*Glossostigma submersum*

*Hydrilla verticillata*

*Isoetes kirkii*

*Isoetes lacustris*

*Lagarosiphon major*

*Lilaeopsis lacustris*

*Limnosella lineata*

*Littorella uniflora*

*Lobellia dortmanna*

*Myriophyllum pedunculatum*

*Myriophyllum propinquum*

*Myriophyllum triphyllum*

*Nymphaea alba*

*Paspalum dilatatum*

*Phragmites australis*

*Polygonum hydropiper*

*Potamogeton cheesemani*

*Potamogeton crispus*

*Potamogeton pectinatus*

*Pratia perpusilla*

*Rotala rotundifolia*

*Ranunculus* sp.

*Ranunculus limosella*

*Ranunculus rivularis*

*Ruppia polycarpa*

*Salvinia cucullata*

*Tillaea sinclarii*

*Zannichellia palustris*

14. From the literature examined by Pennington, it is apparent that sediment nutrient levels are an important factor regulating the occurrence of mycorrhizae on aquatic plants. In general, sediments containing high levels of nutrients, such as nitrogen and phosphorus, retard the occurrence and growth of mycorrhizal fungi. In the case of nitrogen, this has been attributed to a shift in the plant's biosynthetic processes occurring upon increased availability of nitrogen. This shift yields higher levels of protein relative to carbohydrate. Proteins are less available to fungi than carbohydrates, and the capacity of the host plant to support fungi thereby decreases (see Pennington 1986 for details).

15. The beneficial effects of mycorrhizal fungi for the host plant, considered in detail by Pennington (1986), are as follows. Mycorrhizal fungi contribute to the mineral nutrition of the host plant in two ways. First, the root-attached fungal hyphae add to the surface area of plant roots, thereby increasing nutrient absorption in nutrient-deficient soils. Second, the fungi can also increase the availability of soil-bound nutrients. Among the nutrients affected in this manner are phosphorus, potassium, and zinc. Through the development of an extensive mycelium, mycorrhizal fungi also contribute to the stabilization of both the plant and the substrate. The fungi further facilitate this stabilization by production of an amorphous polysaccharide that serves to bind the substrate particles and contribute to their aggregation.

#### Plant Influences on the Rhizosphere Microflora

16. Major plant-associated factors influencing the microbial composition of the rhizosphere community are the amount and nature of organic materials released from root tissue and root hairs and the age and species of the plant. Other important factors, not considered here, are the proximity of the plant roots and rhizosphere microorganisms and the depth of the plant roots.

17. Decomposition products resulting from sloughed-off root tissue and root hairs along with various metabolites are known to contribute to the nutrition of microorganisms in the rhizosphere of terrestrial plants (Vagnerova and Vancura 1962). However, substances excreted by the roots themselves seem to be most important (Macura 1968). Factors affecting the chemical composition of root exudates and their influence on bacteria have been examined by several investigators (Lyon and Wilson 1921; Rovira 1956a,

1956b, 1969; Balasubramanian and Rangaswami 1969; Hale, Foy, and Shay 1971). However, the mycorrhizae have only recently begun to receive attention in this regard (Elias and Safir 1987).

18. The species of plant influences the rhizosphere microflora of terrestrial plants. Areas having the same kind of soil but different types of plants have different microflora, probably as a result of different plants having different excretory products, root tissue composition, and types of rooting habits. By contrast, the same plant species occurring in different soils tends to harbor the same or very similar microorganisms in its rhizosphere. (For a detailed discussion of these influences, see Alexander 1977.)

19. Aging, which results in death and sloughing of root tissue, also influences the structure and composition of the rhizosphere microbial community. Since the excretion of organic materials by plant roots mediates the selection and growth of bacteria in the rhizosphere, any changes that aging brings about in the release of plant materials will also affect the rhizosphere community. Thus, plant root excretory products and the composition of root tissues and tissue products are probably major factors determining the type of microorganisms living in the rhizosphere (Alexander 1977, Reinhold et al. 1986). Products excreted by plants grown under aseptic conditions include amino acids, carbohydrates, organic acids, nucleic acid derivatives, enzymes, various growth factors, and several other materials (Alexander 1977).

#### Microbial Transformations of Nutrients in the Rhizosphere

20. Most of the work in the area of rhizosphere microbiology has focused on nitrogen fixation in terrestrial plants, but a limited amount of work has been conducted with aquatic plant species. Through their role in nutrient regeneration, microorganisms release inorganic forms of carbon, nitrogen, phosphorus, and sulfur. Depending on the oxidation-reduction potential, the inorganic forms of most of these elements become available as nutrients to aquatic plants; however, in the case of sulfur, the inorganic forms can be toxic. Microorganisms also take up considerable quantities of nutrients for their own use and, in this manner, can compete with plants for these substances.

## Nitrogen

21. Watanabe et al. (1979) used acetylene reduction in situ to examine the aerobic nitrogen-fixing bacteria associated with rice in the wetland environment. They reported that nitrogen fixation activities increase with the age of the plant, with maximum rates occurring at the seed production or heading stage. Rice is host for a number of different nitrogen-fixing bacteria, including *Enterobacter* (Watanabe et al. 1977), *Azospirillum* (Watanabe et al. 1979), and *Pseudomonas* (Watanabe and Barraquio 1979; Barraquio and Watanabe 1981; Barraquio, Ladha, and Watanabe 1983). Tjepkema and Evans (1976) have found nitrogen fixation in association with *Juncus balticus* and other wetland plants. McClung et al. (1983) have examined the nitrogen-fixing bacteria associated with *Spartina alterniflora* roots. They report that the responsible organism, a *Spirillum*, not only occurs on root surfaces, but also colonizes the interior of the roots of this plant.

22. Several investigators have studied nitrogen fixation in *Halodule wrightii* in the marine environment. Smith and Hayasaka (1982a) examined nitrogenase activity associated with the roots of this plant. They found that the activity of the enzyme varies seasonally, with the highest rates occurring in the warm summer months. At in situ temperatures, nitrogenase activities associated with surface-sterilized and nonsurface-sterilized roots are similar, indicating that there are nitrogen-fixing microorganisms in the roots themselves. An additional finding is that at 35° C, much of the nitrogenase activity is associated with non-surface sterilized roots, the opposite of what is found for cooler temperatures. This indicates that at cooler in situ temperatures, nitrogenase activity may be associated with bacteria situated within the roots, while at warmer temperatures most activity can be attributed to the rhizoplane (root surface) bacteria. Acetylene reduction assays (a measure of nitrogen fixation) conducted with *Halodule* yield higher rates under aerobic than under anaerobic conditions. This suggests that the oxygen-sensitive nitrogenase may be compartmentalized within the root cells, protecting the nitrogenase from inactivation.

23. In a later study, Schmidt and Hayasaka (1985) described a nitrogen-fixing *Klebsiella* in the roots and rhizomes of *Halodule wrightii*. The bacterium colonizes the interior roots of the plant. Nitrogen-fixing activity also occurs with *Zostera marina* (Smith and Hayasaka 1982b); however, the responsible bacteria have not been identified. The sources of elemental

nitrogen used in fixation have not been identified. Some nitrogen undoubtedly diffuses from the overlying water column into the sediments. Some nitrogen may be transported with oxygen into the roots in those plants that carry out this process. A very likely source of elemental nitrogen, particularly for those plants having an oxidized rhizosphere, is the denitrification process.

