ARTICLE

Ascorbic acid losses in vegetables associated with cook-chill food preparation

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Objective. To assess ascorbic acid (AA) losses in four vegetables (broccoli, peas, cauliflower and cabbage) at each production stage in a cook-chill food service system.

Setting. A long-stay psychiatric hospital in Cape Town.

Design. Cross-sectional analytical study. On two repeat occasions, three samples of each vegetable were taken at the following stages: (*i*) delivery (day 1); (*ii*) after preparation (day 2); (*iii*) after cooking (day 5); (*iv*) after blastchilling (day 5); and (*v*) in the holdroom and after regeneration over a 4-day period (days 6 - 9). AA content of each sample was assessed in duplicate using the 2,6-dichloroindophenol method of titration.

Statistical analyses. Differences between the AA concentration of each vegetable at baseline (delivery) and at subsequent stages in food production were assessed using analysis of variance (ANOVA) methods.

Results. The most dramatic AA losses occurred during the cooking stage (mean loss 58%, standard deviation (SD) 19.5%, range 33 - 81%), with broccoli showing the greatest mean loss of 81% (SD 2.9%). During storage in the holdroom from day 6 to day 9, the average daily loss was 4.3% (SD 4.2%). Further average losses of 6.1% (SD 3.6%) were incurred after regeneration on each day. Mean AA losses on day 6 of production and after regeneration (the earliest time a patient would receive the vegetable) were 87% (SD 5.6%). On day 9 after regeneration (the latest time it could be received by a patient) mean losses were 89% (SD 10.5%).

Conclusion. Use of a cook-chill food service system needs to be carefully controlled in order to minimise AA losses. In particular, prolonged cooking times should be avoided and vegetables should be immersed into rapidly boiling rather than cold water. A reduction of the number of days in the chilled storage holdroom would not substantially reduce overall AA losses.

Serving food to patients has been a primary component of hospital care for centuries. Traditionally, food was prepared in the hospital kitchen and held in heated units until it was served to the patient. However, the cook-chill technology of the 1960s allowed for a distinct break between production and service.[1] In the cook-chill catering system food is fully cooked, then fast-chilled and stored at controlled low-temperature conditions above freezing point until subsequent thorough reheating close to the time and place of consumption.[2]

Vitamin C is the most labile vitamin during chilled storage of food at 0 - 3°C after cooking. It has been suggested that vitamin C losses of between 6% and 10% per day can be expected during cold storage at 2°C.[3] This is particularly relevant in the cook-chill food service system, where the holding period may be as long as 5 days.

Vitamin C loss during the cooking process is due partly to oxidative destruction and partly to leaching of the

vitamin into the water used for cooking. The amount of vitamin destroyed may be quite small compared with the amount lost in leaching. Bender[4] demonstrated that 10% of the vitamin C content of cabbage was lost by heat-associated destruction, while 80% was leached into the cooking water.

In addition to vitamin C losses associated with cooking, substantial losses occur during reheating of chilled food; however, the losses are dependent on the time taken to reheat, as well as the portion size of the foodstuff. Reheating a bulk portion (2 kg) of food results in an average vitamin C loss of 23%, compared with losses of 10 - 15% if individually portioned food is reheated for the same length of time.[5]

Long-stay psychiatric patients have been shown to be a group at high risk of suboptimal vitamin C status. Almost two decades ago, it was shown that the plasma vitamin C status of 855 patients in a psychiatric hospital was lower than that of age-matched controls.[6] Elderly subjects with impaired cognitive function are particularly vulnerable. A study of new geriatric admissions to a psychiatric hospital in Leeds, UK, identified 25 - 54% of patients as being at high risk of vitamin C deficiency, based on their blood levels.[7] Frank vitamin C deficiency is rarely seen in younger adults; however, up to 50% of older adults may have a marginal or even deficient vitamin C status.[8] This is especially true of those living in long-stay institutions who rely on the hospital food to provide all of their nutrient requirements.[9,10] A local study conducted among older residents in a Cape Town old-age home found that none of the subjects had a vitamin C intake exceeding two-thirds of the RDA and that over one-third of subjects had low plasma levels (< 0.4 mg/dl) of the vitamin.[11]

Vitamin C losses in vegetables prepared using a cook-chill system have been studied previously, but not in South African institutions. This information would give rise to hypotheses on effective ways to minimise such losses. A study was therefore undertaken in a long-stay psychiatric hospital in Cape Town, which represents a patient population at high risk of a suboptimal vitamin C status, to assess ascorbic acid (AA) losses in four commonly served vegetables of moderate to high vitamin C content (peas, cauliflower, cabbage and broccoli) at each production stage of the cook-chill food service system.

