even class of enzymes plays a predominant role. In our results, both endo- and exopeptidase activities increased during grain development in the irrigated plants at all pHs examined (Tables 2 and 3). The endopeptidase activities were maximum at alkaline pH and exopeptidases activities maximum at neutral pH. It has been reported in the literature that during monocarpic senescence, RuBisCo is degraded and nitrogen is mobilized to the developing grains[3,4]. The breakdown of RuBisCo has usually been studied with a pH optimum of 4 to 5 (refs 17–19), while a pH optimum of 6.5 has also been reported[20]. The highest rates of degradation have been reported at pH 8.0 for RuBisCo and at pH 6.0 for small subunit (SSU) alone[21]. Differences in the proteolytic properties of SSU may be due to different conformations of the substrate proteins and the true pH of the enzyme can thus be concealed. It has been observed that the highest cleavage of the large subunit of endogenous RuBisCo is at pH 4.0 but maximal degradation of external [^{14}C]-methylated RuBisCo is observed at pH 9.0 (ref. 22). In oat leaf segments, different degrees of degradation and differences in degradation products were demonstrated for both subunits of RuBisCo at pH 4.5, 7.0 and 8.0 and it was suggested that proteases have different pH optima in senescent leaves[23].

Water deficit increased both endo- and exo-proteolytic activities (endopeptidases and exopeptidases) almost ten fold (Tables 2 and 3). In water-stressed plants, the acidic endoprotease activity is maximum till 15 DAA followed by an increase at alkaline pH at 20 DAA, whereas protease activities were more or less same at all pH values tested with an increase at alkaline pH being slightly more than at neutral pH at 20 DAA (Table 3). Water deficit induced an increase in proteolytic activities which could be responsible for the decrease in leaf protein content (Table 1) (ref. 7). Proteolytic activities were assayed using azocasein and water deficit increased the activities at different pH values – 6.0, 8.5, 9.0 and 10 in Vigna and Phaseolus cultivars[6].

In conclusion, it can be said that drought-induced enhancement of monocarpic senescence in flag leaf of wheat during grain development was concomitant with the enhancement of both exopeptidase activity and endopeptidase activity against RuBisCo. Both the endopeptidase activity and exopeptidase activity was enhanced at acidic, neutral and alkaline pH.

1. Dungey, N. O. and Davies, D. D., *J. Exp. Bot.*, 1982, **33**, 12–20.
2. Nooden, L. D., *Physiol. Plant.*, 1984, **62**, 273–284.
3. Feller, U., *Plant Cell Physiol.*, 1979, **20**, 1577–1583.
4. Peoples, M. B. and Dalling, M. J., *Planta*, 1978, **138**, 153–160.
5. Gan, S. and Amasino, R. M., *Plant Physiol.*, 1997, **113**, 313–319.
6. Roy-Macaulay, H., Zuily-Fodil, Y., Kidric, M., Pham Thi, A. T. and Vieira da Silva, J., *Physiol. Plant.*, 1992, **85**, 90–96.
7. Zuily-Fodil, Y., Roy-Macaulay, H. and Pham Thi, A. T., *Physiol. Plant.*, 1990, **A79**, 112.
8. Khanna-Chopra, R., Rao, P. S. S., Maheswari, M., Xiaobing, L. and Shivshankar, K. S., *Ann. Bot.*, 1994, **74**, 503–511.
9. Scholander, P. F., Hammel, H. T., Hemmingsen, E. E. and Bradstreet, E. D., *Proc. Natl. Acad. Sci. USA*, 1964, **52**, 119–125.
10. Barrs, H. D. and Weatherly, P. E., *Aust. J. Biol. Sci.*, 1962, **15**, 413–428.
11. Arnon, D. I., *Plant Physiol.*, 1949, **24**, 1–15.
12. Zivy, M., Thiellement, H., de Vienne, D. and Hofnann, J. P., *Theor. Appl. Genet.*, 1983, **66**, 1–7.
13. Lowry, O. H., Rosebrough, N. J., Farr, A. L. and Randall, R. J., *J. Biol. Chem.*, 1951, **193**, 265–275.
14. Carrasco, P. and Carbonell, J., *Physiol. Plant.*, 1988, **72**, 610–616.
15. Rosen, A., *Arch. Biochem. Biophys.*, 1957, **67**, 54–59.
16. Frith, G. J. T. and Dalling, M. J., in *Senescence in Plants* (ed. Thimann, K. V.), CRC Press, Boca Raton, 1980, pp. 117–130.
17. Peoples, M. B., Pate, J. S. and Atkins, C. A., *J. Exp. Bot.*, 1983, **34**, 563–578.
18. Miller, B. L. and Huffaker, R. C., *Plant Physiol.*, 1981, **68**, 930–936.
19. Whittenbach, V. A., *Plant Physiol.*, 1978, **62**, 604–608.
20. Dalling, M. J., Tang, A. B. and Huffaker, R. C., *Z. Pflanzenphysiol.*, 1983, **111**, 311–318.
21. Otto, S. and Feierabend, J., *J. Plant Physiol.*, 1994, **144**, 26–33.
22. Grima-Pettenati, J., Piquemal, M. and Latche, J. C., *Phs Res.*, 1987, **14**, 179–189.
23. Shurtz-Swirski, R. and Gepstein, S., *Plant Physiol.*, 1985, **78**, 121–125.