24. In a study of ammonification of amino acids by rhizoplane microorganisms, Smith, Dennison, and Alberte (1984) found that the microflora of *Halodule wrightii* and also *Zostera marina* deaminate selected amino acids, including some of those found in plant root exudates and tissues. The ammonium released by this process is subsequently absorbed by the root-rhizomes. Boon, Moriarty, and Saffigna (1986) examined rates of ammonium turnover and the role of amino-acid deamination in seagrass (*Zostera capricorni*) beds of Moreton Bay, Australia. Using a  $^{15}\text{N}$  isotope dilution technique, they found ammonium pool turnover rates of 0.4 to 0.8 day in seagrass bed sediments. Between 35 and 65 percent of added  $^{15}\text{N}$ -glycine is deaminated in 12 hr, and the remainder is most likely utilized by microorganisms. These studies provide support for the contention that deamination of amino acids is of major importance in ammonium regeneration in seagrass-bed sediments.

25. Kenworthy et al. (1987) used epifluorescence and scanning electron microscopy to examine bacteria growing on and in close association with the rhizome detritus of the seagrasses *Zostera marina* and *Thalassia testudinum*. Degradation of below-ground plant materials is a function of the metabolism of microorganisms associated with the detritus. The authors found microbial cells of large size along with the presence of measurable acetylene reduction activity, suggesting an active nitrogen-fixing microbial population associated with the rhizome detritus.

26. Durako and Moffler (1987) examined responses of *Thalassia testudinum* to nitrogen enrichment using axenic (algae and bacteria-free) seedlings. They found that axenic seedlings cultured in media amended with various forms of nitrogen demonstrated no growth enhancement compared to controls. In addition, decreases in green leaf areas and widths after 1 month, together with relatively high carbon:nitrogen ratios after 3 months, suggested consumption and depletion of stored nitrogen reserves and little or no assimilation of external nutrients. Based on this information and the results of previous studies with nonaxenic seedling cultures, they concluded that microbial associations may indicate an obligate microbial role in the nutrient



physiology of the plant. Finally, they noted that some microbial-plant associations have been deemed to be critical in plant survival and fitness in certain habitats.

27. Capone (1983) evaluated the importance of nitrogen fixation in the economy of seagrasses for communities of *Thalassia testudinum* and *Zostera marina*. He concluded that nitrogen fixation is probably more important for tropical than for temperate seagrasses, accounting for 20 to 50 percent of the estimated plant requirements in tropical beds. In contrast, nitrogen fixation meets the needs of only 3 to 28 percent of the plant requirements for a temperate *Zostera marina* community.

28. The rhizosphere of some rooted freshwater plants is also known to support nitrogen-fixing bacteria (Bristow 1974). Blotnick, Rho, and Gunner (1980) studied several microbial processes occurring within the rhizosphere of *Myriophyllum heterophyllum*. They selectively sampled microorganisms associated with processes of ammonification, denitrification, and acid production from glucose in the rhizosphere. The composition of the rhizosphere community differed distinctly from that of the adjacent sediment; however, the population density of heterotrophic bacteria in each environment did not differ. Higher populations of bacteria capable of nitrate reduction and ammonification occurred in the rhizoplane, and nitrogen-fixing activity was significantly higher on the root surface than in the sediment. The higher numbers of organisms involved in nutrient transformations in the rhizoplane suggest that the organisms involved have a greater capacity for providing readily available nutrients to these aquatic plants than do nonrhizoplane populations.

#### Phosphorus

29. Microorganisms are known to be important in the mobilization of phosphorus from terrestrial soils (Alexander 1977), where there are two possible mechanisms for phosphate mobilization. These are production of phosphate-dissolving organic acids and elaboration of phosphoesterase enzymes, which release phosphate through hydrolysis. Craven and Hayasaka (1982) examined inorganic phosphorus solubilization by rhizosphere bacteria in a *Zostera marina* community. They found obligately aerobic bacteria capable of calcium phosphate solubilization and determined that the solubilizing agent is probably acetic acid. Production of acetic acid is accomplished microbiologically by oxidation of alcohols; however, the alcohols are produced under anaerobic conditions. This suggests an environment requiring an

aerobic-anaerobic interface. In aquatic sediments, a third mechanism of phosphate release is known. Nitrate-reducing bacteria catalyze chemical iron reduction, which frees phosphorus from iron oxyhydroxide complexes (Jansson 1987).

#### Sulfur

30. There are very few studies on sulfur cycling in the rhizosphere, despite the plethora of information on sulfur biogeochemistry in both freshwater and marine environments (see, for example, Abram and Nedwell 1978; Engler and Patrick 1973; Gunnison, Engler, and Patrick 1985). Joshi and Hollis (1977) found a *Beggiatoa* capable of oxidizing hydrogen sulfide in rice rhizospheres. Penhale and Wetzel (1983) postulated that a *Beggiatoa* was responsible for sulfide oxidation observed in the roots of the sediments surrounding seagrasses. In view of the strong toxicity of sulfide for rooted plants and the oxygen-conducting capacity of many aquatic plant roots, this finding is not unexpected.

### Role of Rhizosphere Microflora in Rhizosphere Processes

#### General functions

31. The microbial rhizosphere community may have either stimulatory or inhibitory effects on the growth of rooted plants. These influences include: (a) elaboration of substances that exert allelochemical effects on plants, (b) release of substances that directly stimulate plant growth, and (c) conduct of processes that alter the plant root environment in a manner that enhances plant growth. The latter category includes processes that influence plant growth through production of stable soil structure, destabilization of organic complexes, and formation of symbiotic relationships with plant roots. In addition to releasing plant growth stimulators, microorganisms may produce compounds that result in solubilization of materials required by the plant. Calcium phosphate is an example of this, as discussed previously. In addition, assimilation of manganese, iron, zinc, and potassium by rooted plants may be stimulated by microbial activity. The activities of the mycorrhizal fungi typify many of these influences, as discussed earlier.

32. Some compounds elaborated by rhizosphere microorganisms have a protective effect, rather than exerting a growth influence. Many rhizosphere microorganisms are capable of producing antibiotic compounds; thus, the

rhizosphere is a likely site for antibiotic production (Alexander 1971, 1977). Antibiotic compounds have not yet been specifically detected in the rhizosphere; however, they may undergo translocation to aerial portions of the plant (Alexander 1977). While antibiotics are not known to be growth enhancers or inhibitors, they do provide a protective mechanism for the plant. By preventing disease or parasitism, antibiotics prevent diversion of the plant's energies into nonproductive channels, thus enabling the plant to better cope with its environment.

#### Formation of plant growth regulators

33. Microorganisms are known to produce substantial quantities of plant growth regulators. These unique substances have been isolated from media in which soil isolates have been cultured. This phenomenon has apparently been examined only for terrestrial plants; however, its importance to the growth of these plants suggests that it may occur elsewhere. Plant regulator compounds produced by microorganisms include indolacetic acid (Brown and Burlingham 1964; Lee, Breckenridge, and Knowles 1970; Brown 1972; Scott 1972; Barea and Brown 1974; Clark 1974; Tien, Gaskins, and Hubbell 1979), gibberellins (Brown and Burlingham 1964; Katznelson and Shirley 1965; Lee, Breckenridge, and Knowles 1970; Barea and Brown 1974; Tien, Gaskins, and Hubbell 1979), cytokinins (Klamt, Thies, and Skoog 1966; Hegelson and Leonard 1966; Romanov, Chalvignac, and Pochon 1969; Phillips 1970; Phillips and Torrey 1972; Henson and Wheeler 1977; Tien, Gaskins, and Hubbell 1979), kinetin (Tien, Gaskins, and Hubbell 1979), and ethylene (Smith and Russell 1969; Considine, Flynn, and Patching 1977).

