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ASCORBIC ACID (VITAMIN C) IN DEFENCE FEEDING

by

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INTRODUCTION

Although it was known in the 18th Century that citrus fruits and fresh vegetables could prevent and cure scurvy, it was not until the early 20th Century that the factor involved, vitamin C, was purified. Vitamin C, also known as ascorbic acid, was obtained in the pure crystalline form by King and Waugh (1932).

CHEMICAL STRUCTURE

Hirst, Percival, Smith & Haworth (1933) defined the structure of ascorbic acid. Only two forms possess biological activity, L — ascorbic acid and its first oxidation product dehydroascorbic acid (Johnson, 1979).

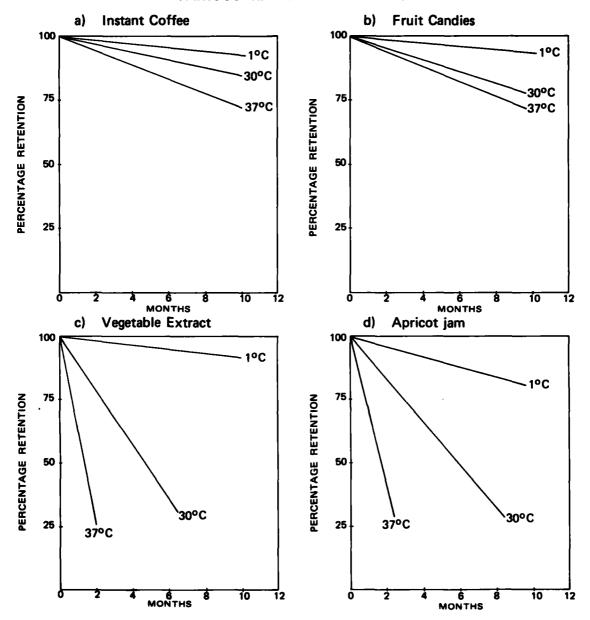
STABILITY

Seasonal variation, storage (particularly at elevated temperature), processing techniques and cooking practices all affect the ascorbic acid content of food as consumed. As a general rule, canned fruit and vegetables contain little ascorbic acid. Retention of 10 - 45% has been reported for canned peaches (Souty, 1972). Ascorbic acid will degrade in the can if oxygen is present and will also degrade faster in a lacquered can than in a plain one. The tin plate of a plain can acts as a reducing agent and rapidly removes residual oxygen, protecting vitamin C from oxidation. Immediately the can is opened, destruction of ascorbic acid by atmospheric oxygen commences and proceeds rapidly.

Ascorbic acid is the most active reducing agent known to occur naturally in living tissues (Davidson and Passmore, 1971). It is therefore very easily oxidised. If the oxidation is continued beyond the stage of dehydroascorbic acid, it is irreversible and the vitamin loses its activity. Many plants contain vitamin C oxidase, an enzyme that catalyses the oxidation of the vitamin in the presence of air. Ascorbic acid is normally protected from contact with the enzyme in the intact plant. Processing methods involving cutting or pounding, release vitamin C oxidase which then quickly destroys the ascorbic acid irreversibly.

Ascorbic acid retention during storage of Australian ration packs is monitored at the Armed Forces Food Science Establishment (AFFSE). The graphs on page 2 illustrate typical vitamin C losses on storage at various temperatures for one year. The items were fortified with Vitamin C initially and stored at 1°C, 30°C and 37°C.

RETENTION OF ASCORBIC ACID DURING STORAGE OF VARIOUS RATION PACK ITEMS



From the graphs, fruit candies and coffee retain significant levels of ascorbic acid even after twelve months storage at 37°C. Fruit candies lose little vitamin C because the solid candy excludes oxygen. Coffee is protected from contact with oxygen by the hermetic packaging. All foods of high moisture level, e.g. vegetable extract and apricot jam show a rapid drop in ascorbic acid level at elevated temperatures.

The Australian Army Service Requirement (A.S.R. 69.1 1980) specifies a shelf life of twelve months above the Tropic of Capricorn, for operational ration packs. Our results suggest that this is a realistic shelf life with respect to ascorbic acid. At present levels of fortification with ascorbic acid, a significant extension of shelf life would not be recommended if the vitamin C levels of operational ration packs are to remain adequate.

Cooking also leads to loss of vitamin C. The quantity depends on duration of cooking, amount of water added, pH of the cooking medium and holding time before serving.

Food which is brought rapidly to cooking temperature, cooked quickly and consumed immediately after cooking loses the least vitamin C (Johnson & Peterson, 1974).

Table I shows average cooking losses of vitamin C for vegetables (from Johnson & Peterson, 1974).

Table I LOSSES OF VITAMIN C FROM VEGETABLES DUE TO COOKING

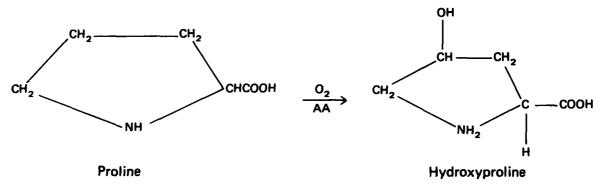
Vegetable	Average % loss of Vitamin C
Leafy, green & yellow	60
Potato	60
Tomato	15
Other Vegetables	60

METABOLIC FUNCTIONS AND FACTORS AFFECTING REQUIREMENTS

1. Wound Healing

In spite of its many known metabolic functions, ascorbic acid is known to act as coenzyme only to certain hydroxylating enzymes (Johnson, 1979).