Received 16 January 1998; revised accepted 10 September 1998

Identification of blocks in chlorophyll biosynthetic pathway in chlorophyll-deficient mutants of *Pennisetum glaucum* (L.) R. Br.

K. Karunasri, M. V. Sailaja and N. C. Subrahmanyam*

School of Life Sciences, University of Hyderabad, Hyderabad 500 046, India

Blocks in chlorophyll biosynthetic pathway among different chlorophyll-deficient phenotypes of *Pennisetum glaucum* (L.) R. Br. were investigated employing 77 K fluorescence spectroscopy. Green seedlings from yellow/white stripe mutants and an *albino* mutant showed emission maxima at 657, 685 and 733 nm corresponding to protochlorophyllide (pchlide), photosystem II and photosystem I respectively. Yellow seedlings showed peaks at 657 nm and 678 nm corresponding to pchlide and chlorophyllide (chlide) respectively, indicating a block in the reduction of chlide, whereas the white seedlings and *albino* accumulated pchlide, suggesting an impairment in the photoreduction of pchlide. Of the four loci identified[20], vi_3 and vi_4 blocks the conversion of pchlide to chl(ide), whereas the conversion of chlide to chlorophyll is under the control of vi_1 and vi_2 loci.

STEPS in chlorophyll biosynthesis are subject to modulation by both light and cell type[1,2]. Information on

*For correspondence. (e-mail: scnsl@uohyd.ernet.in)

chlorophyll biosynthetic pathway mostly comes from the use of normal, etiolated or dark-grown plants[3–6]. Work in seemingly unrelated different plant systems has provided insights into the regulatory interaction between plastid and nuclear genomes[7].

Mutations in nuclear genes that affect plastid biogenesis are of immense value in dissecting the mechanisms controlling plastid development[7]. Mutations that block chlorophyll biosynthesis have major effects on chloroplast biogenesis and thus are lethal. Several nuclear-induced plastid mutants of higher plants and algae are known to block different steps in the chlorophyll biosynthesis[8–10]. In a Mendelian mutant of *Chlamydomonas reinhardtii*, accumulation of protoporphyrin IX was reported[11]. A recessive Mendelian mutant of *Arabidopsis thaliana* though formed protochlorophyllide (pchlide), was unable to photoreduce the protochlorophyllide to chlorophyll[12]. *Albino* mutant *alb-f17*, and yellow mutant, *xan-j59*, of barley were shown to photoreduce chlorophyllide (chlide), but were unable to phytylate the chlorophyllide to form chlorophyll *a*[13,14]. In *albostrains* of barley, absence of $tRNA^{Glu}$ was found to block chlorophyll synthesis[15]. In pearl millet, studies on chlorophyll-deficient (stripe) mutants provided genetic evidence for plastid–nuclear gene interactions in the biosynthesis of chlorophyll[16,17]. Ultrastructural and molecular characterization of plastids in these mutants[18,19] revealed the absence of: (i) chlorophyll, (ii) thylakoid membrane organization, (iii) plastid-coded large subunit of RuBisCo, and (iv) plastid-specific ribosomes and rRNA (23s and 16s) *vis-á-vis* defective plastid protein synthesizing machinery. So far, four independent loci which control the biosynthesis of chlorophyll have been identified based on the segregation patterns in the progenies of intercrosses of different stripe mutants[20]. While any one locus in homozygous condition blocks the biosynthesis of chlorophyll, chlorophyll synthesis is accomplished by the complementary interaction of dominant genes at these loci[20]. We present here the *in vivo* low temperature (77 K) fluorescence emission spectra of five well-defined chlorophyll-deficient (yellow and white) mutants of pearl millet (Table 1), and their respective green siblings among different genotypes. Comparison of the accumulation of intermediates of chlorophyll biosynthesis among different chlorophyll-deficient (yellow or white) and their normal siblings derived from different genotypes enabled the identification of blocks at specific steps in chlorophyll biosynthetic pathway and their control by different loci.