Methods

Sample selection

Four vegetables (cauliflower, cabbage, broccoli and peas) were sampled during the following stages of production in a long-stay (i.e. average minimum patient stay of 6 months) psychiatric hospital in Cape Town utilising a cook-chill catering system.

1. Upon delivery of the product (day 1) from the market.
2. After preparation of fresh vegetables (day 2) (except for peas, which were frozen and therefore needed no preparation).
3. Within 30 minutes after cooking (day 5). Days 3 and 4 fell over a weekend, during which time no food production takes place; the hospital kitchen operates on a 5-day production cycle.
4. Directly after blast chilling to a core temperature of 0 - 3°C (day 5).
5. In the holdroom for up to 4 days (days 6 - 9), and after regeneration on each of these days. The temperature of the vegetable samples was measured with a probe thermometer after regeneration on days 6 - 9, using the standard protocol of 25 minutes of reheating.

Three samples were selected at each of the food production stages for each vegetable, one sample each from the left side, the right side and the centre of the gastronome. Each vegetable sample was immediately macerated and weighed in a measuring cylinder, with 8% acetic acid added to a volume of 100 ml to stabilise the AA content. Filtrate was collected for each sample after the solution was passed through filter paper. Two aliquots were pipetted from each filtrate for duplicate titrations and AA analyses. The mean of the duplicate AA analyses was calculated for each of the three sampled vegetable portions, and an overall mean AA content was calculated for each vegetable. The entire sampling and analytical process was repeated, by the same single, trained observer, for all vegetables except broccoli on a separate occasion (repeat 2) in order to increase reliability of the results (fresh broccoli was not seasonally available at the time of the repeat tests).

Ascorbic acid determination

The 2,6-dichloroindophenol titration method was utilised to detect the AA content of the vegetable samples as it is simple and rapid to perform and its specificity has been shown to be comparable to other oxido-reduction methods.[12]

A standard AA solution was made up daily to standardise the 2,6-dichloroindophenol dye. The following equation was used to assess the mg of AA that corresponded to the ml of dye obtained from an average titre:

$$\text{ml dye (F)} = \frac{\text{mg AA x 10 x 5 x 1 000}}{\text{100 x 250}}$$

The amount of AA equivalent to 1 ml of dye was calculated. The factor (F) was then used in the following formula to estimate the mg of AA per 100 g of a vegetable sample (mg/100 g):

$$\text{mg AA/100 g} = \frac{\text{titre x F x 100 x 100}}{\text{mass x aliquot}}$$

A single observer performed all of the titrations on each occasion.

Comparison of analyses with food composition tables and menu analysis

The average AA content for the portion size of vegetables served at the long-stay institution was compared with reference vitamin C values published in the fruit and vegetable supplement of the South African Food Composition Tables.[13] The average daily vitamin C content of the menus provided was calculated for 1 week of the 4-week menu cycle in operation at the institution at which the study took place. The 'Foodfinder' nutritional analysis software package was used for this purpose, the database of which is the South African Food Composition Tables.[14]

Statistical analyses

Six titrations were performed for each vegetable at each stage of food production on both repeat 1 and repeat 2 occasions. The mean AA content was calculated for the six samples at each repeat. At each stage of food preparation the coefficient of variation (CV = SD/mean x 100) was calculated for the AA concentration of the six samples taken for each vegetable (duplicate titrations for each of the three samples taken on each repeat occasion), as well as the mean CV of the two repeat analyses. The CV allows for assessment of the variability in repeated analytical methods, and provides information on the variation in AA content of food samples taken from different parts of the gastronome.

