(0.25 mg/l). These calli were then tested at 0, 20, 40, 80, 10 and 200 μM glyphosate treatment on EPM medium containing ABA 2.5 mg/l. Somatic embryos formed within two weeks after calli, were transferred to formation EPM containing 100 μM, and 200 μM glyphosate. However on those control explants cultured on medium with ABA alone and without glyphosate, embryos appeared only after three weeks. The number of embryos produced per explant was highest in 200 μM and least in 20 μM glyphosate (Table 1). The lower levels of glyphosate (20, 40 and 80 μM) had an inhibitory effect on embryo initiation and development; this result is difficult to explain and may be due to the interaction of ABA with glyphosate up to 80 μM. Somatic embryos formed in medium containing higher concentrations of glyphosate (100 and 200 μM) clearly appeared desiccated. The epidermal layer of the leaf appeared peeled off, and the embryos arose from the sub-epidermal layer of the leaf. The epidermal layer in the control explants may hinder the somatic embryo development and the corrosion of such layers by glyphosate treatment has perhaps facilitated easier protrusion of somatic embryos. The torpedo-stage embryos differentiated faster in the glyphosate-treated calli, and this was in contrast to the control where most embryos germinated pre- cociously. Such precocious germination hinders the full utilization of somatic embryos in many crop plants[5]. In our study, glyphosate at both 100 μM and 200 μM effectively controlled the precocious germination. Although embryos obtained from 100 μM as well as 200 μM glyphosate treatments appeared normal, those from 200 μM treatment showed the highest percentage of germination and grew more vigorously. Somatic embryos derived from glyphosate treatments of 100 μM and 200 μM also exhibited a higher conversion rate *ex vitro* to normal plantlets (Table 1).

The presence of 2,4-D, BAP and ABA in the medium was essential to somatic embryogenesis and development. When various concentrations of glyphosate were included in the CPM, but 2,4-D and BAP were not included, callus formation was not observed in the sweetpotato explants. Such explants also did not produce any somatic embryos when subsequently transferred on to EPM. Further, the use of glyphosate alone (without ABA) in EPM did not result in embryo production on explants cultured on CPM with 2,4-D and BAP. Thus, glyphosate by itself may not aid either in the initiation or the subsequent development of somatic embryos, but appears to act in conjunction with ABA in enhancing the number and quality of somatic embryos in sweetpotato. Glyphosate inhibits the activity of 5-enolpyruvyl shikimic acid-3-phosphate (EPSP) synthase, a key enzyme of shikimic acid pathway. However, there are many other effects of glyphosate on plant metabolism[9]. Glyphosate is known to rapidly decrease the levels of IAA and inhibits IAA transport[10]. While auxin is required for the initiation of somatic embryos, it is often detrimental to further histodifferentiation[11]. Thus glyphosate may have facilitated somatic embryo development by depleting the auxin reserves in the embryos. It is also possible that glyphosate may have simply facilitated the developmental arrest of somatic embryos by promoting desiccation via synergistic interaction with ABA. Glyphosate may have also enhanced the embryo development through its disruption of epidermal layer so that embryos arising from sub-epidermal layers would develop without any physical interference. Further studies are needed to identify the mechanisms by which glyphosate improves the somatic embryo production in sweetpotato.