34. Some compounds formed by microorganisms are not considered to be growth regulators in the literature. Nonetheless, these substances may have either stimulatory or inhibitory effects on plant growth, depending on the circumstances. These compounds include aliphatic dibasic and tribasic acids, oxy acids, aromatic acids (Takijima 1964a) and nicotinamide, mugeneic acid, and arenic acid (Walker and Welch 1986). Investigations of these compounds have been conducted for purposes other than delineating the influence of rhizosphere microflora on plants. Thus, it is not always known if the formation of these compounds actually occurs in the rhizosphere. However, because the effects are exerted through the roots, they must move through the rhizosphere on their way from the sediments to the roots. Consequently, there is

ample opportunity for the rhizosphere microflora to influence or alter these chemicals.

35. The interactions among soil microorganisms can be complex. For example, certain phenolic acid-utilizing organisms may contribute to total ethylene production in soils having high levels of phenolic polymers (Considine, Flynn, and Patching 1977). Since phenolic acids are toxic (Table 4), consumption of the inhibiting material combined with the stimulation resulting from ethylene production constitutes a twofold benefit from the presence of such organisms. *Penicillium* and *Mucor* spp. are examples of such phenolic acid-utilizing organisms.

36. Allelopathy (biochemical interactions between all types of plants, including microorganisms) within rooted aquatic plants has been reviewed by Szczpanski (1977). The author noted that most types of compounds involved in allelopathy are large molecules, including aldehydes, alkaloids, alcohols, amino acids, enzymes, glycosides, ketones, lactones, nucleic acids, organic acids, sugars, tannins, terpenes, and vitamins. There are several different pathways for release of the compounds, including leaching, guttation, secretion, volatilization, exudation from roots, and release through decomposition. The mechanism of release varies with the environment and the nature of the plant material. Release can occur from leaves, straw, bark, flowers, seeds and fruits, roots, and litter. Some of these compounds are also released directly by microorganisms.

37. Einhellig (1986) provided a description of the relationship between allelopathic activity and processes occurring in the rhizosphere. He indicated that many chemical compounds are volatilized or lost through root exudation and leaching from shoots of actively growing plants and through degradation of residues. Compounds are also released by nonvascular plants and microorganisms. Some of the compounds released into the environment are able to inhibit the growth and development of the receiving plant. Other allelopathic compounds result from microbial transformations. Most allelochemicals are processed through the soil in terrestrial environments, and the compounds reach the target plant by way of the rhizosphere to the roots. This allows the opportunity for direct participation of the rhizosphere microflora in the transformation, enhancement, or weakening of the activity of allelopathic compounds. The extent to which the cycling process holds for aquatic

Table 4  
Plant Growth-Inhibiting Compounds Elaborated by  
Microorganisms in Soils and Sediments

<u>Plant and Associated Microorganisms</u>	<u>Compound(s)</u>	<u>References</u>
Wheat and oats. Several soil microbes	Extracts of rotting wheat straw	Kimber 1967, 1973
Rice. Microbes not specified	Volatile fatty acids	Chandrasekaran and Yoshida 1973
Plants from wet heath. Microbes not specified	Carbon dioxide (negative effect increases as concentration increases)  Hydrogen sulfide	Sheikh 1970
Neither plants nor microbes specified	Phenolic acids (toxic at 50-ppm level)	Wang, Chang, and Chuang 1967
Wheat. <i>Pseudomonas</i> and <i>Syncephalastrum</i>	Phenolic acids	Vaughan, Sparling, and Ord 1983
Barley. Microbes not specified	Compounds in flooded soils and leachates of flooded soils	Takijima 1964b
<i>Hydrilla verticillata</i> . Microbes not specified	Extracts of peatlike sediments	Dooris and Martin 1980
<i>Phragmites</i> . Sediment microbes.	Plant degradation and pollution products	Juttner and Schroeder 1982

sediments has not been established, although some studies have been done with wetland plants.

38. There are several examples of allelopathic activity in wetland and submersed aquatic plants. Szczpanski (1977) noted that autoinhibition occurs in cattail seeds growing on the remains of cattail plants (*Typha latifolia*) and that the same process occurs with *Phragmites australis*. *Myriophyllum spicatum* suppresses growth of *Najas marina* (Agami and Waisel 1985). Yeo and Thurston (1984) reported the effect of spikerush (*Eleocharis coloradensis*) on seven species of submersed aquatic weeds. They found the order of suppression, from most-to-least susceptible, to be horned pondweed (*Zannichella*

*pallustris*), Nuttall's elodea (*Elodea nuttallii*), American elodea (*Elodea canadensis*) hydrilla (*Hydrilla verticillata*), American pondweed (*Potamogeton nodosus*), sago pondweed (*P. pectinatus*), and Eurasian milfoil (*Myriophyllum spicatum*). Spikerush is able to decrease production of subterranean tubers of hydrilla and tubers of sago and American pondweed by more than 50 percent.

39. Zapata and McMillan (1979) found six different phenolic acids in over 50 percent of the seagrasses they examined. The plants studied included *Thalassia testudinum*, *Halodule wrightii*, *Syringodium filiforme*, and *Halophila engelmanni*. The authors noted that the allelopathic role of phenolic compounds in land plants is often cited, while the role in seagrasses is just beginning to be investigated. They suggested that the similarity of phenolic acids in seagrasses indicates that these compounds may play a similar role in both marine and land systems.

40. Several physiological difficulties experienced by plants are a result of the action of low molecular weight metabolites that are produced in or outside of the rhizosphere by noninvasive bacteria and fungi, but affect the plant through the roots. Among the naturally occurring compounds and substances in culture filtrates that have been found to adversely influence plants are hydrogen sulfide, fatty acids, simple alcohols, oxalic acid, phenolic substances including phenylacetic acid and 3-phenyl propionic acids, polysaccharides, and, occasionally, antibiotics (Alexander 1971 and Table 4). Certain habitats allow such toxic substances to persist long enough to affect the plant. These environments include flooded soils and sediments, sites with immediate proximity to decomposing plant litter, and areas where the same crop has been maintained under continuous cultivation for prolonged periods. Among the compounds identified in these habitats are butyric, acetic, p-hydroxybenzoic, p-coumaric, vanillic, and protocatechuic acids, methanol, ethanol, an antibiotic (patullin), and a few cyanogenic (cyanide precursor) substances (Wang, Yang, and Chuang 1967; Alexander 1977). It is important to note that the phytotoxicity of phenolic acids can also be decreased by some soil microorganisms (Vaughan, Sparling, and Ord 1983).

41. The relationship between organic materials derived entirely or in part from the activity of the rhizosphere microflora on sediment substrates and the growth of rooted aquatic plants has only begun to be examined. Dooris and Martin (1980) investigated sediments from two Florida lakes infested with *Hydrilla verticillata*. Using aqueous extracts of sediment from lakes shown to

inhibit the growth of hydrilla in laboratory systems, these workers found that the specific inhibitory fraction of the extract fell in the molecular weight range of 2,000 to 10,000, was highly fluorescent, dark brown in color, and lost activity after passage through an anion-exchange column. The exact nature and origin of the material remains unknown.

#### Factors determining rhizosphere redox status

42. Activities that influence the redox levels in the rhizosphere include those brought about by plants and those brought about by microorganisms. Rooted emergent aquatic plant species often have a lacunar system connecting roots to shoots and leaves. The lacunar system facilitates transport of oxygen to the roots and out into the surrounding sediment, creating an oxidized zone in the rhizosphere. Most often, this oxidized zone is limited to the immediate root vicinity. The oxidized zone surrounding roots of these plants can be visualized in sediments high in iron by a red zone of  $\text{Fe}_2\text{O}_3$  (a rust encrustation) extending out from the root a few millimetres to several centimetres.