Analysis of variance (ANOVA) tests were performed for the average AA content of each of the four vegetables sampled to assess whether differences in AA content were present between the early (i.e. delivery to blast chilling) and late (i.e. blast chilling day 5 - day 9 of regeneration) stages of food production. For each of the four vegetables, a 'group effect' was investigated to assess AA differences in the samples according to whether they were reheated or not; a 'time effect' analysis to assess differences between the stages of sampling of the vegetables (i.e. delivery to day 9 of reheating); and a 'repeat effect' to assess differences between the mean AA values obtained in the repeated analyses. A significant interaction between the early stages ('time') effect and the 'repeat' effect indicates that the magnitude of AA losses associated with each of the production stages differs according to the occasion on which the experiment was performed, and suggests poor reliability of the analytical methodology. A significant interaction between the reheating ('group') effect and the late stages ('time') effect indicates that the magnitude of AA losses associated with reheating differ according to the time the vegetable is stored in the holdroom. Data were analysed using the Statistica computer program.

Results

The mean AA content of the vegetables (mg AA per 100 g) during the stages of food production from delivery to blast chilling are shown in Table I. The mean AA content of the four vegetables, according to different stages in the cook-chill process, is shown in Fig. 1.

The CV for each of the stages of food production on each of the repeat sampling days (i.e. CV of 6 titrations) were substantial for each of the vegetables: 5.3 - 45.8% for cauliflower, 9.8 - 80.7% for broccoli, 7 - 53.2% for cabbage, and 5.1 - 24.3% for peas. The mean CV (for the two repeat occasions) of the AA analyses for all four vegetables at delivery was 16.7% (SD 9.3%, range 9.8 - 29%); after preparation 15.7% (SD 7%, range 6.6 - 21%); after cooking 18.2% (SD 4.8%, range 17 - 21.4%); and after blast chilling 19.2% (SD 12.4%, range 14.6 - 37.6%). During the late stages of food preparation, the mean CV for the AA analyses of the four vegetables kept in the holdroom for various time periods ranged from 12.8% to 25.3%. Mean

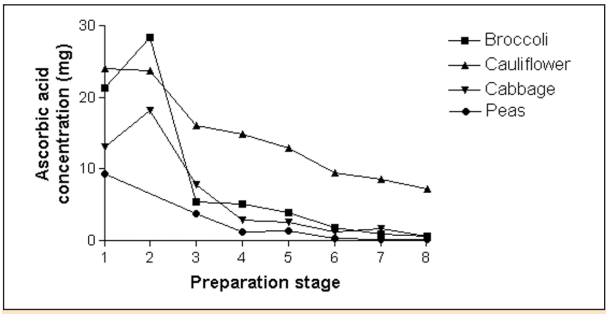


Fig. 1. Ascorbic acid content of four vegetables, at different stages in the cook-chill process.

Table I. Ascorbic acid (AA) content (mg/100 g) and AA retention (%) for broccoli, cauliflower, cabbage and peas at different stages of production*

Production stage (day of sampling)	AA content (% AA retention)			
	Broccoli	Cauliflower	Cabbage	Peas
Delivery (day 1)	21.3	24.0 (100)	12.9	9.2 (100)
Preparation (day 2)	28.43 (100)†	23.8 (99.2)	18.1 (100)	-
Cooked (day 5)	5.47 (19.2)	16.0 (66.7)	7.8 (42.8)	4.8 (40.3)
Blast chilled (day 5)	5.06 (17.8)	14.2 (59.2)	2.9 (15.7)	1.3 (14.1)
Holdroom day 1 (day 6)	3.97 (14.0)	12.9 (53.8)	2.6 (14.4)	1.4 (15.6)
Reheating day 1 (day 6)	1.84 (6.5)	9.5 (39.6)	1.22 (6.7)	0.3 (3.0)

* Mean of repeat 1 and repeat 2 values, except for broccoli for which only one sample was undertaken due to seasonal unavailability.
† The highest concentration of ascorbic acid was selected as the starting concentration (i.e. 100% retention).

CV for the AA analyses of the four vegetables after reheating on days 6 - 9 ranged from 20.1% to 37.3%.