1. Zheng, Q., Porobo Dessai, A. and Prakash, C. S., *Plant Cell Rep.*, 1996, **15**, 381–385.
2. Prakash, C. S., *Biotechnol. Dev. Monitor*, 1994, **18**, 18–22.
3. Finkelstein, R. R. and Crouch, M. L., *Planta*, 1984, **162**, 125–131.
4. Gray, D. J., *Hortic. Sci.*, 1987, **22**, 810–812.
5. Lee, C. W. and Thomas, J. C., *Hortic. Sci.*, 1985, **20**, 267.
6. Carman, J. G., *In Vitro Cell Dev. Biol. Plant*, 1990, **26**, 746–753.
7. Ammirato, P. V., Evans, D. A., Sharp, W .R. and Yamada, Y, *Handbook of Plant Cell Culture*, Macmillan Publishing Co., New York, 1984.
8. Murashige, T. and Skoog, F., *Physiol. Plant*, 1962, **15**, 473–497.
9. Duke, S. O., *Glyphosate in Herbicides: Chemistry, Regulation and Mode of Action* (eds Kearney, P. C. and Kaufman, D. D.), Academic Press, New York, 1987, pp. 2–71.
10. Lee, T. T., *J. Plant Growth Regulators*, 1982, **1**, 37–38.
11. Merkle, S. A., Parrot, W. A. and Flinn, B. S., *In Vitro Embryogenesis in Plants* (ed. Thorpe, T. T.), Kluwer Academic Publishers, Dordrecht, The Netherlands, 1995, pp. 173–203

ACKNOWLEDGEMENTS. We thank Dr Marceline Egnin for her assistance during the study.PHRG was initially supported by a fellowship from UNESCO/BAC. Research supported by USDA/CSREES and NASA is gratefully acknowledged.

Received 10 March 1998; revised accepted 30 June 1998

Soil enzymes in the mangroves: Activities and their relation to relevant soil properties

R. Dinesh, B. R. Shome, Rajeshwari Shome and A. K. Bandyopadhyay

Central Agricultural Research Institute, Indian Council of Agricultural Research, P.B. No. 181, Port Blair 744 101, India

We report here some basic but significant observations on enzyme activities in soils of a few prominent mangroves in South Andamans. In the present study, soil enzymes important in nutrient cycling were

assayed in 5 major mangroves of South Andamans, and their activities related to relevant soil properties to derive relationships, if any. Overall, the mean enzyme activities varied among soils possibly due to variations in the levels of endoenzymes and accumulated enzymes in the soil matrix. Nonsignificant relationship existed between enzyme activities and soil pH. However, all the enzymes showed positive and significant relationships with organic nutrient forms like organic C, organic P and organic S, which indicated that soils with higher organic C stimulated microbial activity and, therefore, provided a more conducive environment for enzyme synthesis and accumulation in the soil matrix of the mangroves.

THE mangrove forests are one of the major ecosystems of the biosphere. About 60–70% of the tropical coasts are covered by this type of ecosystem. The literature on mangroves consists of roughly more than 6000 references[1]. One could, therefore, easily reach to the conclusion that this ecosystem is indeed well documented and understood and that not much is left to be said. However, the great majority of these studies refer to flora, microbial ecology, plant and animal physiology, sedimentology, nutrient status, conservation and reclamation.

It is well known that all biochemical reactions important in nutrient cycling/recycling in the soil are catalysed by enzymes which are proteins with catalytic properties owing to their power of specific activation[2]. Surprisingly, there is no literature that has documented the activity of soil enzymes in the mangroves. Since microbial aspects of nutrient cycling in mangrove soils is a vital and crucial field of study if we are to fully understand and appreciate the mangrove ecosystem, assessing soil enzyme activities assumes greater significance. We, therefore, report here some significant observations on selected enzyme activities that play an active role in nutrient cycling in soils of the mangroves. Besides, we have also tried to bring out the degree of relationship between relevant soil properties and enzyme activities.

About 15 soil cores each (0–30 cm) were collected from 5 prominent mangroves of South Andamans during periods of high tide. The soil cores were then packed in polythene bags and brought to the laboratory. The individual core samples from each mangrove were then combined, cleared of any organic debris, and stored at 4°C. While a portion of each soil sample (< 2 mm) was used for determining relevant soil properties (Table 1), another portion was stored at 4°C for no longer than 1 week before enzyme assay. Soils were analysed for pH (soil to solution ratio 1 : 2.5), and the other parameters were determined using standard methods[3]. The enzyme assays (Table 2) were performed on moist samples throughout. All values reported are means of 15 determinations expressed on dry weight basis.

