BRIEF COMMUNICATION

Changes in leaf mineral composition and chloroplast proteins induced by K-deficiency and increased UV-B radiation

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Abstract

Solar UV-B radiation increased to 20 % over ambient level at Madurai was given to cowpea (*Vigna unguiculata* L. cv. Pusa-152) seedlings sufficiently supplied by potassium (0.88 mM K_2SO_4) and K-deficient (0.05 mM K_2SO_4). Leaf mineral composition was significantly changed due to both increased UV-B radiation and K-deficiency imposed independently or jointly for 12 d. A severe reduction in 23 kDa chloroplast protein was seen only in seedlings encountered combined stress.

Additional key words: cadmium, calcium, chromium, copper, iron, lead, magnesium, manganese, nickel, potassium, sodium, zinc.

Large uncertainty exists regarding the realistic impacts of UV-B (280-320 nm) radiation on plants (Fiscus and Booker 1995). For instance, some of the field studies have shown little or no UV-B effects on physiological parameters (*e.g.* Drilias *et al.* 1997, Visser *et al.* 1997) while other ones (*e.g.* Dillenberg *et al.* 1995, Nedunchezhian and Kulandaivelu 1997) have shown large reduction of them. In addition, changes in other abiotic factors like nutrient availability (Musil and Wand 1994, Premkumar and Kulandaivelu 1996), CO_2 concentration (Visser *et al.* 1997), precipitation (Drilias *et al.* 1997) and tropospheric ozone level (Zeuthen *et al.* 1997) are known to alter the effectiveness of UV-B radiation.

In line of our previous study (Premkumar and Kulandaivelu 1996), we examined the interactive influence of increased solar UV-B radiation and K-deficiency on leaf mineral content and chloroplast proteins in field grown cowpea seedlings.

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Cowpea (*Vigna unguiculata* L. cv. Pusa-152) seedlings were grown in pots filled with vermiculite under natural conditions. The vermiculite was moistened daily with standard nutrient solution prepared according to Marschner and Cakmak (1989) with some modifications. For K-deficiency the seedlings were supplied with 0.05 mM K_2SO_4 and P and Cl were supplied as NaH_2PO_4 and NaCl, respectively. The increased solar UV-B irradiance (12.0 kJ m^{-2} d^{-1}) was eqivalent to that anticipated with 20 % ozone depletion at Madurai (10 °N, India). Other conditions and experimental design were the same as described previously (Premkumar and Kulandaivelu 1996). After 12 d, chloroplasts were isolated following an extraction procedure described earlier (Noorudeen and Kulandaivelu 1982) and SDS-PAGE analysis of chloroplast proteins was made according to the method of Laemmli (1970). Leaf mineral composition in the dried samples was determined using Integrated Coupled Spectrophotometer (*Applied Research Lab. 3410,* Switzerland). Samples were prepared by adopting the method of Sivaguru and Paliwal (1993).

In contrast to photosynthetic activities (Premkumar and Kulandaivelu 1996), the most pronounced change under K-deficiency and increased solar UV-B radiation was significant reduction of 43 and 23 kDa proteins in chloroplasts (Fig. 1). In addition,

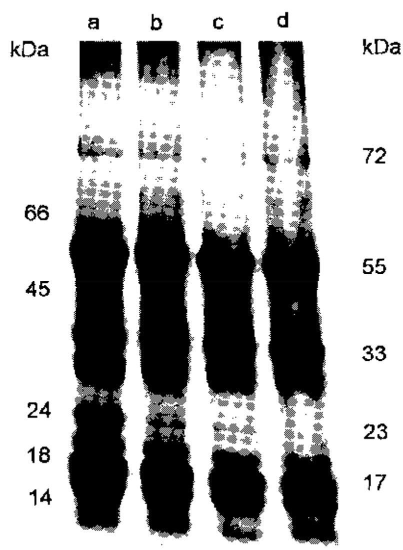

Fig. 1. Coomassie blue stained protein profiles of chloroplast protein isolated from primary leaves of 12-d-old cowpea seedlings grown under sufficient K supply (*lane a*), K-deficiency (*lane b*) enhanced solar UV-B radiation (*lane c*) and combination of both K-deficiency and enhanced solar UV-B radiation (*lane d*). An equal amount (125 μg) of protein sample was loaded in all lanes and *arrows* on left side indicate the position of markers.

among the three extrinsic proteins (33, 24 and 16 kDa) of oxygen evolving complex (Barber *et al.* 1997) only the level of 24 kDa has decreased under all treatments. The latter is responsible for maintaining the structural integrity of oxygen evolving complex. Although 24 kDa was significantly decreased in chloroplasts of K deficient cowpea seedlings, the loss of PS 2 mediated DCPIP photoreduction was only 8 % (Premkumar and Kulandaivelu 1996). Similar to these results, Ono and Inoue (1984) in a specific removal and reconstitution of the extrinsic proteins of PS 2 particles, found that the 24 kDa protein may not always be essential for O_2 evolution.

