

habitants of Leyden, The Netherlands, who were selected at random from the population. Ten per cent were suffering from famine edema. None of the individuals examined showed corneal vessels.

From this, it would appear that while deficiencies of any of three different indispensable amino acids or of protein may result in corneal vascularization in the rat, further investigation is necessary before the significance of these findings with reference to human nutrition becomes clear.

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Dehydroascorbic Acid in Cabbage

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In analyzing school lunches for ascorbic acid content using the method of Roe and Oesterling (3), dehydroascorbic acid was found to be present in appreciable amounts. The average content for 55 plate lunches was 14.8 mg. of reduced ascorbic acid and 5.9 mg. of dehydroascorbic, giving a total of 20.7 mg. (2). Thus, there was 40 per cent more ascorbic acid present than would have been accounted for by the usual indophenol procedure.

These findings prompted us to question whether the reported losses of ascorbic acid in cabbage, when prepared as salad and allowed to stand, could be accounted for, at least partially, as dehydroascorbic acid. The destructive effect of metallic catalysts and time of chopping have been noted by several investigators, but in few studies has the reversibly oxidized form of the vitamin been reported. The method of Roe and Oesterling is sensitive to small amounts of ascorbic acid and is not open to some of the criticisms of the hydrogen sulphide method.

Values for fresh cabbage were obtained from a wedge cut from the intact head and immediately immersed in acid and then ground in a Waring blender. The remainder of the head was cut within five minutes, either with a knife or a hand shredder, and samples taken for analyses at regular intervals. Reduced ascorbic acid was determined by the method of

Loeffler and Ponting (1) and dehydroascorbic acid and total ascorbic acid by the diphenylhydrazine procedure of Roe and Oesterling (3).

Typical results are given in Table 1. When the

TABLE 1
ASCORBIC ACID CONTENT OF CABBAGE AFTER CUTTING AND SHREDDING (MG./100 GRAMS)

Flat Dutch Cabbage	Knife			Shredder		
	Reduced	Dehydro	Total	Reduced	Dehydro	Total
Before cutting	47.3	4.3	50.1	47.6	6.1	53.1
Immediately after cutting	41.5	10.4	49.4	35.5	15.1	52.2
15 minutes after cutting	40.6	10.8	46.3	35.7	17.1	52.5
30 minutes after cutting	40.4	9.1	46.1	34.9	15.7	51.5
60 minutes after cutting	41.1	8.8	49.2	35.3	15.6	52.5
120 minutes after cutting	40.9	8.5	47.2	36.3	14.3	50.6

cabbage was chopped with a knife there was a 13.5-per cent loss of reduced ascorbic acid after standing 120 minutes. However, the values for dehydroascorbic acid indicated that this was not a true loss but that a large portion was changed to the reversibly oxidized form. The total ascorbic acid at the end of 120 minutes was 5.8 per cent less than at the beginning; thus, there was a small destruction of the vitamin during this holding period. Shredding with a hand grater caused a total destruction of 4.7 per cent of the vitamin; with this method of preparation there was an increase in the amount converted to dehydroascorbic acid.

The average values for all heads of cabbage are shown in Table 2. There was a 5-per cent loss of total

TABLE 2
PER CENT OF THE TOTAL ORIGINAL ASCORBIC ACID PRESENT AS REDUCED, DEHYDRO- AND TOTAL ASCORBIC ACID

Treatment	Knife cut (4 heads)			Shredded (5 heads)		
	Reduced	Dehydro	Total	Reduced	Dehydro	Total
Before cutting	99.9	8.4	100.0	92.9	10.3	100.0
After cutting	88.0	10.9	96.3	67.7	30.7	97.0
15 minutes after cutting	86.8	20.0	97.2	66.1	32.5	95.2
30 minutes after cutting	87.7	19.4	93.8	65.5	29.9	96.1
60 minutes after cutting	87.5	17.9	97.2	66.2	29.2	95.2
120 minutes after cutting	89.3	16.6	98.4	67.6	26.8	95.1

ascorbic acid when shredded and held for 120 minutes. The maximum loss occurred in the first 15 minutes and did not increase on standing.