43. Activities of microbes that influence redox potential include consumption of dissolved oxygen, denitrification, reduction of manganese, iron, and sulfur, and methanogenesis. Microbial formation of organic redox couples resulting from degradation of organic matter is also known to influence redox status (Brannon et al. 1978, 1984).

44. Plant effects. Teal and Kanwisher (1966) examined gas transport in *Spartina alterniflora*. They found oxygen levels in the roots as low as 3 percent, but levels increased toward the stem. Oxygen diffusing outward from the roots oxidized the sediments immediately surrounding the roots. Carpenter, Elser, and Olsen (1983) examined the effects of the roots of *Myriophyllum verticillatum* on sediment redox conditions. They found that roots of actively photosynthesizing plants gave redox values of +40 to +160 mV, with rates of oxygen release per unit mass similar to ranges reported by other investigators for different plants (0.8 to 5.4 mg  $\text{O}_2$ /mg/hr). Sorrell and Dromgoole (1987) studied oxygen transport in *Egeria densa*. When shoots were illuminated, the average oxygen loss rate from plant roots was 2.97  $\mu\text{l O}_2/\text{hr}/\text{cm}^2$  of plant root surface area. In contrast, root oxygen release rates in the dark were much lower (0.49  $\mu\text{l O}_2/\text{hr}/\text{cm}^2$  root surface area). Penhale and Wetzel (1983) examined the structural and functional adaptations of



eelgrass (*Zostera marina*) to the anaerobic sediment environment. They found that internal gas volumes of leaves and roots increased along a transect of increased sediment anaerobiosis. Physiological adaptations were also characteristics of tolerance to anaerobiosis. The authors concluded that the oxidizing capacity of roots is associated with the survival of these plants. Survival is thought to involve either chemical or biological oxidation of reduced substances in anaerobic sediments. Penhale and Wetzel (1983) noted that a root bacterium, *Beggiatoa*, is capable of decreasing  $H_2S$  concentrations and causing an increase in rate of root oxygen releases (see Table 3).

45. Microbial effects. Microbial consumption of dissolved oxygen followed by utilization of alternate inorganic electron acceptors in sediments causes development of anaerobic conditions and a decrease in the oxidation-reduction potential (see Ponnampertuma 1972; Gunnison, Engler, and Patrick 1985). Under normal circumstances in permanently saturated sediments, anaerobic conditions prevail. Nitrogen release is in the form of ammonium, the favored form of nitrogen for uptake by rooted aquatic plants. Accumulation of carbon dioxide and organic acids from decomposition of organic matter increases the availability of nutrients such as iron, manganese, phosphate, and potassium (Gunnison, Engler, and Patrick 1985). However, hydrogen sulfide and phytotoxic fatty acids and phenolic materials can also accumulate under these conditions (Szczepanski 1977).

46. Sediment oxidation in the rhizosphere of plants that transport an excess of oxygen to the roots is believed to benefit the plants. Oxidized conditions promote chemical and microbial oxidation of harmful levels of ferrous iron, sulfides, and organic compounds (Green and Etherington 1977; Gunnison, Engler, and Patrick 1985). However, oxidation may also reduce levels of dissolved nutrients around the roots (Tessenow and Baynes 1978). An increase in the oxidation-reduction potential may also influence plant establishment and survival (Barko and Smart 1983).

#### Gas formation

47. Gases formed in the sediment are sometimes conducted through the roots of aquatic plants, up the stems, and out into the atmosphere. The gases result from microbial processes occurring in the sediments, although not necessarily in the rhizosphere. Nonetheless, microbial oxidation of the methane, hydrogen, and other labile compounds in the gases can consume some of the oxygen supplied by the plant to the rhizosphere. Data generated through the



study of gas movement from the root up through the waterlily (*Nuphar* sp.) indicate that the waterlily may contain up to 37 percent methane and 6 percent carbon dioxide (Dacey and Klug 1979). Only limited data concerning gas movement through other aquatic species are available.

48. Oremland and Taylor (1977) examined fluctuations of oxygen, nitrogen, and methane in the rhizosphere of *Thalassia testudinum* and also in the surrounding sediment. They found the composition of samples varied in their contents of these gases, depending on the time of collection. Distinctive release patterns were related to the diurnal samples. Oxygen content was lowest in early morning, increased during the day, then decreased again by evening. Methane levels within the rhizome were lower than in the surrounding sediment; this was ascribed to oxidation occurring at the rhizome surface. Since some methane-oxidizing bacteria can fix nitrogen and nitrogen fixation is associated with the rhizomes of *Thalassia testudinum*, the sediment/rhizome interface is probably an environment suitable for development of the methane-oxidizing, nitrogen-fixing bacteria.

49. Ethylene production in the soil and rhizosphere of terrestrial plants is of interest because this gaseous compound is believed to stimulate plant growth (see, for example, Considine, Flynn, and Patching 1977; Samarakoon, Woodrow, and Horton 1985). However, to date there have been no studies of this substance in relation to rooted aquatic plants.

#### Miscellaneous effects

50. The interactions among soil microorganisms can be complex. For example, certain phenolic acid-utilizing organisms may contribute to total ethylene production in soils having high levels of phenolic polymers (Considine, Flynn, and Patching 1977). Since phenolic acids are toxic (Table 4), consumption of the inhibiting material combined with the stimulation resulting from ethylene production constitutes a twofold benefit from the presence of such organisms. *Penicillium* and *Mucor* spp. are examples of such phenolic acid-utilizing organisms.

51. A final area, one that is just beginning to be examined, is that of the protective benefits of microbial associations with plant roots. Colonization of crop plant roots by saprophytic fungi has been implicated in the suppression of fusarium wilt disease of muskmellon (Rouxel, Alabouvette, and Louret 1977, 1979), fusarium yellows of celery (Schneider 1984), and fusarium wilt of cucumber (Paulitz, Park, and Baker 1987). Louvet, Alabouvette, and

Rouxel (1981) have concluded that the mechanism of suppression involves competition between pathogenic and saprophytic varieties of *Fusarium* spp. for ecological occupation of soil niches and for ability to infect the host root. Once the saprophytic strain has colonized the root, the pathogenic strain is unable to move in. Polonenko et al. (1987) pointed out that results from many terrestrial studies indicate certain rhizosphere microorganisms, termed plant growth-promoting rhizobacteria (PGPR), are able to dramatically improve plant growth development and yields in nonlegume crops, such as potato, radish, and sugar beet. Promotion of plant growth results from the ability of PGPR to dominate native soil microbial populations by rapid and aggressive multiplication through the host plant root areas. Certain PGPR are also able to suppress or completely prevent activity of pathogenic soil bacteria and fungi. Polonenko et al. (1987) observed that the PGPR did not interfere with the ability of a nitrogen-fixing bacterium to form nodules, and certain PGPR strains may actually enhance nodulation and plant growth. It is not presently known how commonly this phenomenon occurs or if the colonization of plant roots by fungi influences plant root colonization by bacteria. Whether the phenomenon occurs in the aquatic plant rhizosphere is also unknown.