Enzyme assay indicates wide variation in enzyme activities among soils (Table 3). This might be due to variation in the amount of endoenzymes in the viable microbial population and variation in the levels of accumulated enzymes in the soil matrix. Because enzyme activity has shown to be highly correlated with both microbial respiration and total biomass in soil[6], higher

Table 1. Relevant properties of soils of some prominent mangroves of South Andamans

Mangroves	pH	Organic C (g/kg)	Total N (g/kg)	Total P (mg/kg)	Organic P (mg/kg)	Total S (mg/kg)	Organic S (mg/kg)
Shaitankadi	3.8	23.7	2.04	201.2	166.0	544.0	475.8
Sippighat	5.2	6.5	0.56	100.8	83.2	321.0	239.6
Wandoor	3.5	21.7	1.87	195.2	168.8	549.6	481.4
Manjeri	4.0	16.1	1.39	182.6	162.0	509.8	439.6
Chidiatapu	8.7	14.4	1.24	137.6	112.4	411.4	333.6

Table 2. Details of the methods used for enzyme assay

Soil enzyme	Method	Substrate	End product	Incubation period (h)*
Urease	2	Urea	NH_4-N	2
Phosphatase	2	*p*-Nitrophenyl phosphate	*p*-Nitrophenol	1
Aryl sulphatase	2	*p*-Nitrophenyl sulphate	*p*-Nitrophenol	1
Dehydrogenase	2	2,3,5-Triphenyl tetrazolium chloride	Triphenyl formazan	24
β-Glucosidase	2	*p*-Nitrophenyl β-D-glucoside	NH_4-N	1
Inorganic pyrophosphatase	2	Sodium pyrophosphate	Pyrophosphate	5
Amidase	4	Acetamide	NH_4-N	24
L-glutaminase	5	L-glutamine	NH_4-N	3

*All assays were done at incubation temperature of 37°C.

Table 3. Enzyme activities in some prominent mangrove soils of South Andamans

Mangrove	Urease[a]	Amidase[b]	Phosphatase[c]		Dehydro-genase[d]	β-Glucosidase[e]	L-Glutaminase[f]	Aryl sulphatase[g]	Inorganic pyro-phosphatase[h]
			Acid	Alkaline					
Shaitankadi	290.8	301.1	1053.0	600.0	71.6	262.4	613.8	63.2	369.9
Sippighat	177.4	170.3	798.7	476.7	37.3	173.9	394.6	32.6	103.3
Wandoor	278.3	283.7	945.9	532.8	76.3	258.2	594.6	52.4	304.5
Manjeri	254.2	259.8	877.4	482.3	71.5	244.6	613.5	63.0	266.3
Chidiatapu	222.6	222.2	796.6	459.8	53.3	209.9	491.9	48.2	203.7

[a]μg NH_4-N/g soil/2 h; [b]μg NH_4-N/g soil/24 h; [c]μg *p*-nitrophenol/g soil/h; [d]μg TPF/g soil/24 h; [e]μg NH_4- N/g soil/1 h; [f]μmol NH_4-N/g soil/3 h; [g]μg *p*-nitrophenol/g soil/h; [h]μg *p*-nitrophenol/g soil/5 h

Table 4. Simple correlation coefficients (r)[a] between enzyme activities and soil properties

	pH	Organic C	Total N	Total P	Organic P	Total S	Organic S
Urease	−0.536	0.987**	0.987**	0.989**	0.964**	0.985**	0.985**
Amidase	−0.512	0.984**	0.984**	0.989**	0.951**	0.973**	0.977**
Acid phosphatase	−0.680	0.880*	0.880*	0.850*	0.800	0.823*	0.825*
Alkaline phosphatase	−0.627	0.787	0.786	0.711	0.643	0.678	0.680
Dehydrogenase	−0.566	0.913*	0.914*	0.984**	0.993**	0.992**	0.992**
β-Glucosidase	−0.564	0.963**	0.964**	0.998**	0.988**	0.988**	0.998**
L-Glutaminase	−0.545	0.895*	0.896*	0.981*	0.986**	0.977**	0.977**
Aryl sulphatase	−0.396	0.820*	0.821*	0.903*	0.898*	0.885*	0.884*
Inorganic pyrophosphatase	−0.507	0.982**	0.983**	0.971**	0.933**	0.958**	0.958**