Losses during cooking and blast chilling

Mean AA losses for the repeat analyses between delivery/preparation and the end of the cooking process were as follows (the highest AA concentration at either of these stages was taken as the starting concentration): broccoli 80.8%, cauliflower 33.2%, cabbage 57.2%, and peas 60% (losses between delivery/preparation and cooking were significant ($p < 0.05$) for all vegetables sampled). The interaction of the 'repeat' and the 'time' (i.e. early stage) effects are not significant for the cabbage and cauliflower samples, which indicates that the methodology used to assess AA concentrations during these stages was reliable. However, in the case of peas, a significant ($p < 0.001$) interaction effect was found, which is probably explained by the much higher AA content of peas at delivery at repeat 2 compared with repeat 1 (11.8 v. 6.71 mg/100 g, respectively).

A significant difference ($p < 0.001$) was found between the AA content after cooking and after blast chilling for cabbage and peas, and a similar trend was found for cauliflower ($p = 0.053$); however, no loss in AA associated with blast chilling was found for broccoli. Additional AA losses (as a percentage of starting AA concentration) incurred at this stage of production were 27% for cabbage and peas, and 7.5% for cauliflower.

Losses during chilled storage

A steady decline in AA levels was detected during chilled storage in the holdroom (days 6 - 9), with a mean AA loss of 4.5% (SD 2.2%) per day (range 2.3 - 7.6% per day) (Table II, Fig. 2).

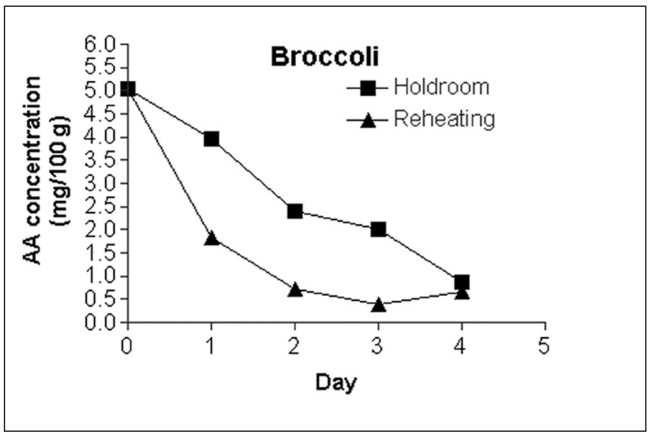
Losses during regeneration

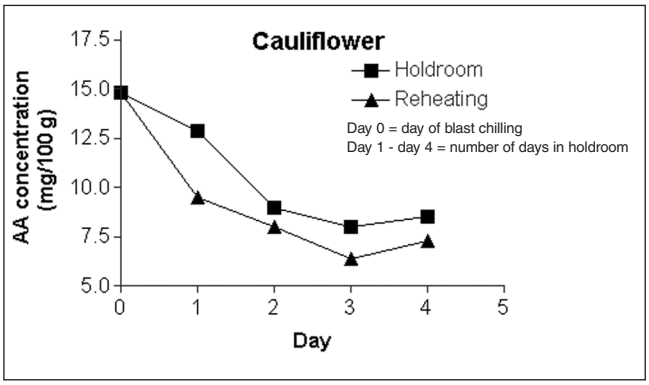
The regeneration process had a significant negative effect on the AA content of the vegetable samples. The vegetable samples in this study were reheated in bulk portions (approximately 2 kg per tray) for 25 minutes at 180°C, as is standard practice in the ward kitchens. The temperature of the samples after 25 minutes of reheating was below the recommended temperature of 70°C (mean 65°C and 60°C for repeat 1 and repeat 2, respectively),

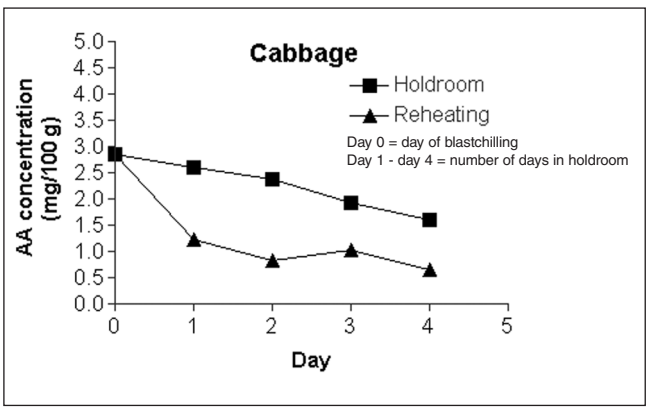
Table II. Mean ascorbic acid retention (%) in cooked vegetables during chilled storage in the holdroom, before regeneration*