### Discussion

52. The zone of intense microbial activity in the plant rhizosphere is of interest because an understanding of the processes taking place here may be valuable in developing new measures for management of nuisance plant species. For example, if the presence of nitrogen-fixing bacteria is a requirement for growth of a nuisance plant in a nitrogen-poor sediment, then altering the rhizosphere environment to produce conditions unsuitable for growth of these bacteria will probably make the sediment unsuitable for the growth of the nuisance plant. In like manner, colonization of the root surface by certain microorganisms has been observed to prevent colonization by less desirable or even pathogenic microorganisms. If the rhizosphere environment can be managed to discourage proliferation of the protective microbes or encourage the multiplication of antagonistic species, the beneficial effects of the protection microbes may be prevented. If the nuisance aquatic plant in question is sensitive to the presence of reduced organic compounds or phenolics, then amending the sediment with labile organics may stimulate the formation of the

compounds and retard the growth of the plant. If the rooted aquatic plant is a desirable species, it may be possible to encourage its growth by making sediment conditions favorable to the growth of microorganisms capable of producing plant growth stimulators. Alternatively, it may be possible to stimulate growth at the "wrong" time and allow natural consequences to kill or injure the noxious plant.

53. Sediment is a very complex chemical and microbiological environment, even in the absence of plant roots. The tendency of sediment to develop anaerobic conditions, coupled with the presence of toxic constituents and/or oxygen-demanding conditions, yields a substrate that places considerable stress on any plant using this material as a rooting medium. The situation is complicated by the fact that the physical properties (fine texture) of many sediments make them very unstable media for plants to use for rooting. Plant roots have developed a number of strategies for coping with the physical and chemical aspects of life in aquatic sediments. Certain of the coping strategies utilized by plants have direct impacts on the rhizosphere microflora. For example, the development of root and rhizome systems capable of conducting oxygen sometimes results in more oxygen than necessary to meet the demands of the roots being present in the rhizosphere. The excess oxygen is available to both oxidize the sediments immediately around the roots and to supply rhizosphere microorganisms with oxygen to conduct aerobic processes. Life in an aerobic-anaerobic double layer at or near the root surface has several advantages. Many rhizosphere microorganisms are facultatively anaerobic or microaerophilic. Thus, many resident microbes are more likely to find their preferred concentrations of oxygen in the aerobic-anaerobic double layer. They will also be able to utilize organic compounds released from the plant, while having available reduced inorganic species from the surrounding reduced sediments.

54. Durako and Moffler (1987) and others have suggested that the rhizosphere microflora-plant root interrelationship may be obligatory for nutrition of certain plants, particularly for nitrogen. In some cases, microorganisms and plant roots have an active symbiotic relationship that becomes apparent upon examination of the rhizosphere. This has long been known to be the case with nitrogen-fixing nodules on the roots of leguminous plants. In the case of mycorrhizae, the fungus-plant root structure is easily observed with the use of a microscope. In other situations, a symbiotic relationship may occur,

but may not be easily discerned because of the lack of an obvious physical structure on the plant roots. The occurrence of mutually beneficial interactions within the rhizosphere has been demonstrated for terrestrial and wetland plants, and to a lesser extent for marine plants. This has been demonstrated repeatedly with the nitrogen-fixing *Rhizobium* and leguminous crop plants, where the relationship has been encouraged and developed using many different approaches. In the case of the marine plant *Halodule wrightii* and the bacterium *Klebsiella pneumoniae*, Smith and Hayasaka (1982a) demonstrated the occurrence of this nitrogen-fixing bacterium in surface sterilized roots; this indicated that the microbe was housed within the root and suggested that the microorganism was dependent on the host plant for its source of nutrients. In other cases, the relationship may be fortuitous. Growth of the sulfide-oxidizing *Beggiatoa* in the rhizosphere of rice or a marine plant such as *Zostera marina* may not be mandatory for the bacterium and may or may not be mandatory for the host (sulfide is extremely labile in the presence of oxygen). Alternatively, the host plant in this situation may supply the bacterium with some necessary organic substance, and the bacterium may oxidize enough sulfide to prevent injury to some sulfide-sensitive process. The relationship has not been investigated in detail sufficient to supply the answers.

55. The presence of plant-microbe interactions has not been examined in much detail for freshwater plants. A fair amount is known about wetland plants, although considerably less is known about marine plants. These two habitats are not very far removed from freshwater habitats; wetland plants are often emergent aquatic plants, and several marine species are capable of growing in freshwater, e.g., *Ruppia*. Based on what is known about other environments, these interactions probably also occur in rooted freshwater plants. However, until aquatic plant rhizospheres are thoroughly investigated, there will be uncertainty about what is actually occurring and what role these interactions play in the community structure of nuisance aquatic plant species.

### Conclusions

56. A considerable amount is known about the rhizosphere microbiology of terrestrial and wetland plants, particularly those plants used as crops. It is important to note that wetland plants are aquatic plants, and therefore the

information is probably transferable to submersed aquatic plants. Very little is known about the rhizosphere microbiology of marine plants and very little about the rhizosphere microbiology of freshwater plants.

57. There are several areas of major importance that appear to promise a high level of information return. Based on the literature examined, the following areas appear to be the most promising.

- a. Role of rhizosphere microflora in aquatic plant nutrition.
- b. Production of plant growth-regulating substances by the rhizosphere microflora.
- c. Characterization of the differences in rhizosphere microflora between different rooted aquatic plants occurring in the same sediment and between the same rooted aquatic plant occurring in different sediments.
- d. Evaluation of the role of the rhizosphere microflora in aquatic plant senescence.
- e. Examination of the possible role of plant growth-promoting rhizosphere bacteria in plant growth, development, and productivity and in inhibition of plant pathogens.
- f. Assessment of the role of the rhizosphere microflora in the formation and stabilization or destabilization of organic complexes.

58. Much attention has been given to the process of nitrogen fixation in terrestrial plants, wetland plants, and marine grasses. However, nitrogen fixation in freshwater rooted aquatic plants has not been examined in detail.

#### References

- Abram, J. W. and D. B. Nedwell. 1978. Inhibition of methanogenesis by sulfate reducing bacteria competing for transferred hydrogen. Archives of Microbiology. 117:89-92.
- Agami, M. and Y. Waisel. 1985. Inter-relationships between *Najas marina* L. and three other species of aquatic macrophytes. Hydrobiologia. 126:169-173.
- Alexander, M. 1971. Microbial Ecology. John Wiley and Sons, New York, pp 116-117.
- Alexander, M. 1977. Introduction to Soil Microbiology. 2d ed. John Wiley and Sons, New York, pp 423-437.
- Armstrong, W. 1967. The use of polarography in the assay of oxygen diffusing from roots in anaerobic media. Physiologica Plantarum. 20:540-553.
- Bagyaraj, C. J., A. Manjunath, and R. B. Patil. 1979. Occurrence of vesicular-arbuscular mycorrhizas in some tropical aquatic plants. Transactions of the British Mycological Society. 72:164-167.

