

## High Nicotinic Acid Content in Two Ethiopian Sorghum Lines

Two recently discovered high-lysine, high-protein, Ethiopian sorghum lines, IS 11758 and IS 11167, have been found to contain high amounts of nicotinic acid also. This finding is of signifi-

cance in improving the niacin status of populations consuming sorghum in the developing countries.

Sorghum or jowar is one of the important cereal crops in the world, and is a dietary staple of poor populations living in the semi-arid tropics of Asia and Africa. The proteins of sorghum, like those of other cereals, are deficient in lysine. In addition, a specific deficiency disease, pellagra, has been shown to be associated with the consumption of sorghum grain in the Deccan Plateau of India (Gopalan and Srikantia, 1960). The presence of excess leucine in sorghum proteins has been implicated in the etiology of pellagra of sorghum eaters (Gopalan, 1969).

One of the approaches to solve the nutritional problems of cereal-consuming populations is the identification and selective propagation of strains of cereals with a genetic potential for increasing the content of specific nutrients, as has been exemplified by the discovery of high-lysine, opaque-2 maize (Mertz et al., 1964). In order to improve the nutritional quality of grain sorghum, a search is being made to identify strains that are high in protein and lysine, but low in leucine. Since pellagra can be ameliorated by increased intakes of nicotinic acid, an additional approach to the control and prevention of this disease among sorghum consuming populations appears to be the identification of sorghum strains with high amounts of nicotinic acid. Nicotinic acid has been found to be in available form in sorghum (Belavady and Gopalan, 1966).

Recently, two new lines of sorghum of Ethiopian origin, IS 11167 and IS 11758, have been reported to contain exceptionally high amounts of lysine at relatively high levels of protein (Singh and Axtell, 1973). Their improved biological value is reported to be controlled by a single gene (Singh and Axtell, 1973). Their leucine content is slightly reduced (Singh and Axtell, 1973) but is not very low, and therefore from this angle these strains cannot be considered as non-pellagrigenic. It was therefore of interest to examine them for their nicotinic acid content. This paper reports the finding of high nicotinic acid content of these two lines of sorghum.

### MATERIALS AND METHODS

Parent seed material of IS 11167 and IS 11758, the two Ethiopian strains of sorghum, from Purdue were grown at I.A.R.I. Research station at Rajendranagar, Hyderabad, during 1973-1974 and the resulting seeds were made available to this Institute for analysis. These, along with check samples of three Indian varieties of sorghum (CSH 1, PJ 160, and G 4), were analyzed in duplicate or replicates for nicotinic acid content by a microbiological procedure (Association of Vitamin Chemists, 1966) using *L. arabinosus* as the test organism. The nicotinic acid levels in powdered seeds were determined subsequent to hydrolysis with 1 N H<sub>2</sub>SO<sub>4</sub> in an autoclave at 15 lb pressure for 30 min. Growth of the organisms was measured turbidimetrically after 18 hr incubation at 37°.

Single analysis for nicotinic acid was done in composite seeds from 17 individual plant heads of one of the lines (IS 11758) grown at Rajendranagar, Hyderabad.

### RESULTS AND DISCUSSION

From the results presented in Table I, it can be seen that the two Ethiopian lines of sorghum have a remarkably high content of nicotinic acid, 10.5-11.5 mg %, as compared to the values for common sorghums. Most of

Table I. Nicotinic Acid Content of New Sorghums

Sample	Sorghum line <sup>a</sup>	Total nicotinic acid, mg %
High lysine, high protein sorghums	IS 11167 (1)	10.5 (mean of duplicate analysis)
	IS 11758 (17)	11.5 (range 9.27-13.45)
Normal sorghums	CSH 1 (1)	2.9 <sup>b</sup> (range 2.8-3.0)
	PJ 160 (1)	3.0 <sup>b</sup> (range 2.85-3.2)
	G 4 (1)	4.9 <sup>b</sup> (range 4.5-5.2)

<sup>a</sup> Numbers in parentheses indicate number of samples analyzed.

<sup>b</sup> Mean of four replicate analyses.

the nicotinic acid values for different sorghum varieties fall between 1.3 and 5.0 mg % (Chitre et al., 1955; Bressani and Rios, 1962; Rao and Ramasastry, 1969; Gopalan et al., 1971; Watt and Merrill, 1963; Leung, 1968). There are, however, occasional reports of findings of high nicotinic acid values in some of the sorghum varieties (Knox et al., 1944; Tanner et al., 1947, 1949; Adrian and Sayerse, 1957; Seljametov and Massino, 1970). The highest value of 12.4 mg % nicotinic acid ever recorded for a sorghum variety was reported by Tanner et al. (1949) in one head of a sorghum cross.