Vegetable	AA retention (%)				Mean loss (%) (SD) per day in holdroom
	Day 6	Day 7	Day 8	Day 9	
Broccoli	14.0	8.4	7.1	3.1	3.6 (2.2)
Cauliflower	53.8	37.5	33.4	35.9	7.6 (7.5)
Cabbage	14.4	13.1	10.7	8.8	2.3 (1.2)
Peas	15.6	7.7	4.1	2.4	4.4 (3.2)

* Mean of repeat 1 and repeat 2 values, except for broccoli for which only 1 sample was undertaken.







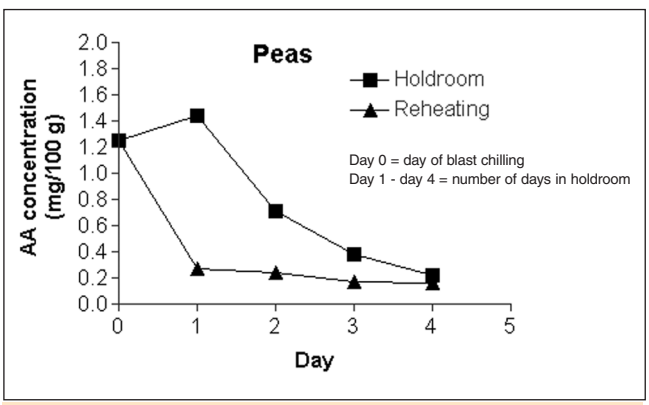


Fig. 2. Ascorbic acid losses in vegetables, according to time in the holdroom after blast chilling and after subsequent reheating.

except on day 7 of the first repeat when a temperature of 75°C was reached.

The average AA loss for the vegetable samples from the holdroom to after reheating was 6.1% (SD 3.6%), with a range of 5.0 - 7.6%. On day 6, after regeneration, the total mean loss of AA from baseline was 86.0% (SD 17.2%) (Table III), with the peas showing a particularly high mean loss of 97% (SD 1.3%). On day 9, after regeneration, the total mean AA loss was 90.4% (SD 13.9%), with the peas again showing the highest mean loss of 98.2% (SD 0.007%).

The mean AA losses associated with the late stages of the cook-chill food preparation system (i.e. according to number of days in the holdroom and after subsequent regeneration on each of these days) are demonstrated in Fig. 2. Two-way ANOVA found a significant ($p < 0.05$) interaction for the 'group' (i.e. reheating) and 'time' (i.e. late stage) effect for all four vegetables, which indicates that AA declines at a more rapid rate during reheating than in the holdroom. For the three vegetables for which repeat analyses were performed (cabbage, cauliflower and peas), the interaction between 'repeat', 'group' (i.e. reheating), and 'time' (i.e. late stage) effect was significant, which suggests either inherent variation associated with the days of sampling or metho-dological inconsistencies between days of analysis.

Vitamin C content of menus

The vitamin C content of serving sizes of each of the four vegetables, as indicated on the patients' menu at the long-stay institution, was compared using reference values derived from the South African Food Composition Tables[13,14] and the AA concentration measured in the present study (Table IV). Even if the vegetables were served to patients immediately after cooking, large discrepancies between the two values are evident. The mean measured AA content of the serving sizes of the four vegetables sampled, after cooking, was 7.0 mg, compared with the calculated value of 36 mg (i.e. 19.5%). The average daily provision of AA, as estimated from the 1-week menu analysis, is 81.4 mg per day (range: 31 mg - 113 mg/day). However, based on the assumption that only 19.5% of the reference value would be present in food served to the patients, the daily provision would equate to a mean of 16 mg, which is less than one-fifth of the dietary reference intake of 90 mg.[15]

Table III. Total ascorbic acid (AA) losses (%) after reheating following 1 - 4 days in the holdroom (i.e. days 6 - 9)*

	Day 6	Day 7	Day 8	Day 9	% Difference between days 9 and 6
Broccoli	93.5	97.5	98.6	97.6	4.1
Cabbage	93.3	95.4	94.3	96.4	3.1
Cauliflower	60.4	66.7	73.5	69.6	9.2
Peas	97.0	97.4	98.2	98.2	1.2
All vegetables	86.0	89.3	91.2	90.4	4.4

* Mean of repeat 1 and repeat 2 analyses, except for broccoli for which only one sample was undertaken.

