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A COMPARISON OF METAPHOSPHORIC AND OXALIC ACIDS AS EXTRACTANTS SOLUTIONS FOR THE DETERMINATION OF VITAMIN C IN SELECTED VEGETABLES

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ABSTRACT

Two extractant solutions, metaphosphoric and oxalic acids were tested in the determination of vitamin C in both, raw and cooked, broccoli, green pepper, potato, cauliflower, cabbage, and green collard. The results did not show any significant difference (P)0.05) between the two solutions. Metaphosphoric acid could be advantageously replaced by oxalic acid.

INTRODUCTION

Vegetables are important contributors to the diet as a source of vitamins and minerals. They are responsible for most of the vitamin C consumed by the population(1).

The vitamin C activity in foods is associated with L-ascorbic acid content. Its determination is usually based on the reducing properties of L-ascorbic acid or on its reaction with hydrazin; chromatographic methods have also been used. A common step in all the procedures is the vitamin extraction from the samples(2,3,4,5,6,7). Several solutions have been suggested for this purpose. Among them, the most cited in the literature are the solutions of metaphosphoric, oxalic and trichloroacetic acids, alone or in combinations(3,4,5,6). The most frequently employed are the solutions of metaphosphoric and oxalic acids, the former being the preferred one in most of the published papers reviewed. It is claimed that it is an effective inhibitor of the ascorbic acid oxidase activity and an efficient protein

precipitant, useful in the extract clarification. Oxalic acid shows these properties but it is not as efficient as metaphosphoric acid as a protein precipitant agent Considering that the products analysed in the present investigation have a low protein content this limitation can be disregarded.

Oxalic acid has been satisfactorily used for the determination of vitamin C when model systems are studied(9,10). A few papers have described the use of oxalic acid solution as an extractant agent, nevertheless, neither justification nor comparison with metaphosphoric acid solution has been made(11,12,13,14,15,16,17,18,19,20, 21).

The objective of this study was to compare the efficiency of oxalic acid solution as an extractant agent in comparison with the metaphosphoric acid solution. The determinations were carried out in raw and cooked vegetables.

MATERIAL AND METHODS

The vegetables selected for the study were: broccoli, green pepper, potato, cauliflower, cabbage, and green collard. In all cases fresh, unwilted vegetables of the best quality obtainable were purchased from local markets in amounts that made possible an uniform sample for each procedure tested. All the vegetables were cleaned, washed and rinsed with distilled water.

The samples taken for direct analysis and those used for cooking were as homogeneous as possible, being selected morphologically similar pieces. Samples were cooked in 100 ml of distilled water for 20 minutes. Immediately after cutting and weighing the samples were dip into boiling water contained in a pyrex beaker.

After cooking the vegetables were drained and the

volumes of liquid were measured and recorded. This was followed by homogenization of 25 grams sample with 50 grams of extractant solution (oxalic acid 2% and metaphosphoric acid 3%) in a waring blender for approximately 2 minutes. An aliquot of 10 grams was taken and diluted to 50 ml with the extractant solution in a volumetric flask. From this an aliquot of convenient volume was taken for the titration with 2,6-dichlorophenolindophenol 0.01%. A modification of the titration method(2) was carried out by using a more diluted 2,6-dichlorophenolindophenol solution (0.01%). The titration end point was detected visually. All the analyses were conducted in triplicated samples.

RESULTS AND DISCUSSION

The only visualized difference between the extracts obtained with the two acids (metaphosphoric and oxalic) was the color and a turbid appearance of the oxalic acid extracts. These observations are summarized in Table 1.

TABLE 1. Appearance of the extracts obtained with metaphosphoric and oxalic acids.

VEGETABLE	Metaphosphoric acid	Oxalic acid
Broccoli	light green, transparent	dark green, turbid
Green pepper	light green, transparent	dark green, turbid
Potato	whitish, transparent	yellowish, slightly turbid
Cauliflower	whitish, transparent	yellowish, slightly turbid
Cabbage	yellowish, transparent	yellow, turbid
Green collard'	light yellow, transparent	greenish, turbid

To avoid any difficulty in visualizing the titration end point the extracts were diluted (1:30). At this dilution the color and turbidity did not interfere with the titration. Moreover, it was found that the sample dilution to 50 ml made the extract filtration quite easy. A

10 grams aliquot was used, but when the vitamin C content is low an amount of 20-30 grams is sufficient.

After cooking it was easier to filtrate the oxalic acid extract than the metaphosphoric one, especially for potato, broccoli and cauliflower.

The end point of the 2,6-dichlorophenolindophenol titration is visualized when the solution color changes from colorless to pink. Due to the selected procedure where the samples are already diluted we choose a more diluted solution of 2,6-dichlorophenolindophenol (0.01%) than the recommended by AOAC (2)(0.025%).

As far as stability is concerned, metaphosphoric acid is considered as not so stable, making necessary the preparation of a new solution every week(3). Oxalic acid solution, stable for several weeks has to be kept in the dark to avoid peroxide formation which will destroy the vitamin. It was observed that if stored for short periods none of the solutions (metaphosphoric acid 3% and oxalic acid 2%) changed when kept in the refrigerator. After a ten days period the metaphosphoric acid solution showed some change, while the oxalic acid solution remained unchanged.

According to the literature the acids concentration varies in the range of 0.5-2%. Oxalic acid solution in a concentration of 2% was tested and it was stable and efficient as an extractant agent. No vitamin losses were detected even if the solution is kept at room temperature for several hours, which represents a convenience, especially when many samples have to be analyzed. We also found that it was not necessary to keep an innerte atmosphere during the extraction procedure(21).

As shown in Table 2, when oxalic acid was used higher values were obtained than those for the metaphosphoric acid, but, sometimes, this tendency was reversed. Statistical analysis of variance showed that no

significant differences (P)0.05) (22) were found.

Our data shows that is perfectly viable the use of oxalic acid, which can replace efficiently the metaphosphoric acid, being less expensive and showing a greater stability It can be suggested as an extractant solution of choice, at least to determine vitamin C in the vegetables selected for this study. We think that would be interesting to test the applicability of oxalic acid solution to determine vitamin C in other vegetables.

TABLE 2. Vitamin C content of raw and cooked vegetables extracted with metaphosphoric and oxalic acid solutions (mg vitamin C/100 grams sample).*

VEGETABLE		Oxalic acid $\bar{X}$	Oxalic acid s	Metaphosphoric acid $\bar{X}$	Metaphosphoric acid s
Broccoli	Raw	105.91	(2.32)	112.50	(1.74)
	Cooked	47.09	(1.74)	50.21	(1.93)
Green pepper	Raw	120.60	(1.87)	111.92	(1.84)
	Cooked	83.57	(1.24)	85.08	(1.32)
Potato	Raw	13.81	(0.89)	10.22	(0.73)
	Cooked	8.32	(0.45)	7.56	(0.36)
Cauliflower	Raw	112.67	(3.43)	113.50	(1.60)
	Cooked	46.65	(1.53)	54.07	(1.95)
Cabbage	Raw	65.51	(1.51)	78.20	(1.83)
	Cooked	20.71	(1.38)	25.80	(1.80)
Green collard	Raw	126.56	(2.39)	136.41	(2.53)
	Cooked	40.13	(1.63)	31.22	(1.36)

* No significant difference (P)0.05) between the means were detected.

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