Ascorbic Acid Stability in Certain Aqueous and Fruit Juice Vehicles Subjected to Elevated Temperature

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The effect of heating ascorbic acid in fruit juices for 6 hours in a boiling water bath in the pH range 2.0 to 8.0 was investigated. Pure lime juice, lime juice plus sorbitol (1:1), and lime juice plus glycerol (1:1) were found to be good vehicles. At pH 3.0 the retention was fairly good. Under controlled conditions there was no significant loss even in distilled water. Storage of vitamin C in distilled water at room temperature showed a rapid loss, except at pH 3.0 where the retention was 44 per cent.

IT IS WELL KNOWN that liquid vitamin mixtures gradually deteriorate in their biological potency during storage and during the process of heating. The loss in vitamin potency is dependent on many factors. Various workers have investigated the problem of stability, yet much remains to be done in this direction. Vehicles (1–3) greatly influence the stability of vitamins; an ideal vehicle which may be acceptable to both children and adults has been the subject of long search. With this idea in mind, the present study was undertaken employing fruit juices as vehicles. Fruit juices are preferred because they are natural and palatable. The various formulations used are listed in Table I. Ascorbic acid was subjected to heat in a boiling water bath in the formulations as given in Table I for 6 hours. On the basis of the retention values of ascorbic acid, the suitable vehicles offering maximum protection were employed for further storage studies.

EXPERIMENTAL

Vitamin.—Ascorbic acid (Hoffmann-La Roche and Co., Ltd., Basle, Switzerland) was used.

Vehicles.—Fruit juices, syrup (67% w/v), sorbitol (E. Merck 70% w/v), and glycerol (BDH 75% v/v) were employed. Some of these bases contained 0.15% potassium metabisulfite (E. Merck, henceforth referred to as KMS) and 0.15% cysteine hydrochloride (E. Merck) Table I.

Procedure.—A 5.0 mg./ml. quantity of ascorbic acid was added to the base and the solution was filled in Pyrex test tubes. The pH of all these vehicles was adjusted between 2.0 to 8.0. After recording the initial readings, the test tubes were plugged with cotton and kept in boiling water for 6 hours. Ascorbic acid was estimated by titrating it against standard 2,6-dichlorophenol-indophenol (BDH) solution.

The percentage retention of ascorbic acid after 6 hours of boiling is given in Table I. Another study was made to see the effect of storage on ascorbic acid in glass distilled water at room temperature for a period of 30 days, the results are presented in Table II.

Received January 3, 1963, from the Government Fruit Preservation and Canning Institute, Lucknow, India.
Accepted for publication March 27, 1963.
The authors express their thanks to Dr. D. N. Srivastava, Director, and to Dr. W. B. Daté, Deputy Director, Fruit Utilization, U. P., Ranikhet, for their keen interest in this work.

RESULTS AND DISCUSSION

It is clear from Table II that in distilled water vitamin C was lost to the extent of 70% in all the pH ranges between 2.0 to 8.0, except at pH 3.0 where there was 44% retention. Between pH range 4.0 to 6.0, the destruction was to the same extent, indicating that pH values 4.0, 5.0, and 6.0 behave alike as far as retention of vitamin C in distilled water is concerned.

A slight drop in pH occurred in the sample originally adjusted to pH 8.0. No changes were observed in other sets having a pH 2.0 to 7.0.

At pH 2.0, 7.0, and 8.0 the retention is practically the same. Table II suggests that vitamin C can be stabilized at pH 3.0 in distilled water with the inclusion of some antioxidants and metal binders. Experiments are in progress to stabilize the solution of ascorbic acid at pH 3.0 in distilled water.

At the end of the experimental period (30 days), browning occured at all the pH values under trial. The intensity of the browning was more with corresponding higher pH values. At pH 3.0, browning was comparatively less. It is assumed that the oxidation products of vitamin C were responsible for this browning and probably also because of polymerization of ascorbic acid to furfural. Further work on this aspect is in progress.

It is evident from Table I that even in distilled water ascorbic acid was retained to the extent of 91% at all the pH values, except at pH 4.0 and 7.0, where the retention was 78.0% and 61%, respectively. Hence, it can be concluded that heating does not completely destroy ascorbic acid under controlled conditions of pH in distilled water. This may be explained on the presumption that chances of metallic ion contamination in distilled water are less.

KMS has a stabilizing tendency in the retention of ascorbic acid at all pH values except at pH 2.0. Loss of ascorbic acid in distilled water at pH 4.0 and 7.0 after 6 hours of heating can be counteracted by the addition of KMS at the 0.15% level. Cysteine hydrochloride also has a stabilizing effect on vitamin C at all pH values except pH 2.0.