Table IV. Vitamin C content of vegetable serving sizes as provided to patients in a long-stay institution — comparison between food composition reference values and measured ascorbic acid (AA) concentration

Vegetable	Serving size on menu (g)	Vitamin C content (mg) (Food Composition Tables)	Measured ascorbic acid content (mg)*	
			After cooking	After reheating on day 6
Broccoli	80	67	4.4	1.5
Cabbage	100	24	7.8	1.2
Cauliflower	80	44	12.8	7.6
Peas	80	8.4	3.0	0.2
Mean AA content (mg) per serving of vegetable		35.9	7.0	2.6

* Mean of repeat 1 and repeat 2 analyses, except for broccoli for which only one sample was undertaken.

Discussion

The most significant AA losses occurred between the preparation and cooking stage in all of the vegetables sampled. The method of cooking used for the four vegetables was boiling, which resulted in an average AA loss of 58%, with the greatest AA loss occurring in the broccoli samples (81%).

AA losses during cooking can be reduced by at least half if vegetables are only one-quarter covered by water, rather than being completely immersed.[16] Oxidative destruction of the AA content of foodstuffs further contributes to losses of the vitamin. Oxygen is present in cold water, but not in rapidly boiling water.[17] The vegetables in this study were cooked according to the usual practice in the hospital kitchen, which includes placing the vegetables in large pots of cold water while the water is being brought to the boil and cooking the vegetables for an unspecified period of time.

Surprisingly, the AA content of the broccoli and cabbage samples increased from delivery to the preparation stage. At the delivery stage the samples selected included outer leaves, in which AA destruction may have been excessive, and which were removed during the preparation stage.

In the early stages of production (i.e. delivery to blast chilling), no differences in AA losses were found between the first and second repeat analyses for cabbage and cauliflower samples (data not shown). This suggests that the methodology employed was reliable. In the case of peas, the significant 'repeat' effect could be due to a higher starting AA concentration at delivery in the first analysis, which resulted in a different magnitude of AA loss, despite a similar trend between the two analyses. The frozen peas were kept at room temperature before analysis for all the titrations. It is possible that the amount of AA that may have leached into the surrounding defrosting water may have differed between analyses. Alternatively, natural variation in AA content between vegetable crops may have explained the 'repeat' effect. A 5-fold range in vitamin C has been demonstrated in tomatoes, a 20-fold difference in different varieties of mangoes and a 35-fold range in varieties of grape, while the loss of AA in peas has been shown to vary according to variety and degree of

maturation.[4] For all vegetables, the CV for the six AA titrations performed for each vegetable on each of the two sampling days was substantial for some of the stages of food production. This highlights the importance of taking food samples from different parts of the gastronome, in which the AA content may vary, as well as performing duplicate analyses in order to improve the validity of the results.

A steady decline in AA levels was detected during the chilled storage (days 6 - 9), with a mean loss per day in the holdroom of 4.2% (SD 4.2%) and a range of 2.3 - 7.6% per day, depending on the type of vegetable. These values are slightly lower than those reported in the literature. Bognar *et al*.[3] reported a daily vitamin C loss of 6% during chilled storage at 2°C, while Williams[5] reported a 7.5% vitamin C loss per day at storage temperatures below 3°C.

The regeneration process had a significantly greater negative effect on the AA content of the vegetable samples than the losses associated with cold storage. The vegetable samples in this study were reheated in bulk portions of approximately 2 kg per tray. The average loss of AA during reheating was 6.1%, with a range of 5 - 7.6%, depending on the type of vegetable. It has previously been reported[3,4] that losses of AA during reheating of chilled meals are considerable, and that the losses depend on the time taken to reheat, as well as the size of the sample. Bognar *et al*.[3] demonstrated that AA losses decreased from 36% to 17% in mashed potatoes and from 33% to 11% in boiled potatoes when reheating sample size was reduced from 2 kg to 200 g. In both bulk (i.e. at least 2 kg per tray) and pre-portioned foods, AA losses of at least 14% have been reported during reheating from 3°C to 70°C.[5]

The lower AA loss during reheating in the present study may be due to the fact that the samples only reached an average temperature of 65°C and 60°C during the first and second repeat experiments respectively, despite adherence to the recommended practice of reheating food for 25 minutes at 180°C in the ward kitchens.