- Balasubramanian, A. and G. Rangaswami. 1969. Studies on the influence of foliar nutrient sprays on the root exudation pattern in four crop plants. Plant and Soil. 30:210-220.
- Barea, J. M. and M. E. Brown. 1974. Effects on plant growth produced by *Azotobacter paspali* related to synthesis of plant growth regulating substances. Journal of Applied Bacteriology. 40:583-593.
- Barko, J. W. and R. M. Smart. 1983. Effects of organic matter addition to sediment on the growth of aquatic plants. Journal of Ecology. 71:161-175.
- Barko, J. W., M. S. Adams, and N. L. Clesceri. 1986. Environmental factors and their consideration in the management of submersed aquatic vegetation: A review. Journal of Aquatic Plant Management. 24:1-10.
- Barko, J. W., R. M. Smart, R. L. Chen, and D. G. McFarland. Interactions between macrophyte growth and sediment nutrient availability. Technical Report in preparation, US Army Engineer Waterways Experiment Station, Vicksburg, Miss.
- Barraquio, W. L., J. K. Ladha, and I. Watanabe. 1983. Isolation and identification of  $N_2$ -fixing *Pseudomonas* associated with wetland rice. Canadian Journal of Microbiology. 29:867-873.
- Barraquio, W. L. and I. Watanabe. 1981. Occurrence of aerobic  $N_2$ -fixing bacteria in wetland and dryland plants. Soil Science and Plant Nutrition. 27:121-125.
- Blotnick, J. R., J. Rho, and H. B. Gunner. 1980. Ecological characteristics of the rhizosphere microflora of *Myriophyllum heterophyllum*. Journal of Environmental Quality. 9:207-210.
- Boon, P. I., D. J. W. Moriarity, and P. G. Saffigna. 1986. Rates of ammonium turnover and the role of amino-acid deamination in seagrass (*Zostera capricorni*) beds of Moreton Bay, Australia. Marine Biology. 91:259-268.
- Brannon, J. M., D. Gunnison, P. L. Butler, and I. Smith, Jr. 1978. Mechanisms that regulate the intensity of oxidation-reduction in anaerobic sediments and natural water systems. Technical Report Y-78-11. US Army Engineer Waterways Experiment Station, Vicksburg, Miss.
- Brannon, J. M., D. Gunnison, R. M. Smart, and R. L. Chen. 1984. Effects of added organic matter on iron and manganese redox systems in sediment. Geomicrobiology Journal. 3:319-341.
- Bristow, J. M. 1974. Nitrogen fixation in the rhizosphere of freshwater angiosperms. Canadian Journal of Botany. 52:217-221.
- Brown, M. E. 1972. Plant growth substances produced by microorganisms of soil and rhizosphere. Journal of Applied Bacteriology. 43:443-451.
- Brown, M. E. and S. W. Burlingham. 1964. Production of plant growth substances by *Azotobacter chroococcum*. Journal of General Microbiology. 53:135-144.
- Burrell, D. C. and J. R. Schubel. 1977. Seagrass ecosystem oceanography, pp 195-232. In: C. P. McRoy and C. Hefferich (eds.), Seagrass Ecosystems: A Scientific Perspective. Marcel Dekker, New York.
- Capone, D. G. 1983.  $N_2$  fixation in seagrass communities. Marine Technology Society Journal. 17:32-37.

- Carignan, R. 1985. Nutrient dynamics in a littoral sediment colonized by the submersed macrophyte *Myriophyllum spicatum*. Canadian Journal of Fisheries and Aquatic Science. 42:1303-1311.
- Carpenter, S. R., J. J. Elser, and K. M. Olsen. 1983. Effects of roots of *Myriophyllum verticillatum* L. on sediment redox conditions. Aquatic Botany. 17:243-249.
- Chandrasekaran, S. and T. Yoshida. 1973. Effect of organic acid transformations in submerged soils on growth of the rice plant. Soil Science and Plant Nutrition. 19:39-45.
- Chaubal, R., G. D. Charma, and R. R. Mishra. 1982. Vesicular-arbuscular mycorrhiza in subtropical aquatic and marshy plant communities. Proceedings of the Indian Academy of Sciences. 91:69-77.
- Clark, A. G. 1974. Indole acetic acid production by *Agrobacterium* and *Rhizobium* species. Microbios. 11A:29-35.
- Clayton, J. S. and D. J. Bagyaraj. 1984. Vesicular-arbuscular mycorrhizas in submerged aquatic plants of New Zealand. Aquatic Botany. 19:251-262.
- Considine, P. J., N. Flynn, and P. W. Patching. 1977. Ethylene production by soil microorganisms. Applied and Environmental Microbiology. 33:977-979.
- Coult, D. A. 1964. Observations on gas movement in the rhizome of *Menyanthes trifoliata* with comments on the role of the endodermis. Journal of Experimental Botany. 15:205-218.
- Craven, P. A. and S. S. Hayasaka. 1982. Inorganic phosphate solubilization by rhizosphere bacteria in a *Zostera marina* community. Canadian Journal of Microbiology. 28:605-610.
- Dacey, J. W. H. 1980. Internal winds in water lilies: An adaptation for life in anaerobic sediment. Science. 210:1017-1019.
- Dacey, J. W. H. and M. J. Klug. 1979. Methane efflux from lake sediments through water lilies. Science. 203:1253-1255.
- Dooris, P. M. and D. F. Martin. 1980. Growth inhibition of *Hydrilla verticillata* by selected lake sediment extracts. Water Resources Bulletin. 16:112-117.
- Drew, M. C. and J. M. Lynch. 1980. Soil anaerobiosis, microorganisms, and root function. Annual Review of Phytopathology. 18:37-66.
- Durako, M. J. and M. D. Moffler. 1987. Nutritional studies of the submerged marine angiosperm *Thalassia testudinum*. I. Growth responses of axenic seedlings to nitrogen enrichment. American Journal of Botany. 74:234-240.
- Einhellig, F. A. 1986. Mechanisms and modes of action of allelochemicals, pp 171-188. In: A. Putman and C. S. Tang (eds.), The Science of Allelopathy. John Wiley and Sons, New York.
- Elias, K. S. and G. R. Safir. 1987. Hyphal elongation of *Glomus fasciculatus* in response to root exudates. Applied and Environmental Microbiology. 53:1928-1933.
- Engler, R. M. and W. H. Patrick, Jr. 1973. Sulfate reduction and sulfide oxidation in flooded soils as affected by chemical oxidants. Proceedings of the Soil Science Society of America. 37:685-688.



- Gardes, M., J. Bousquet, and M. Lalonde. 1987. Isozyme variation among 40 *Frankia* strains. Applied and Environmental Microbiology. 53:1596-1603.
- Ginsburg, R. N. and H. A. Lowenstam. 1958. The influence of marine bottom communities on the depositional environment of sediment. Journal of Geology. 66:310-318.
- Green, M. S. and J. R. Etherington. 1977. Oxidation of ferrous iron by rice (*Oryza sativa* L.) roots: A mechanism for waterlogging tolerance? Journal of Experimental Botany. 28:678-690.
- Gregg, W. W. and F. L. Rose. 1982. The effect of aquatic macrophytes on the stream microenvironment. Aquatic Botany. 14:309-324.
- Gunnison, D., R. M. Engler, and W. H. Patrick, Jr. 1985. Chemistry and microbiology of newly flooded soils: Relationship to reservoir water quality, pp 39-57. In: D. Gunnison (ed.), Microbial Processes in Reservoirs. Dr. W. Junk, Publishers, The Hague, Netherlands.
- Hale, M. G., L. L. Foy, and F. J. Shay. 1971. Factors affecting root exudation. Advances in Agronomy. 23:89-109.
- Hegelson, J. P. and N. J. Leonard. 1966. Cytokinins: Identification of compounds isolated from *Corynebacterium fascians*. Proceedings of the National Academy of Science of the United States of America. 56:60-63.
- Henson, J. E. and C. T. Wheeler. 1977. Hormones in plants bearing nitrogen fixing root nodules: Partial characterization of cytokinins from root nodules of *Alnus glutinosa* (L.) Gaertn. Journal of Experimental Botany. 28:1076-1086.
- Iizumi, H., A. Hattori, and C. P. McRoy. 1980. Nitrate and nitrite in interstitial waters of eelgrass beds in relation to the rhizosphere. Journal of Experimental Marine Biology and Ecology. 47:191-201.
- Jansson, M. 1987. Anaerobic dissolution of iron-phosphorus complexes in sediment due to the activity of nitrate-reducing bacteria. Microbial Ecology. 14:81-89.
- Joshi, M. M. and J. P. Hollis. 1977. Interaction of *Beggiatoa* and rice plant: Detoxification of hydrogen sulfide in the rice rhizosphere. Science. 195:179-180.
- Juttner, F. and R. Schroeder. 1982. Microbially-derived volatile organic compounds in the recent sediment of the *Phragmites australis* bed of the Bodensee (Lake Constance). Archives of Hydrobiology. 94:172-181.
- Katznelson, H. and E. C. Shirley. 1965. Production of gibberellin-like substances by bacteria and actinomycetes. Canadian Journal of Microbiology. 11:733-741.
- Kenworthy, W. J., C. Currin, G. Smith, and G. Thayer. 1987. The abundance, biomass and acetylene reduction activity of bacteria associated with decomposing rhizomes of two seagrasses, *Zostera marina* and *Thalassia testudinum*. Aquatic Botany. 27:97-119.
- Kenworthy, W. J., J. C. Zieman, and G. W. Thayer. 1982. Evidence for the influence of seagrasses on the benthic nitrogen cycle in a coastal plain estuary near Beaufort, North Carolina (USA). Oecologia. 54:152-158.