When cabbage was cooked by boiling for 12 minutes and then held on the steam table for two hours, there was marked destruction of the vitamin. Only 25 per cent of the total ascorbic acid present in the freshly cooked cabbage was retained, and 50 per cent of the

total amount present was in the form of dehydroascorbic acid.

The importance of analyzing raw and cooked foods for dehydroascorbic acid is demonstrated. The general assumption has been that this form of the vitamin is equally as well utilized as is reduced ascorbic acid. Since evidence on this point is incomplete, the utilization of dehydroascorbic acid by human subjects is now being investigated in this laboratory.

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Buckwheat as a Source of Rutin

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The recent discovery that the flavonol glucoside, rutin, is effective in the treatment of increased capillary fragility associated with hypertension (2) in man has led to a widespread demand for supplies of the drug by physicians and pharmacologists. Preliminary reports indicate that rutin therapy is successful in controlling conditions due to this type of fragility, such as certain cases of retinal hemorrhage and apoplexy.

In this laboratory, rutin was first isolated from tobacco, and the glucoside supplied for the early clinical experiments was prepared from the flue-cured type of high quality. The low yields from an expensive raw material were reflected in a high cost for the product. It was, therefore, desirable to find a more economical source for the glucoside.

A number of plants were examined for rutin content in the course of this research. Of all the species examined, buckwheat is the most promising source yet discovered.

Rutin was first discovered in buckwheat by E. Schunck (3), who states that he isolated 240 grains of glucoside from 30 pounds of fresh leaves, a yield of 0.11 per cent. Wunderlich (4) obtained "more than 2 per cent" from the dried blossoms of the plant. Brandl and Schärtel (1) reported 1.78 per cent from fresh leaves, 0.71 per cent from fresh flowers, 0.09 per cent from the stems, and 1.02 per cent from the dried whole plant.

During the Summer of 1944, forty-six collections of buckwheat of the Japanese variety were made from

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four scattered localities and examined for rutin.² The results showed that the content of rutin was often considerably in excess of that indicated in the earlier reports. In four cases, yields of more than 6 per cent were recorded, and the over-all average for all samples was definitely higher than previously reported.

TABLE 1
RUTIN CONTENT OF FRESH BUCKWHEAT

Part	Number of samples	Rutin content*		
		Average	Maximum	Minimum
Whole plant†	28	Per cent 2.07	Per cent 8.56	Per cent 0.43
Leaves and blossoms	13	2.50	6.37	1.16

* Moisture-free basis.
† Exclusive of roots.

The leaves contain more rutin than other tissues of the plant. In one case, the leaves and blossoms together contained 6.37 per cent rutin, the leaves 7.92 per cent, and the blossoms 4.15 per cent. The stems contain only small quantities, 0.4 per cent being the largest found. The seeds and flour were free of rutin.

An experiment conducted with one crop and in a single season indicated that the rutin content varies with the age of the plant, being greatest in the early blossoming stage. Collections of the whole plant, minus roots, were made weekly throughout the growing season until the plant had gone to seed. The rutin content was determined for each collection. The results are presented in Table 2.

TABLE 2
VARIATION OF RUTIN CONTENT OF BUCKWHEAT WITH AGE OF PLANTS

Time from planting (days)	Stage of maturity of plants	Moisture, per cent	Rutin,*† per cent	Rutin per plant, mg.	Rutin, per acre, lbs.
12	4-leaf	91.2	0.92	0.87	1.86
19	6-leaf, flower buds forming	89.5	2.50	5.4	10.86
26	1-3 blossom heads in bloom	87.9	2.98	6.9	14.18
33	24"-30" tall, in bloom	91.9	2.47	19.1	39.3
40	36" tall, in bloom	86.9	1.76	24.2	50.25
47	Seeds setting	85.0	1.21	23.5	48.5
54	All seeds set, one-fourth dark	78.6	0.99	27.3	56.3
61	About one-half of seeds dark	80.2	0.62	19.0	39.2
68	All seeds dark	77.8	0.47	19.5	40.2

* Average of duplicate analyses.
† Moisture-free basis.

The data show the rapid increase in rutin content, which reached a maximum in 23 days after emergence

² The method of analysis was essentially that of C. E. Sando and J. U. Lloyd. *J. biol. Chem.*, 1924, **58**, 737.