Lime juice without preservative can retain ascorbic acid to the extent of 100% at pH values 3.0, 6.0, 7.0, and 8.0. At pH values 2.0, 4.0, and 5.0 a slight destruction occurred. Preservative seems to have no influence on the stability of ascorbic acid in lime juice at pH 6.0, 7.0, and 8.0; but at pH

TABLE I.—PERCENTAGE RETENTION OF ASCORBIC ACID AFTER 6 HOURS IN BOILING WATER

Soln. No.	Vehicle	pH 2	3	4	5	6	7	8
1	Water without preservative	97.3	94.4	78.3	91.8	91.8	61.2	91.8
2	Water with preservative	94.4	97.3	91.8	100.0	100.0	100.0	97.2
3	Water with cysteine hydrochloride	94.4	102.8	89.3	94.0	102.8	97.3	102.8
4	Lime[a] juice with preservative	98.0	93.3	89.4	98.5	100.0	100.0	100.0
5	Lime juice without preservative	89.9	100.0	84.9	93.1	100.0	100.0	100.0
6	Lime juice (50%) plus 50% sorbitol	100.0	100.0	91.6	91.6	93.0	100.0	100.0
7	Lime juice (50%) plus 50% glycerol	92.1	100.0	100.0	96.2	97.2	100.0	97.2
8	Lime juice (50%) plus 50% syrup	94.4	88.4	94.4	88.3	97.1	97.2	97.3
9	Sorbitol, 70%	92.8	100.0	91.7	96.5	100.0	100.0	100.0
10	Syrup, 67%	50.0	92.9	77.3	95.4	97.4	98.5	100.0
11	Glycerol, 75%	74.3	80.3	95.8	83.3	90.0	91.9	94.4
12	Mango[e] pulp (50%) plus 50% water with preservative	67.7	98.8	89.3	80.5	82.6	74.3	81.7
13	Mango pulp (50%) plus 50% water without preservative	62.5	74.3	64.4	90.1	94.7	97.3	91.6
14	Pure apple juice[b]	100.0	98.5	88.9	93.2	100.0	93.9	78.0
15	Pure mosambi juice[c]	89.9	89.5	89.5	86.5	92.9	95.2	97.4
16	Pure pineapple juice[d]	95.3	100.6	80.1	95.2	100.0	95.3	95.3

[a] Lime—*Citrus aurantifolia* Swingle. [b] Apple—*Prunus malus*. [c] Mosambi—A variety of *Citrus sinensis osbeck* (sweet orange), grown in Western India. [d] Pineapple—*Ananas sativa* schutt. [e] Mango—*Mangifera indica* L.

TABLE II.—PERCENTAGE RETENTION OF ASCORBIC ACID DURING STORAGE AT ROOM TEMPERATURE FOR 30 DAYS

Soln. No.	Vehicle	pH 2	3	4	5	6	7	8
1	Distilled water	9.2	44.5	27.3	25.3	25.9	9.8	13.3

3.0 preservative seems to have a destructive effect on the stability of ascorbic acid in lime juice. At pH 2.0 and 5.0, preservative seems to have a stabilizing action. Experiments are in progress to confirm this postulation.

In 50% lime juice plus 50% sorbitol at pH values 2.0, 3.0, 7.0, and 8.0 there was no loss of ascorbic acid. At pH values 4.0, 5.0, and 6.0 the loss was to the extent of 10%.

In 50% lime juice plus 50% glycerol vitamin C, retention was 100% at pH values 3.0, 4.0, and 7.0, while at other pH values the loss was not more than 8.0%.

The combination of 50% lime juice plus 50% syrup was a poor vehicle compared to vehicles No. 6 and 7 (see Table I).

In 70% sorbitol there was no loss of vitamin C after 6 hours of boiling, at pH 3.0, 6.0, 7.0, and 8.0. A slight loss was observed at pH 2.0, 4.0, and 5.0.

Sixty-seven per cent syrup and 75% glycerol do not prove to be good vehicles compared to 70% sorbitol. At pH 2.0 in 67% syrup, there was loss of 50% of vitamin C after 6 hours of boiling. Sorbitol proved to be a good vehicle. This finding to some extent is in conformity with Gerber, *et al.* (4), who showed that in 70% sorbitol and in presence of vitamin B_{12} and ferrous gluconate the degradation of vitamin C can be checked.

Vitamin C deteriorated in 50% mango pulp plus 50% water vehicle at all pH values, both in the presence and absence of KMS. It is interesting to note that 0.15% KMS has a stabilizing effect on the retention of vitamin C after 6 hours of boiling at pH 2.0, 3.0, and 4.0; but the same substance has a deleterious effect at higher pH values, *viz.* 5.0, 6.0, 7.0, and 8.0. This finding is being subjected to repeated trials.

There was no significant loss of vitamin C in both apple juice and pure pineapple juice after 6 hours of boiling at all pH values except pH 4.0. At pH 8.0 there was 20% loss of ascorbic acid in apple juice but none in pineapple juice. Mosambi juice is not a suitable vehicle compared to apple and pineapple juice.

For retention of ascorbic acid, pH 3.0, 6, and 7 were found suitable. pH 4.0 appeared to be on the border line for retention values of ascorbic acid after 6 hours of boiling in all the vehicles tried except vehicles No. 7, 8, 11, and 15. Fifty per cent lime juice plus 50% sorbitol was found to be most suitable for ascorbic acid retention, both from the palatability and economic points of views, although 50% lime juice plus 50% glycerol was better. Apparently there is not much difference between bases No. 6 and 7.

After 6 hours of boiling no change in pH values was observed in the samples previously adjusted to pH value 2.0 and 7.0, but a slight drop in the pH was observed in the test tube originally adjusted to pH 8.0. The result of this investigation indicates that the loss sustained by ascorbic acid during heating for 6 hours in different media is not considerable.

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