- Khan, A. G. 1974. The occurrence of mycorrhizas in halophytes, hydrophytes, and xerophytes, and of endogone spores in adjacent soils. Journal of General Microbiology. 81:7-14.
- Kimber, R. W. L. 1967. Phytotoxicity from plant residues. I. The influence of rotted wheat straw on seedling growth. Australian Journal of Agricultural Research. 18:361-374.
- Kimber, R. W. L. 1973. Phytotoxicity from plant residues. II. The effect of time of rotting of straw from some grasses and legumes on the growth of wheat seedlings. Plant and Soil. 38:347-361.
- Klamt, D., D. Thies, and F. Skoog. 1966. Isolation of cytokinins from *Corynebacterium fascians*. Proceedings of the National Academy of Sciences of the United States of America. 56:52-59.
- Laing, E. H. 1940. The composition of the internal atmosphere of *Nuphar advenum* and other water plants. American Journal of Botany. 27:861-868.
- Lee, M., C. Breckenridge, and R. Knowles. 1970. Effect of some culture conditions on the production of indole-3-acetic acid and gibberellin-like substances by *Azotobacter vinelandii*. Canadian Journal of Microbiology. 16:1325-1330.
- Lifshitz, R., J. W. Kloepper, F. M. Scher, E. M. Tipping, and M. LaLiberte. 1986. Nitrogen-fixing pseudomonads isolated from roots of plants grown in the Canadian High Arctic. Applied and Environmental Microbiology. 51:251-255.
- Louvet, J., C. Alabouvette, and F. Rouxel. 1981. Microbiological suppressiveness of some soils of fusarium wilts, pp 261-275. In: P. E. Nelson, T. A. Toussoun, and R. J. Cook (eds), Fusarium: Diseases, Biology, and Taxonomy. The Pennsylvania State University Press, University Park.
- Lynch, J. M. 1979. The terrestrial environment, pp 67-91. In: J. M. Lynch and N. J. Poole (eds.), Microbial Ecology: A Conceptual Approach. Halstead Press Division of John Wiley and Sons, New York.
- Lyon, T. L. and J. K. Wilson. 1921. Liberation of organic matter by roots of growing plants. Memoirs of the New York Agricultural Experiment Station. 40:1-44.
- Macura, J. 1968. Physiological studies of rhizosphere bacteria, pp 379-395. In: T. R. G. Gray and D. Parkinson (eds.), The Ecology of Soil Bacteria. University of Toronto Press, Toronto.
- McClung, C. R., P. van Berkum, R. E. Davis, and C. Sloger. 1983. Enumeration and localization of N<sub>2</sub>-fixing bacteria associated with roots of *Spartina alterniflora* Loisel. Applied and Environmental Microbiology. 45:1914-1920.
- Muller, P. J. 1977. C/N ratios in Pacific deep-sea sediments: Effect of inorganic ammonium, and organic nitrogen compounds sorbed by clays. Geochimica Cosmochimica Acta. 41:765-776.
- Oremland, R. S. and B. F. Taylor. 1977. Diurnal fluctuations of O<sub>2</sub>, N<sub>2</sub>, and CH<sub>4</sub> in the rhizosphere of *Thalassia testudinum*. Limnology and Oceanography. 22:566-570.
- Paulitz, T. C., C. S. Park, and R. Baker. 1987. Biological control of fusarium wilt of cucumber with nonpathogenic isolates of *Fusarium oxysporium*. Canadian Journal of Microbiology. 33:349-353.

- Penhale, P. A. and R. G. Wetzel. 1983. Structural and functional adaptations of eelgrass (*Zostera marina* L.) to the anaerobic sediment environment. Canadian Journal of Botany. 61:1421-1428.
- Pennak, R. W. 1971. Toward a classification of lotic habitats. Hydrobiologia. 38:321-334.
- Pennington, J. C. 1986. Feasibility of using mycorrhizal fungi for enhancement of plant establishment on dredged material disposal sites: A literature review. Miscellaneous Paper D-86-3. US Army Engineer Waterways Experiment Station, Vicksburg, Miss.
- Phillips, D. A. 1970. Cytokinin production of *Rhizobium japonicum*. Physiologica Plantarum. 23:1057-1063.
- Phillips, D. A. and J. G. Torrey. 1972. Studies on cytokinin production by *Rhizobium*. Plant Physiology. 49:11-15.
- Polonenko, D. R., F. M. Scher, J. W. Kloepper, C. A. Singleton, M. LaLiberte, and I. Zaleska. 1987. Effects of root colonizing bacteria on nodulation of soybean roots by *Bradyrhizobium japonicum*. Canadian Journal of Microbiology. 33:498-503.
- Ponnamperuma, F. N. 1972. The chemistry of submerged soils. Advances in Agronomy. 24:29-88.
- Prentki, R. T. 1979. Depletion of phosphorus from sediment colonized by *Myriophyllum spicatum* L., pp 161-176. In: J. E. Breck, R. T. Prentki, and O. L. Loucks (eds.), Aquatic Plants, Lake Management, and Ecosystem Consequences of Lake Harvesting. Institute for Environmental Studies, University of Wisconsin, Madison.
- Read, D. J., et al. 1976. Vesicular-arbuscular mycorrhizae in natural vegetation systems. New Phytologist. 77:641-653.
- Reinhold, B., T. Hurek, E. G. Nieman, and I. Fendrik. 1986. Close association of *Azospirillum* and diazotrophic rods with different root zones of Kallar grass. Applied and Environmental Microbiology. 52:520-526.
- Romanov, I., M. A. Chalvignac, and J. Pochon. 1969. Recherches sur la production d'une substance cytokinique pour *Agrobacterium tumefaciens* (Smith et Town) Conn. Annals Institute Pasteur (Paris). 117:58-63.
- Rosenfeld, J. K. 1979. Amino acid diagenesis and adsorption in nearshore anoxic sediments. Limnology and Oceanography. 24:1014-1021.
- Rouxel, F., C. Alabouvette, and J. Louvet. 1977. Recherches sur la resistance des sols aux maladies. II. Incidence de traitements thermiques sur la resistance microbiologique d'une sol a la Fusariose vasculaire du melon. Annals of Phytopathology. 9:183-192.
- Rouxel, F., C. Alabouvette, and J. Louvet. 1979. Recherches sur la resistance des sols aux maladies. IV. Mise en evidnece du role des *Fusarium* autochtones dans la resistance d'une sol a la Fusariose vasculaire du melon. Annals of Phytopathology. 11:199-207.
- Rovira, A. D. 1956a. Plant root excretions in relation to the rhizosphere effect. I. The nature of root exudate from oats and peas. Plant and Soil. 7:178-194.