On day 6 after regeneration, in other words the earliest day on which patients would receive the chilled food that had been reheated and therefore the maximum amount of AA provided by the vegetables, the mean AA loss was 86%, with peas showing a particularly high mean loss of 97%. Day 9 is the final day on which patients would receive chilled food that has been reheated. The mean loss of AA after regeneration on day 9 was 90%, with the peas again showing the highest mean loss of 98%. These reported values for the extent of AA loss after the reheating of the vegetables are unacceptably high. It has been recommended that if the holding temperature requirements are achieved (0 - 3°C), then the maximum life of cooked products in the cook-chill system is 5 days, including the day of cooking and the day of consumption.[2] However, these recommendations are mainly concerned with the microbiological safety of the food rather than the nutritional quality.

We have demonstrated that the large discrepancies between indirect methods of assessment of the vitamin C content of cyclical menus used in the long-stay institution under study and the direct measurement of the vitamin content in food as served to patients, would grossly overestimate the provision of this micronutrient. Even if the vegetables were served to patients immediately after cooking, the food table-derived values would overestimate vitamin C content by between a half and two-thirds, and by 90% in the case of broccoli.

The vitamin C content of foods consists of two biologically active forms of the vitamin — dehydroascorbic acid (DHAA) and ascorbic acid (AA). DHAA is the oxidised form of the vitamin, while AA is the reduced form of the vitamin. In the South African Food Composition Tables,[13,14] the values for vitamin C refer to the total vitamin C (DHAA + AA). The DHAA content of vegetables comprises approximately 10 - 20% of the total vitamin C content, but this value can vary from vegetable to vegetable.[18] DHAA is biologically active as it can be easily reduced back to AA in the body; however, once DHAA is irreversibly oxidised to 2,3-diketogulonic acid it has no antiscorbutic activity. The relatively small content of DHAA in foodstuffs would not explain the large differences in value obtained in this study (in which only AA was determined) and published reference values.

In order to obtain a composite value for both forms of vitamin C, analysis by high performance liquid chromatography is required. However, the method is costly and the objective of the present study was to assess the magnitude of AA losses associated with each of the food production stages, rather than to assess absolute concentrations of vitamin C.

It is also feasible that the vegetables sampled in this study may be poor-quality raw material due to crop variation or prolonged storage before purchase, which would have resulted in extensive oxidation to DHAA, even before preparation, and therefore the much lower

than expected AA content. State-funded institutions, such as the one investigated in the current study, are required to adhere to a strict food budget, which results in the purchase of the cheapest fresh produce available. Post-harvest losses of AA were not addressed in this study. However, in a study in which a maximum period of commercial transport, distribution and retail shelf-life was simulated, vitamin C losses in freshly harvested broccoli were minimal, despite major losses in phenolic compounds and other health-promoting compounds.[19]

The present study did not attempt to measure the vitamin C status of patients in the long-stay psychiatric institution. Future studies that investigate whether changes in the cooking methods employed within a cook-chill system will result in a higher retention of AA in vegetables should include an assessment of the change (or not) in the vitamin C status of the patient population consuming the food.

Conclusions and recommendations

The cooking stage of food production showed the most significant AA losses. In institutions that use a cook-chill catering system, it is recommended that particular attention be paid to the cooking stage in terms of reduced cooking time, minimal quantities of cooking water used for boiling or the employment of alternative cooking methods, and the addition of vegetables to water only once it has reached boiling point. Use of the vegetable cooking water in soups and gravies would also substantially increase the amount of AA that patients receive. For some vegetables such as frozen peas, the cooking stage could be eliminated until regeneration at ward level. Reducing the holding period in chilled storage from a maximum of 5 days to 2 days (with the day of cooking and blast chilling taken as day 1) would reduce AA losses by only an average of 3.2%. Additional ways to increase the vitamin C intake of long-stay patients may include the provision of fresh fruit or juice and salads.

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