Genotypes of stripe mutants of *Pennisetum glaucum* (L.) R. Br. (Table 1) and their breeding behaviour were well characterized[20,21]. Following self-pollinations, yellow stripe mutants IP 5009 and IP 9712 gave green, yellow stripe and yellow progeny, while white stripe mutants GWS-14 and VCM-36 gave green, white stripe and white progeny. Seeds from the yellow stripe mutants of IP 5009 and IP 9712, white stripe mutants of VCM 36 and GWS 14 and *albino* mutants were sown in plastic trays under glasshouse conditions (14 h light and 10 h dark). Low temperature (77 K) fluorescence measurements of upper-expanded portion of six-day-old green and non-green (yellow and white) seedlings were recorded, using Hitachi fluorescence spectrophotometer (model 4010). Seedlings were harvested 2–3 h after sunrise, and the fluorescence spectra were recorded immediately using seedlings of approximately similar size. For each set of measurements, five samples were taken from green and yellow siblings of the yellow stripe mutants (IP 5009 and IP 9712), green and white siblings of the white

Table 1. Accessions of *Pennisetum glaucum* (L.) R. Br. used, their phenotype, genotype and source

Accession	Phenotype (genotype)	Source
IP 5009	Green yellow stripe ($vi_1\ vi_1$)*	S. Appa Rao and M. H. Mengesha, Genetic Resources Division, ICRISAT, Patancheru
IP 9712	Green yellow stripe ($vi_2\ vi_2$)	
VCM 36	Green white stripe ($vi_3\ vi_3\ vi_4\ vi_4$)*	
GWS 14	Green white stripe ($vi_3\ vi_3\ vi_4\ vi_4$)*	J. L. Minocha, Genetics Department, Punjab Agricultural University, Ludhiana
Albino	Albino seedlings (*al al*)**	Isolated in earlier study**

*Karunasri and Subrahmanyam[20], **Reddy[21].

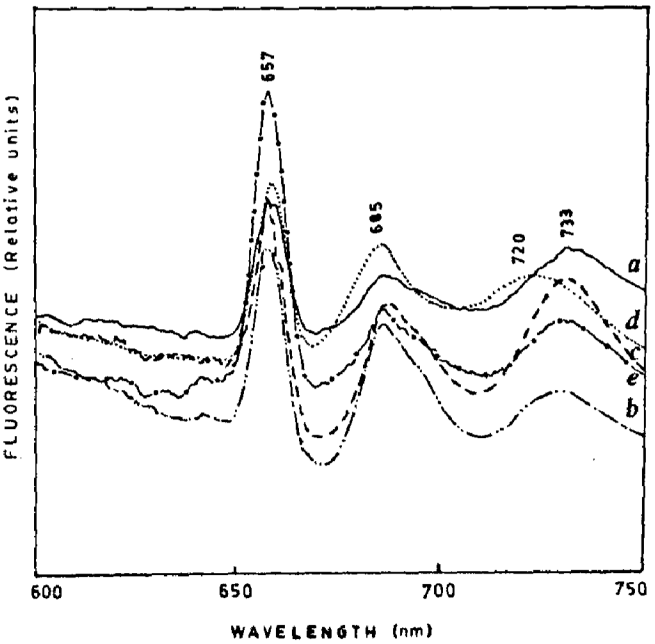

Figure 1. Low temperature (77 K) fluorescence emission spectra of green seedlings from different genotypes (*a*, *b*, *c*, *d*, *e*) at 438 nm excitation wavelength: *a*, IP 5009; *b*, IP 9712; *c*, VCM 36; *d*, GWS 14; *e*, Albino.

Table 2. Accumulation of protochlorophillide/chlorophillide and proposed block(s) in chlorophyll biosynthetic pathway in chlorophyll-deficient phenotypes of *Pennisetum glaucum* (L.) R. Br.