Both K-deficiency and increased solar UV-B radiation produced significant nutritional disorders in the seedlings whether imposed independently or jointly. K-deficiency reduced the concentrations of Zn, Ca, Mn, Mg, Ni, Cr, K, Pb and Fe in the primary leaves and increased concentration of Na, Cd and Cu (Table 1). UV-B radiation reduced the content of Fe, Zn, Mg, Na, Cu, Cr, Pb, Ni, Ca, Cd and Mn. In contrast intracellular K concentration was higher in seedlings grown under increased solar UV-B radiation. UV-B irradiated leaves of K-deficient cowpea seedlings showed higher concentration of Ca, Mg, Fe, Na, Zn, Ni and Pb than those of seedlings stressed with K-deficiency alone. Similarly the concentrations of Mg, Fe, Ca, Na, Zn, Cd, Ni and Pb in the leaves of seedlings under combined stressed were higher when compared with seedlings received only increased solar UV-B radiation. An additive adverse effect was noticed in the concentrations of K and Cu.

Table 1. Mineral composition of leaves [ng kg^{-1}(f.m.)] of 12-d-old cowpea seedlings grown under K-deficiency and increased UV-B radiation. Means ± SE, n = 3.

	Control	K-deficient	UV-B	K-def. + UV-B
K	369.90 ± 6.15	288.80 ± 3.60	462.40 ± 2.70	240.70 ± 5.20
Ca	1307.40 ± 9.62	602.40 ± 3.96	1172.90 ± 6.41	1272.70 ± 4.80
Na	118.40 ± 1.35	135.00 ± 1.83	101.20 ± 2.06	153.40 ± 1.21
Mg	300.30 ± 5.15	195.40 ± 4.99	256.40 ± 2.29	270.30 ± 1.95
Fe	9.45 ± 0.20	8.70 ± 0.41	3.78 ± 0.19	9.81 ± 0.21
Mn	0.24 ± 0.01	0.15 ± 0.00	0.22 ± 0.04	0.18 ± 0.04
Zn	2.88 ± 0.11	1.20 ± 0.13	1.51 ± 0.11	1.53 ± 0.17
Cd	0.12 ± 0.03	0.15 ± 0.01	0.11 ± 0.01	0.13 ± 0.01
Cr	0.72 ± 0.04	0.55 ± 0.01	0.56 ± 0.02	0.81 ± 0.01
Ni	2.08 ± 0.06	1.42 ± 0.15	1.50 ± 0.18	1.93 ± 0.09
Cu	0.28 ± 0.02	0.29 ± 0.05	0.19 ± 0.02	0.16 ± 0.02
Pb	0.52 ± 0.01	0.47 ± 0.01	0.41 ± 0.01	0.54 ± 0.03

Concentration of several elements was different in levels and shoots (Tables 1, 2). For instance the concentration of Pb was higher in shoots of stressed seedlings as compared with control seedlings. A completely reverse trend was seen in the concentration of Na.

Both stresses significantly lowered the concentration of Zn. The lowered photosynthesis in UV-B irradiated K-deficient seedlings (Premkumar and Kulandaivelu 1996) could be correlated with lower level of Zn which enhanced the formation of reactive oxygen species in chloropasts (Cakmak *et al.* 1989).

Table 2. Mineral composition of shoots [ng kg^{-1}(f.m.)] of 12-d-old cowpea seedlings grown under K-deficiency and increased UV-B radiation. Means ± SE, n = 3.

	Control	K-deficient	UV-B	K-def. + UV-B
K	110.90 ± 3.20	72.60 ± 2.90	99.80 ± 1.40	61.60 ± 1.60
Ca	96.20 ± 1.90	93.60 ± 1.40	84.70 ± 1.40	81.70 ± 1.60
Na	97.30 ± 1.40	102.00 ± 1.70	74.20 ± 1.10	101.20 ± 2.40
Mg	126.60 ± 3.10	162.60 ± 4.80	95.70 ± 2.70	126.60 ± 2.90
Fe	8.80 ± 0.62	13.20 ± 1.00	8.70 ± 0.82	13.40 ± 0.91
Mn	0.06 ± 0.01	0.06 ± 0.01	0.03 ± 0.00	0.03 ± 0.02
Zn	1.12 ± 0.14	2.10 ± 0.43	1.36 ± 0.18	1.84 ± 0.32
Cd	0.09 ± 0.01	0.09 ± 0.01	0.09 ± 0.00	0.10 ± 0.00
Cr	0.47 ± 0.20	0.42 ± 0.19	0.70 ± 0.43	0.50 ± 0.29
Ni	1.09 ± 0.11	1.17 ± 0.13	1.04 ± 0.29	1.21 ± 0.34
Cu	0.06 ± 0.01	0.06 ± 0.00	0.09 ± 0.01	0.13 ± 0.09
Pb	0.32 ± 0.04	0.39 ± 0.02	0.35 ± 0.09	0.50 ± 0.06

UV-B irradiation is also known to stimulate the generation of free radicals (Hideg *et al.* 1997). The inhibition of plant growth in UV-B irradiated K deficient cowpea seedlings (Premkumar and Kulandaivelu 1996) was linearly correlated with reduced level of important macroelements in the leaves, particularly Mg, Mn and Zn.

It is concluded from these results that both K deficiency and increased solar UV-B radiation had direct effects on nutrient status of cowpea plants. Combination of both stresses produced significant nutritional disorders and the persistence nature of such additive effects should be characterized in long-term field study. The experimental design used in this study was very preliminary where only young seedlings were used for impact analysis.

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