- Rovira, A. D. 1956b. Plant root excretions in relation to the rhizosphere effect. II. A study of the properties of root exudate and its effect on the growth of micro-organisms isolated from the rhizosphere and control soil. Plant and Soil. 7:195-208.
- Rovira, A. D. 1969. Plant root exudates. Botanical Review. 35:35-57.
- Russell, E. W. 1968. The agricultural environment of soil bacteria. In: T. R. G. Gray and D. Parkinson (eds.), The Ecology of Soil Bacteria. University of Toronto Press, Toronto, Canada.
- Samarakoon, A. B., L. Woodrow, and R. F. Horton. 1985. Ethylene- and submergence-promoted growth in *Ranunculus scleratus* L. petioles: The effect of cobalt ions. Aquatic Botany. 21:33-41.
- Sanderson, P. L. and W. Armstrong. 1980. Phytotoxins in periodically waterlogged forest soils. Journal of Soil Science. 31:643-653.
- Sand-Jensen, K. and C. Prahl. 1982. Oxygen exchange with the lacunae and across leaves and roots of the submerged vascular macrophyte *Lobelia dortmanna* L. New Phytologist. 91:103-120.
- Sand-Jensen, K., C. Prahl, and H. Stockholm. 1982. Oxygen release from roots to submerged aquatic macrophytes. Oikos. 38:349-354.
- Sargent, L., S. H. Huang, B. G. Role, and M. A. Djordjevic. 1987. Split-root assays using *Trifolium subterraneum* that includes a systemic response that can inhibit nodulation of another invasive *Rhizobium* strain. Applied and Environmental Microbiology. 53:1611-1619.
- Schmidt, M. A. and S. S. Hayasaka. 1985. Localization of a dinitrogen-fixing *Klebsiella* sp. isolated from root-rhizomes of the seagrass *Halodule wrightii* Aschers. Botanica Marina. 28:437-442.
- Schneider, R. W. 1984. Effects of nonpathogenic strains of *Fusarium oxysporum* f. sp. *appi* and a novel use of the Lineweaver-Burke double reciprocal plot technique. Phytopathology. 74:646-653.
- Scott, T. K. 1972. Auxins and roots. Annual Review of Plant Physiology. 23:235-258.
- Sculthorpe, C. D. 1967. The Biology of Aquatic Vascular Plants. Edward Arnold Publishers, Ltd., London.
- Sheikh, K. H. 1970. The responses of *Molina caerulea* and *Erica tetralix* to soil aeration and related factors. III. Effects of different gas concentrations on growth in solution culture, and general conclusions. Journal of Ecology. 58:141-154.
- Smith, G. W. and S. S. Hayasaka. 1982a. Nitrogenase activity associated with *Halodule wrightii* roots. Applied and Environmental Microbiology. 43:1244-1248.
- Smith, G. W. and S. S. Hayasaka. 1982b. Nitrogenase activity associated with *Zostera marina* from a North Carolina estuary. Canadian Journal of Microbiology. 28:448-451.
- Smith, G. W., S. S. Hayasaka, and G. W. Thayer. 1984. Ammonification of amino acids by the rhizoplane microflora of *Zostera marina* L. and *Halodule wrightii* Aschers. Botanica Marina. 27:23-27.

- Smith, K. A. and R. S. Russell. 1969. Occurrence of ethylene, and its significance, in anaerobic soil. Nature. 222:769-771.
- Smith, R. D., W. C. Dennison, and R. S. Alberte. 1984. Role of seagrass photosynthesis in root aerobic processes. Plant Physiology. 74:1055-1058.
- Sondergaard, M. and S. Laegaard. 1977. Vesicular-arbuscular mycorrhizae in some aquatic vascular plants. Nature. 268:232-233.
- Sorrell, B. K. and F. I. Dromgoole. 1987. Oxygen transport in the submerged freshwater macrophyte *Egeria densa* Planch. I. Oxygen production, storage and release. Aquatic Botany. 28:63-80.
- Szczepanski, A. J. 1977. Allelopathy as a means of biological control of water weeds. Aquatic Botany. 3:193-197.
- Takijima, Y. 1964a. Studies on organic acids in paddy field soils with reference to their inhibitory effects on the growth of rice plants. Part 1. Growth inhibitory action of organic acids and absorption and decomposition of them by soils. Soil Science and Plant Nutrition. 10:14-21.
- Takijima, Y. 1964b. Studies on organic acids in paddy field soils with reference to their inhibitory effects on the growth of rice plants. Part 2. Relations between production of organic acids in waterlogged soils and the root growth inhibition. Soil Science and Plant Nutrition. 10:22-29.
- Teal, J. M. and J. W. Kanwisher. 1966. Gas transport in the marsh grass *Spartina alterniflora*. Journal of Experimental Botany. 17:355-361.
- Tessenow, V. U. and Y. Baynes. 1978. Redoxchemische Einflüsse von *Isoetes lacustris* L. im Litoralsediment des Feldsees (Hochschwarzwald). Archives of Hydrobiology. 82:20-48.
- Tien, T. M., M. H. Gaskins, and D. H. Hubbell. 1979. Plant growth substances produced by *Azospirillum brasilense* and their effect on the growth of pearl millet (*Pennisetum americanum* L.). Applied and Environmental Microbiology. 37:1016-1024.
- Tjepkema, J. D. and H. J. Evans. 1976. Nitrogen fixation associated with *Juncus balticus* and other plants of Oregon wetlands. Soil Biology and Biochemistry. 8:505-509.
- Vagnerova, K. and V. Vancura. 1962. Production and utilization of amino acids by various species of rhizosphere bacteria. Folia Microbiologia. 7:55.
- Vaughan, D., G. P. Sparling, and B. G. Ord. 1983. Amelioration of the phytotoxicity of phenolic acids by some soil microbes. Soil Biology and Biochemistry. 15:613-614.
- Vesper, S. J. 1987. Production of pili (Fimbriae) by *Pseudomonas fluorescens* and correlation with attachment to corn roots. Applied and Environmental Microbiology. 53:1397-1405.
- Walker, C. D. and R. M. Welch. 1986. Nicotinamide and related siderophores: Their physiological significance and advantages for plant metabolism. Journal of Plant Nutrition. 9:523-524.
- Wang, T. S. C., T.-K. Yang, and T.-T. Chuang. 1967. Soil phenolic acids as plant growth inhibitors. Soil Science, 103:239-246.

- Watanabe, I. and W. L. Barraquio. 1979. Low levels of fixed nitrogen required for isolation of free-living  $N_2$ -fixing organisms from rice roots. Nature. 277:565-566.
- Watanabe, I., K. K. Lee, B. V. Alimagno, M. Sato, D. C. del Rosario, and M. R. de Guzman. 1977. Biological nitrogen fixation in paddy field studied by in situ acetylene-reduction assays. IRRI Research Paper Series. 3:1-16.
- Watanabe, I., W. Barraquio, M. Guzman, and D. Cabrera. 1979. Nitrogen-fixing (acetylene reduction) activity and population of aerobic heterotrophic nitrogen-fixing bacteria associated with rice. Applied and Environmental Microbiology. 37:813-819.
- Wiley, M. J., R. W. Gordon, S. W. Waite, and T. Powless. 1984. The relationship between aquatic macrophytes and sport fish production in Illinois ponds: A sample model. North American Journal of Fishery Management. 4:111-119.
- Yeo, R. R. and J. R. Thurston. 1984. The effect of dwarf spikerush (*Eleocharis coloradoensis*) on several submersed aquatic weeds. Journal of Aquatic Plant Management. 22:52-56.
- Zapata, O. and C. McMillan. 1979. Phenolic acids in seagrasses. Aquatic Botany. 7:307-317.