Phenotype	Genotype	Pchlide	Chlide	Block in chlorophyll biosynthesis
Yellow	vi_1 vi_1 Vi_2 Vi_2 Vi_3 Vi_3 Vi_4 Vi_4	+	+	CS Chlide → Chl
Yellow	Vi_1 Vi_1 vi_2 vi_2 Vi_3 Vi_3 Vi_4 Vi_4	+	+	CS Chlide → Chl
White	Vi_1 Vi_1 Vi_2 Vi_2 vi_3 vi_3 vi_4 vi_4	+	–	PCOR Pchlide → Chlide
White	Vi_1 Vi_1 Vi_2 Vi_2 vi_3 vi_3 vi_4 vi_4	+	–	PCOR Pchlide → Chlide
Albino	*al al*	+	–	Pchlide → Chlide

Green siblings had protocholorphyllide and chlorophyll irrespective of their genotype.
Pchlide – protochlorophyllide; Chlide – chlorophyllide; Chl – chlorophyll;
(+) accumulation; (–) absent.
CS – chlorophyll synthetase; PCOR – protochlorophyllide oxidoreductase.

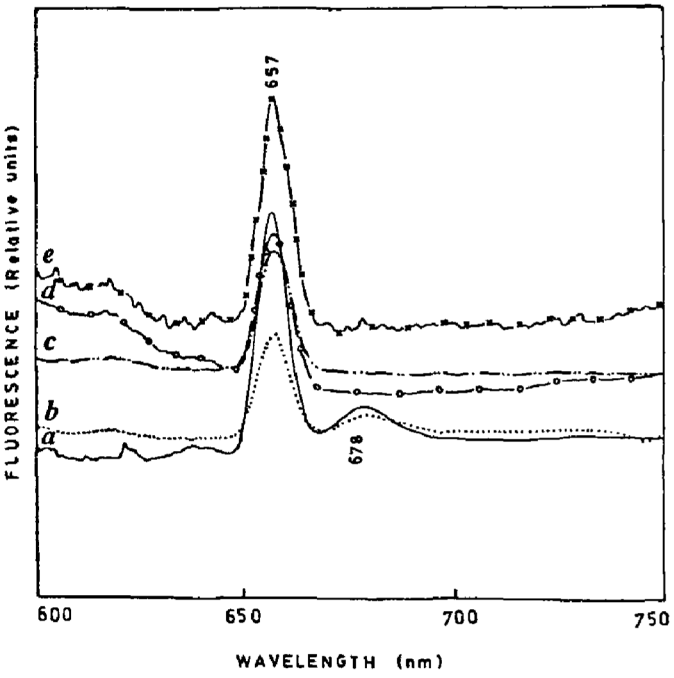

Figure 2. Low temperature (77 K) fluorescence emission spectra from seedlings at 438 nm excitation wavelength. (*a*, IP 5009 yellow seedlings; *b*, IP 9712 yellow seedlings; *c*, VCM 36 white seedlings; *d*, GWS 14 white seedlings; *e*, *al al* white seedlings).

stripe mutants (VCM 36 and GWS 14) and *albino*. Fluorescence emission spectra were recorded at excitation wavelength of 438 nm with 5 nm excitation slit and 4 nm emission slit. The integration time was 0.18 s. The mean spectrum was smoothened before recording. The spectra were not normalized since the aim of the study was qualitative and not quantitative.

In vivo low temperature fluorescence emission spectra of green seedlings from the different genotypes are presented in Figure 1 *a–e*, while the spectra from the corresponding yellow or white siblings from each mutant are presented in Figure 2. The results are summarized in Table 2. The emission spectra were recorded at 438 nm excitation wavelength which preferentially excites pchlide[22].

The fluorescence emission maxima in green seedlings were at 657 nm, 685 nm and 733 nm (Figure 1 *a–e*). Emission spectra revealed the invariable fluorescence emission peak positions in accordance with the elegant demonstration by Böddi *et al.*[23] on the repeatability of the technique and no additional peaks appeared (data not presented). The emission spectrum from the green seedlings, irrespective of their genotype, showed a broader peak in the 720–733 nm range (Figure 1 *a–e*).

