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 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
		Per cent	Per cent	Per cent
Whole plant†	28	2.07	8.56	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

[1] One of the laboratories of the Bureau of Agricultural and Industrial Chemistry, Agricultural Research Administration, U. S. Department of Agriculture.

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

from the ground (26 days after planting) and thereafter declined gradually as the plant went to seed. A part of the decrease in rutin concentration was due to the relatively faster growth of stem tissues, which contain less rutin than the leaves and blossoms, and later in the season, to atrophying of leaf tissues and replacement of blossoms by seeds. The weight of rutin per plant, however, reached a maximum in 37 to 51 days from emergence and then was 3.5 to 4 times as great as at the time of greatest concentration, the plants having increased 6 to 12 times in weight during the two- to four-week period. On an over-all yield basis, one acre of buckwheat in 26 days from planting would produce 14.2 pounds of rutin, while in 40 days the yield would be 50.25 pounds, or approximately 3.5 times as much.

Storage of the crop presents some difficulties because of the tendency of rutin to disappear as the plant dries. Buckwheat exposed to the sun as in haymaking does not dry quickly or thoroughly and loses rutin rapidly. Experiments conducted to determine the effect of drying conditions upon the rutin content showed that the loss of rutin usually increased as the drying process was prolonged, especially at moderate temperatures. Some typical cases are presented in Table 3.

TABLE 3

LOSS OF RUTIN ON DRYING BUCKWHEAT

Sample number	Manner of drying	Rutin content* of		Loss Per cent
		Undried plant Per cent	Dried plant Per cent	
32	Dried in air 4 days, then at 110° overnight	2.50	0.71	71.6
34	Dried† at 135° for 22 minutes	2.12	1.36	35.8
34	Dried† at 71° for 135 minutes	2.12	.00	100.
34	Dried at 110° for 19 hours	2.12	0.59	72.2
35	Dried† at 105° for 50 minutes	2.98	0.84	71.9
35	Dried at 92–100° for 4 hours	2.98	0.81	72.8
37	Dried at 92–100° for 6 hours	2.47	1.53	38.1
37	Dried at 105° for 40 minutes	2.47	1.52	38.1

* Moisture-free basis.
† Chopped.

When the buckwheat is thoroughly dried the rutin content appears to be stable, no loss being observed in specimens stored for six months or more.

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Thiamine Depletion of Human Subjects on a Diet Rich in Thiamine[1]

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Studies on thiamine balance in human subjects have usually been carried out with restrictions in dietary intake, with recovery or therapeutic test doses administered after deficiency symptoms have been produced, or, more recently, with enzymatic destruction of thiamine in the digestive tract.

In this laboratory it has been found that by supplementation of an adequate basal diet with certain viable fresh bakers' yeasts it is possible to produce within a period of days a strikingly low excretion of urinary thiamine. In a series of nine tests, five college women in a satisfactory nutritional state were fed a basal diet consisting of bread, pineapple, and dairy products which provided 1.6 mg. of thiamine per day. In order to further insure satisfactory body stores 2 mg. of thiamine hydrochloride were added daily to the subjects' self-selected diets for a 3-day period 24 hours prior to ingestion of the basal diet. The diet sequence was one of three periods: a yeast-supplement period during which either 15 or 150 grams of the live, fresh bakers' yeast containing 7 µg. of thiamine per gram, of a yeast type previously shown not to yield its thiamine for absorption (*4*), was ingested in addition to the basal diet; the yeast period was immediately preceded and followed by a yeast-free basal period.

The interference of the yeast with the availability of dietary thiamine from other sources and the resulting decrease in thiamine output are recorded in Table 1. The trend was obvious during the 15-gram dosage as well as on 150 grams of yeast. The sharpest decline occurred in all cases within the first 4 days after which fluctuations at the low level of approximately 50 µg. of thiamine excretion persisted in the case of the larger dosage, a decline twice as great as that produced by the smaller amount. In like manner, when the larger amount was continued for 10 instead of 6 days, the recovery of higher urinary concentrations upon resumption of the yeast-free diet was measurably slow, indicating a possible depletion of body stores. That the interference was a function of the viability of the yeast was indicated not only by the greater decrease in urinary thiamine excretion on the larger dosage, but also by the decided increases in the

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