Nicotinic acid content of cereals appears to be less influenced by soil and environment than by heredity (Sebrell and Harris, 1954). Work reported from this Institute (Rao and Ramasastry, 1968) also showed that the range of variation in the nicotinic acid values of two sorghum hybrids, CSH 1 and CSH 2, from four locations was narrow, about 25% for CSH 1 and negligible for CSH 2. Thus, the high values of nicotinic acid found in the two Ethiopian lines in the present study are more likely to be due to their heredity traits rather than to the effect of soil or environment.

The present finding has an added significance for the nutritional superiority of the Ethiopian lines not only from the point of view of their improved protein quality but also for their non-pellagrigenic character. However, further work is necessary to evaluate this aspect.

While these lines may not be agronomically superior and their acceptance by the consumers may be poor because of the dimpled nature and color of the seeds, it may be possible to transfer the high niacin character by crossing to the high yielding varieties of sorghum.

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## Turbidimetric Determination of Ascorbic Acid in Foods

A previously reported qualitative test for ascorbic acid has been found to provide a turbidimetric procedure for the determination of this vitamin in foods. Selenious acid reacts only with ascorbic

acid and stannous ion among some 50 food ingredients tested to date at low pH and room temperature.

The current direct methods for analysis of ascorbic acid in foods (Freed, 1966; Horwitz, 1970) suffer from nonspecificity. The most widely used method, 2,6-dichlorophenol-indophenol titration, gives high results if tannins, reductones, ferrous ion, stannous ion, betanin, and bisulfite are present in the sample. This communication reports a quantitative method for ascorbic acid based on the qualitative test using selenious acid reported by Levine (1936) nearly 40 years ago. The quantitative nature of the reaction between selenious acid (1 mol) and ascorbic acid (2 mol) to form selenium has been reported (Deshmukh and Bapat, 1955). The gravimetric and/or volumetric methods proposed by these workers for the determination of ascorbic acid in vitamin tablets are not useful for analysis of food extracts; many food components will reduce selenious acid to selenium at temperatures of 50–100°.

We have found that stable selenious colloids are formed when food extracts containing ascorbic acid are treated with selenious acid, and the resulting turbidity is proportional to the ascorbic acid content.

### MATERIALS AND METHODS

The ascorbic acid is extracted from foods as described in established published methods (Freed, 1966; Horwitz, 1970); it is not necessary to use acetic acid if ferrous ion is expected in the sample. The clarified, strongly acidic solution is diluted with deionized water to give an ascorbic acid level of 0–4 mg/25–50 ml. The ascorbic acid solution is treated with 5 ml of a solution of 1 g of reagent grade selenium dioxide in 80 ml of water and 20 ml of 37% hydrochloric acid. After 5–15 min the reddish suspension is diluted to 100 ml. The optical density of the suspension is measured in a spectrophotometer at 425 nm using the same volume of diluted food extract with 5 ml of dilute hydrochloric acid, and dilution to 100 ml, in the reference cuvette. A calibration curve is prepared using solutions of ascorbic acid in 3% *m*-phosphoric acid at the 1–4 mg/25–50 ml level with 5 ml of the above described selenious acid reagent and dilution to 100 ml after a 5–15 min colloid development time. See Figure 1 for a typical calibration curve.

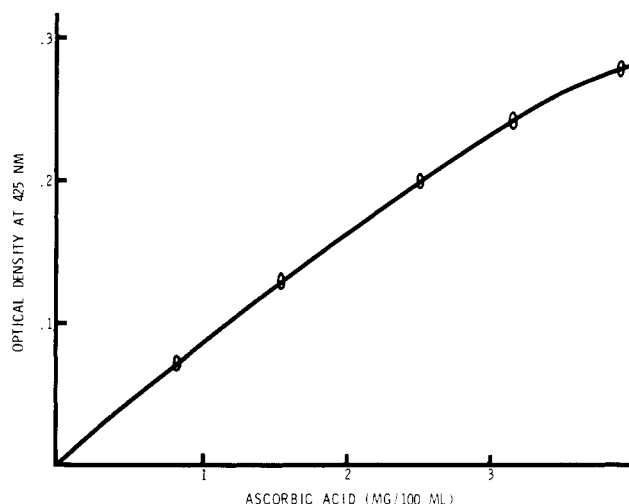


Figure 1. Optical density of selenium colloids prepared from the reaction of ascorbic acid and excess selenious acid.

### RESULTS AND DISCUSSION

The method is highly specific for ascorbic acid when conducted at 20–25°. Levine tested 40 commonly occurring organic compounds and found no selenium formation; we find that betanin, sorbic acid, catechol, and acetoin do not reduce selenious acid to metallic selenium. The levels of ferrous ion and bisulfite ion found in food products do not significantly influence the turbidity caused by the ascorbic acid reduction of selenious acid at room temperature. Stannous ion reacts with selenious acid and, if present, must be corrected for by independent analysis and calibration of its reaction with selenious acid (see Figure 2).

The precision of the turbidimetric determination is illustrated by the data in Table I. The selenium colloid is stable for several hours if no more than 1 mg of selenium/100 ml of solution is formed. Naturally occurring polymeric materials in the commodities tested appear to