Yellow seedlings from the yellow stripe mutants (IP 5009 and IP 9712) exhibited fluorescence emission maxima at 657 nm and 678 nm (Figure 2 *a, b*). The 678 nm peak was found to be sensitive to the light conditions at the time of harvesting. Freshly-harvested leaves showed 678 nm peak, while few hours of storage in dark resulted in the loss of this peak. The second derivatives of the spectra did not alter the number and position(s) of the peaks (data not presented). White seedlings from the white stripe mutants (GWS 14 and VCM 36) and *albino* showed fluorescence emission maxima at 657 nm.

Fluorescence emission spectra of green progeny irrespective of the source (Figure 1 *a–e*) showed emission maxima at 657 nm, 685 nm and 733 nm corresponding to pchlide, chlorophyll from antennae of PS II and PS I respectively when excited at 438 nm (refs 22–24). F_{720} observed from green seedlings of IP 9712 indicates chlorophyll fluorescence from PS I reaction centre complex. Emission band at 678 nm corresponding to chlide[24] was not detectable in the green seedlings, thereby confirming the transient presence of chlide, followed by its rapid conversion to chlorophyll and its concomitant integration into PS II and PS I membrane protein

complexes. However, we cannot completely account for the presence of pchlide in green seedlings which is generally absent in mature leaves because of rapid conversion into chlide/chlorophyll.

A comparison of emission spectra of the white seedlings (Figure 2 *c, d, e*) with those of the green seedlings (Figure 1 *c, d, e*) from three different mutants (GWS 14, VCM 36 and *albino*) revealed a common emission peak at 657 nm which corresponded to pchlide, while the emission peaks at 685 nm and 733 nm which corresponded to the chlorophyll of PS II and PS I respectively, were absent in the white seedlings. Furthermore, peak at 678 nm corresponding to chlide observed in the yellow seedlings (Figure 2 *a, b*) is absent in the white seedlings. This indicates that white seedlings are defective in the conversion of pchlide to chlide, thereby resulting in the accumulation of pchlide. The conversion of pchlide to chlide is catalysed by plastid-specific NADPH–PCOR which accumulates in the plastids of dark-grown plants and eventually becomes the dominating polypeptide of the plastid envelope[25–27]. The white stripe mutants (GWS 14, and VCM 36) are homozygous recessive at the two loci ($vi_3\ vi_3\ vi_4\ vi_4$), while the yellow stripe mutants (IP 5009 and IP 9712) carry dominant alleles at these loci[20]. It is therefore likely that the conversion of pchlide to chlide is controlled by one or both of these loci in pearl millet (Table 2). Further confirmation about the nature of the gene products is awaited.

The presence of an emission peak at 678 nm (Figure 2 *a, b*) in the yellow seedlings signifies the accumulation of chlide[24] and the absence of chlorophyll in PS I and PS II complexes in yellow seedlings indicate a block in the conversion of chlide to chlorophyll. The two yellow stripe mutants are homozygous recessive at vi_1 or vi_2 loci, and their dominant alleles are complementary in the production of chlorophyll[20]. We propose that these loci control the production and/or the activity of chlorophyll synthetase which catalyses the conversion of chlide to chlorophyll (Table 2).

When the leaves are illuminated, pchlide is immediately converted to chlide which is thereafter esterified. In the present study, a major $pchlide_{657}$ form was found in the white seedlings, whereas three spectral forms with fluorescence emission maxima at 632 nm, 645 nm and 657 nm were reported in etioplasts[24]. Emission peak at 678 nm in the yellow seedlings from yellow stripe mutants of pearl millet represents single chlide form (an esterified form – phytyl chlide) unlike different chlide forms in etiolated (dark-grown) seedlings[24].

Henningsen and Thorne[14] have observed that the newly-formed chlide in barley mutants directly emit fluorescence at 678 nm, and the rate of esterification was more rapid than in the wild type. Rüdiger *et al.*[25] have suggested that such esterification of chlide in etioplasts is catalysed by an enzyme or a group of enzymes, termed chlorophyll synthetase. The occurrence of a single emission peak at 678 nm and the absence of chlorophyll (685 nm and 733 nm) peaks in the yellow seedlings of pearl millet in the present study suggests that the esterification of chlide is catalysed by an enzyme different from that catalysing the conversion of chlide to chlorophyll. Since the green and yellow siblings of each accession carry the same genotype, the chlide in the yellow seedlings is likely to be phytyl chlide. Each of these mutants is homozygous recessive at one of the loci ($vi_1\ vi_1$ or $vi_2\ vi_2$) (ref. 20) and block the conversion of phytyl chlide to chlorophyll, but dominant alleles at these loci are complementary in restoring the chlorophyll synthesis. This emphasizes that both these loci control the conversion of chlide to chlorophyll. Further experiments are needed to establish the nature of the gene product(s) and their regulation.