[a]$n = 5$; *at $P < 0.05$; **at $P < 0.01$.

enzyme activities in some soils suggest that greater biological activity has been established in these soils. The data on activity of dehydrogenase also support this claim because dehydrogenase activity is more dependent on the biological activity of the microbial population than on any free enzyme present[7].

Simple correlations obtained between enzyme activities and relevant soil properties (Table 4) indicate a non-significant relationship between enzyme activities and soil pH. Except for phosphatases (acid and alkaline), which are sensitive to soil pH (ref. 8), the relationship between the other enzymes and soil pH was found to be nonsignificant in a number of earlier studies[9–11]. Significant and positive relationship, however, existed between enzyme activities and other soil properties like soil organic C, total N, organic P and organic S (Table 4). Higher organic C levels stimulate microbial activity and, therefore, enzyme synthesis and accumulation. Besides, higher organic C levels in soils may also provide a more conducive environment for accumulation of enzymes in the soil matrix, since soil organic constituents are thought to be important in forming stable complexes with free enzymes[12].

In conclusion, it can be deduced that soil pH apparently did not influence soil enzyme activities in the mangroves. However, organic nutrient forms like organic P, organic S and more importantly organic C markedly influenced enzyme activity, synthesis and accumulation in the soil matrix. Though the data presented give only a broad idea on enzyme activities in soils of the mangroves and their relation to certain soil parameters, the results will, however, be useful in keeping a tab on the potential of mangrove soils in harnessing the capabilities of these enzymes for the benefit and betterment of human life.

1. Singh, V. P. and Garge, G., in *Ecology of Mangrove Swamps of Andaman Islands*, International Book Distributors, Dehradun, 1993.
2. Tabatabai, M. A., in *Methods of Soil Analysis, Part 2, Chemical and Microbiological Properties*, 2nd edn, Agron 9, ASA, SSSA, Madison, WI, 1982, p. 903.
3. Page, A. L., Miller, R. H. and Keeney, D. R., in *Methods of Soil Analysis, Part 2, Chemical and Microbiological Properties*, 2nd edn, Agron 9, ASA, SSSA, Madison, WI, 1982.
4. Frankenberger, W. T. and Tabatabai, M. A., *Soil Sci. Soc. Am. J.*, 1980, **44**, 282–287.
5. Omura, H., Sato, F. and Hayano, K., *Soil Sci. Plant Nutr.*, 1983, **29**, 295–303.
6. Frankenberger, W. T. and Dick, W. A., *Soil Sci. Soc. Am. J.*, 1983, **47**, 945–951.
7. Ross, D. J., *Soil Biol. Biochem.*, 1971, **3**, 97–110.
8. Eivazi, F. and Tabatabai, M. A., *Soil Biol. Biochem.*, 1977, **9**, 167–172.
9. Frankenberger, W. T. and Tabatabai, M. A., *Soil Sci. Soc. Am. J.*, 1981, **44**, 282–287.
10. Bremner, J. M. and Mulvaney, R. L., in *Soil Enzymes*, Academic Press, London, 1978, pp. 149–196.
11. Baligar, V. C., Wright, R. J. and Smedley, M. D., *Commun. Soil Sci. Plant Anal.*, 1988, **19**, 367–384.
12. Burns, R. G., *Soil Biol. Biochem.*, 1982, **14**, 423–427.

ACKNOWLEDGEMENT. We are grateful to the Department of Biotechnology, India, for sponsoring the project.

Received 13 December 1997; revised accepted 25 May 1998