STABILITY OF VITAMIN C IN DRUMSTICK LEAF

THE leaf of the drumstick tree (*Moringa oleifera*) has long been known to be a rich source of ascorbic acid,[1,2] providing 900 to 1,100 mgs. per cent. of this vitamin. The edible portion of the pod also contains nearly one per cent. of the vitamin and, being free from interfering substances, was at one time suggested as a suitable standard for titrating indo-phenol indicator solutions.[1] The leaf, however, contains, in addition, 100 to 110 mgms. per cent. of β-carotene[2,3] as determined spectrophotometrically after phase separation and chromatographic adsorption over Brockman's alumina[4] and is, therefore, richer in this respect than either lucerne[5] or rose hip,[6] two of the natural sources of ascorbic acid and carotene which have been successfully processed[7,8] to yield rich concentrates.

In the course of investigations on the processing of drumstick leaf by fractionation and dehydration methods so as to obtain rich concentrates of ascorbic acid and carotene, interesting results were obtained in regard to the relative stability, under ordinary conditions of exposure, of ascorbic acid in aqueous extracts when examined during pre-flowering (October-November) and flowering stages (January-February). Some typical data are reported in Table I where the results for lucerne leaf sampled during the two periods and for an aqueous solution of pure ascorbic acid are included for comparison.

TABLE I

Stability of ascorbic acid in plant sources

Source	Mgm. per cent. ascorbic acid in			
	HPO_3 (5 per cent. Extract)	Aqueous extract at the end of		
		0 hrs.	24 hrs.	72 hrs.
Drumstick leaf–pre-flowering stage	1,143	888	840	666
Drumstick leaf-flowering stage.	985	112	64	..
Drumstick flowers I	1,250	50	..	..
Drumstick flowers II	1,665	66	..	..
Lucerne leaf sampled in October	960	841	328	294
Lucerne leaf sampled in February	920	800	278	237
Ascorbic acid (pure)	25	21	9	..

These results show that a water extract of drumstick leaf is remarkably stable when

sampled during the period of maximum vegetative growth, there being only 25 per cent. loss in ascorbic acid even after three days' storage. On the other hand, a powerful oxidase system is apparently developed in the leaf when the tree is in flower, more than 80 per cent. of the vitamin being lost during extraction alone. This oxidase activity is even more with the flowers where practically the whole of the ascorbic acid is destroyed instantaneously in the aqueous extract.

Whether the destruction of the ascorbic acid during extraction with water and subsequent storage has been due to the specific oxidase elaborated during flowering has not been fully investigated. But, in experiments with aqueous ascorbic acid, addition of a heated water extract of the leaf sampled at flowering stage resulted in a destruction, during 24 hours, of only 44 per cent. of the original vitamin C content as against 74 per cent. with a corresponding quantity of unheated water extract. These results as also a comparison of the relative stabilities of water extracts of the leaf and of aqueous ascorbic acid would suggest that, in addition to the oxidative heat-labile enzyme, plants contain a thermostable protective factor as postulated by Krishnamurthy and Giri.[10] It cannot, however, be stated on the basis of existing knowledge that the activities of the oxidase and of the protective factor could vary independently in different plants and during different seasons.

The foregoing observations on drumstick leaf have been amply confirmed from an examination of a large number of flowering trees. Besides, the non-existence of a powerful oxidase system was demonstrated during the same season in two instances where the trees were not in flowers due either to immaturity or to excessive defoliation.

In contrast with drumstick leaf, the results obtained with lucerne do not show any wide variation in oxidase activity during the two consecutive seasons; but, it must be borne in mind that this plant is grown largely for its fodder and that, therefore, cuttings are taken frequently, as was the case in the present instance, thereby preventing its reproductive phase. Presumably, this modifies the effect of season on the ascorbic acid-ascorbic oxidase system.

That seasonal variations in the concentrations of ascorbic oxidase and of dehydro-ascorbic acid reductase[11] may be considerable is shown by the results of Crook and Morgan[12] for cauliflower. The evidence recorded here would strongly suggest that these enzyme systems concerned in the reversible oxidation-reduction of ascorbic acid are of possible significance in the biosynthesis of this vitamin in plants. Support is lent to this view by the recent findings of Zilva, Kidd and West[13] that there is a decrease in the ratio of dehydro-ascorbic acid to ascorbic acid in the total vitamin C content of the apple as the fruit approaches maturity. No attempt has been made to follow up the dehydro-ascorbic acid and its reductase contents in drumstick leaf during the different developmental stages of the plant, but there is little doubt that such a study will eventually yield information of physiological importance.

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