The present study established the genetic control for the reduction of pchlide and/or chlide to chlorophyll. Green and chlorophyll-deficient seedlings from each stripe mutant exhibited differences, implying differences among the plastomes of the two phenotypes. RFLP analysis with plastid-specific probes could be used for establishing the physical basis[26]. Further studies on *in vitro* complementation to determine the nature of interactions are underway.

1. Spano, A. J. and Timko, M. P., *Biochim. Biophys. Acta*, 1991, **1076**, 29–36.
2. Witty, M., Wallace-Cook, A. D. M., Albrecht, H., Spano, A. J., Michel, H., Shabanowitz, J., Hunt, D. F., Timko, M. P. and Smith, A. G., *Plant Physiol.*, 1993, **103**, 139–147.
3. Castelfranco, P. A. and Beale, S. I., *Annu. Rev. Plant Physiol.*, 1983, **34**, 241–278.
4. Rüdiger, W. and Schoch, S., *Chemistry and Biochemistry of Plant Pigments* (ed. Goodwin, T. W.), Academic Press, New York, 1988, pp. 1–59.
5. Senge, M. O., *Photochem. Photobiol.*, 1993, **57**, 189–206.
6. von Wettstein, D., Gough, S. and Kannangara, C. G., *Plant Cell*, 1995, **7**, 1039–1057.
7. Taylor, W. C., *Annu. Rev. Plant Physiol. Plant Mol. Biol.*, 1989, **40**, 211–233.
8. Kirk, J. T. O. and Tilney-Bassett, R. A. E, *The Plastids*, Elsevier, Amsterdam, 1978, pp. 1–960.
9. Thompson, D., Walbot, V. and Coe Jr., E. H., *Am. J. Bot.*, 1983, **70**, 940–950.
10. Taylor, W. C., Barkan, A. and Martienssen, R. A., *Dev. Genet.*, 1987, **8**, 305–320.
11. Sager, R., *Genetics*, 1955, **40**, 476–489.
12. Robbelen, G., *Z. Pflanzenphysiol.*, 1966, **55**, 387–403.
13. von Wettstein, D., Henningsen, K. W., Boynton, J. E., Kannangara, G. C. and Nielsen, O. F., *Autonomy and Biogenesis of Mitochondria and Chloroplasts* (eds Boardman, N. K., Linnane, A. W. and Smillie, R. M.), Elsevier, Amsterdam, 1971, pp. 205–223.
14. Henningsen, K. W. and Thorne, S. W., *Physiol. Plant.*, 1973, **30**, 82–89.
15. Hess, W. R., Schendel, R., Rüdiger, W., Fieder, B. and Börner, T., *Planta*, 1992, **188**, 19–27.
16. Reddy, M. K. and Subrahmanyam, N. C., *Can. J. Bot.*, 1988, **66**, 179–182.

17. Reddy, M. K. and Subrahmanyam, N. C., *Genome*, 1988, **30**, 147–151.
18. Reddy, M. K., Subrahmanyam, N. C., Appa Rao, S. and Mengesha M. H., *Hereditas*, 1988, **109**, 253–260.
19. Sujatha, M. and Subrahmanyam, N. C., *Plant Sci.*, 1991, **73**, 55–64.
20. Karunasri, K. and Subrahmanyam, N. C., *Hereditas*, 1994, **120**, 203–209.
21. Reddy, M. K., Ph D thesis, University of Hyderabad, 1986.
22. Lebedev, N. N., Siffel, P. and Krasnovskii, A. A., *Photosynthetica*, 1985, **19**, 183–187.
23. Böddi, B., Ryberg, M. and Sundquist, C., *J. Photochem. Photobiol. B*, 1992, **12**, 389–401.
24. Hendrich, W. and Bereza, B., *Photosynthetica*, 1993, **28**, 1–16.
25. Rüdiger, W., Benz, J. and Guthoff, C., *Eur. J. Biochem.*, 1980, **109**, 193–200.
26. Karunasri, K., Ph D thesis, University of Hyderabad, 1996.
27. Sundqvist, C. and Dahlin, C., *Physiol. Plant.*, 1997, **100**, 748–759.

ACKNOWLEDGEMENTS. We thank Prof. V. S. Rama Das for providing fluorescence spectrophotometer. Financial assistance in the form of fellowship to K.K. from UGC and Research Associateship to M.V.S. from CSIR, Government of India, is gratefully acknowledged.

Received 12 January 1998; revised accepted 17 September 1998

Foliar levels of rare earth elements and thorium in coconut palm in relation to root (wilt)-disease

P. A. Wahid*, N. V. Kamalam†, S. Vijayalakshmi§, R. Krishna Prabhu§, T. R. Mahalingam§, John T. Eapen‡, H. S. Dang‖, D. D. Jaiswal‖, P. V. Balachandran†, V. K. Venugopal†, P. R. Suresh*, V. K. G. Unnithan† and Jacob Thomas*

*Kerala Agricultural University, College of Agriculture, Nileshwar 671 328, India
†Kerala Agricultural University, College of Horticulture, Vellanikkara 680 654, India
§Materials Chemistry Division, Indira Gandhi Centre for Atomic Research, Kalpakkam 603 102, India
‡Division of Cellular and Molecular Cardiology, Sree Chitra Tirunal Institute for Medical Sciences and Technology, Thiruvanthapuram 695 011, India
‖Health Physics Division, Bhabha Atomic Research Centre, Mumbai 400 085, India

Concentrations of major nutrients and micronutrients, rare earth elements (REEs), Th and nutrient/REE ratios in the leaves of diseased and apparently healthy coconut palms of the root (wilt)-disease-affected tract and healthy palms of the disease-free tract, covering three major soil types of Kerala, namely alluvial (entisols), laterite (ultisols), and sandy (entisols), were examined in relation to the disease. Accumulation of major nutrients, especially K, was generally observed in the leaves of diseased palms. Mg content of leaves of palms growing on laterite soil in disease-affected tract was lower than that of palms in the disease-free tract. The leaf concentrations of La, Ce, Pr, Nd, Sm, Gd and Th did not show significant differences between healthy and diseased palms. The only exception to this trend was Gd whose concentration was less in the diseased and apparently healthy palms growing on laterite soil of the disease-endemic area than that in the healthy palms of the disease-free area. Some of the essential plant nutrients (EPN)/La, and EPN/Ce ratios were significantly different in palms of the disease-affected tract compared to that in palms of the disease-free tract indicating imbalances in the relative concentrations of EPNs and REEs. These results call for more detailed study of the geochemical differences between the disease-affected and disease-free tracts for identification of the soil chemical factors associated with the incidence of the disease.

Coconut root (wilt)-disease has a history of over a century. It was first reported in 1874 in Erattupetta of Meenachil taluk in Kottayam district of Kerala State. It is a debilitating disorder affecting about 0.41 million hectares of coconut gardens in the State, causing an estimated yield loss of 968 million nuts annually[1]. Presently, the disorder is seen mostly in the central and southern districts of Kerala.

Over the past few decades, considerable research has been done on various aspects of the root (wilt)-disease[2]. Etiology of the disease has been examined from several angles namely, bacteriological, fungal, virological, nematological, and nutritional aspects. The work of Solomon *et al.*[3] revealed the presence of mycoplasma-like organisms (MLOs) in the phloem element of root (wilt)-disease affected coconut palms. Valiathan *et al.*[4] reported that presence of high Ce and low Mg levels in leaf may be responsible for the incidence of the disease. They also proposed a common geochemical basis for endomyocardial fibrosis in human beings and root (wilt)-disease of coconut palm. Their study was based on analysis of a few leaf samples collected from healthy and diseased palms. In view of the abundance of monazite deposits (the source of rare earth elements and Th) in Kerala soils, the possibility of absorption of these elements, especially Ce, in large amounts by coconut palms cannot be overlooked. The present study was therefore taken up to examine in detail the levels of rare earth elements (REEs) and Th in coconut palms growing in disease-affected and disease-free tracts of the State.

Based on the prevalence of the disease[1], the districts of Kollam, Pathanamthitta, Idukki, Kottayam, Alappuzha and Ernakulam, and parts of Thrissur district, were considered disease-affected areas. The districts of Thiruvananthapuram, Palakkad, Malappuram, Wayanad, Kozhikkode, Kannur, Kasaragod, and parts of Thrissur were considered disease-free areas. Leaf samples were collected from coconut palms growing in root (wilt)-endemic areas and root (wilt)-free areas, repre-