Wound healing involves the production of connective tissue or collagen. Ascorbic acid is involved in the hydroxylation of proline (an amino acid) to form hydroxyproline, an essential component of collagen (Peterkofsky & Undenfriend, 1965).



Proline is incorporated in the polypeptide chain and then oxidised to hydroxyproline in the presence of ascorbic acid. The hydroxyl group so added can then form hydrogen bonds with the C = O groups of the peptide chains. This is a cross linking effect which strengthens the collagen. In the absence of ascorbic acid, this crosslinking is absent, because proline does not possess the hydroxyl group. Therefore, vitamin C deficiency will lead to defective collagen with little or no tensile strength and wounds will not heal properly (West and Todd, 1963).

Ascorbic acid is known to be involved in other aspects of protein metabolism, e.g. the hydroxylation of lysine to hydroxylysine (Jennings 1970). It may be that a high protein intake necessitates a high ascorbic acid intake, just as increased carbohydrate intake requires additional thiamin (Davidson and Passmore, 1971).

Metabolic Defence Mechanisms

Ascorbic acid appears to have a role in defence against toxic substances. Animal experiments have shown increased resistance to pesticides after supplementation of feed with ascorbic acid (Wagstaffe & Street, 1971). Fuller, Henson and Shannon (1971) demonstrated increased resistance to bacterial endotoxins (toxins produced by some pathogenic

bacteria, e.g. salmonellae and shigellae). Spivey, Fox, Fry, Harland, Schertel & Weeks (1971) showed increased resistance to cadmium poisoning after supplementation with ascorbic acid.

Ascorbic acid may be involved with protection against cancer (Yamafugi, Nakamura, Omura, Soeda & Gyotoku, 1971). Pelletier (1970) discussed the possible association of vitamin C and lung cancer: smokers use more ascorbic acid than non-smokers, suggesting that an interaction occurs between ascorbic acid and inhaled smoke. Also, women have higher leucocyte ascorbic acid levels than men (Loh and Wilson, 1970) and women contract lung cancer at about 20% of the male rate (Rohan and Christie, 1980). It has been hypothesised (Johnson, 1979) that carcinogenic free radicals in cigarette smoke may be inactivated by high levels of ascorbic acid.

3. Ascorbic acid and the Common Cold

The use of large doses of ascorbic acid, about one gram per day, as a prophylactic against colds has been proposed by Pauling (1970).

However, little confirmation has come from other sources. Badcock (1973) reviewed twenty seven trials of ascorbic acid as a prophylactic or therapeutic agent towards colds. The results were ambiguous, some trials reporting a positive effect, some showing no effect, and one trial demonstrating a negative effect. Badcock concluded that the problem is unresolved.

Johnson (1979) concluded:

"It seems fair to say that ascorbic acid deficiency lowers resistance to disease generally. Once ascorbic acid requirements have been met, little evidence exists to show that megadosage of ascorbic acid is nutritionally beneficial during normal health.

On the other hand, acute illness is often associated with sudden changes in plasma and leucocyte levels of ascarbic acid. Hence megadosage may be effective as a therapeutic measure during stress."

4. Hot/Cold Acclimatization

Studies in Japan (Nakamura, 1967), the Arctic (Popov and Osetrov, 1963) and the Antarctic (van der Merwe, 1962) indicated greater need for Vitamin C in cold climates. The general conclusion was that acclimatization to cold becomes greater as the blood ascorbic acid level rises until the latter reaches approximately 1.0 mg/ml. This is regarded as the "critical level for maintaining sufficient resistance to cold" (Nakamura, 1967). Such a concentration of blood ascorbic acid is achieved by an intake of 60-100 mg/day (Ralli, 1939). However, similar results were not obtained by Ryer (1954 a, 1954 b).

The evidence concerning vitamin C requirements in hot climates is also conflicting; Henschel (1944) found no benefit in ascorbic acid supplementation. Hinson (1968) reported significant improvement in heat acclimatization after ascorbic acid supplementation.

5. Physical Exertion

Again the results are ambiguous. Harper (1943) demonstrated an increase in vital capacity, breath-holding time and endurance time with supplementary vitamins A, D, and C. However, Keys and Henschel (1942), Johnson (1945), and Fox (1940) found no evidence of improved performance as a result of supplementation of normal diets with ascorbic acid.

TOXICITY

In view of Pauling's suggestion (Pauling, 1970) that megadosage, i.e. intakes of Ig or more, of ascorbic acid may prevent or assist in curing colds, some comments on the possible toxicity of ascorbic acid are relevant.

Ascorbic acid has little acute toxicity (Davidson & Passmore, 1971). However, all materials are toxic if taken in sufficient quantity. In some individuals some physiological effects have been reported due to excessive intake of ascorbic acid. Stein, Hassan & Fox (1976) related megadosing (4g per day) to changes in uric acid metabolism. This may have significance in the formation of uric acid stones and in the management of gout.

Baker (1968) showed that ascorbic acid metabolism varies with intake. One metabolite of ascorbic acid is oxalate (Kallner, 1977). In the human being, high ascorbic acid intake leads to a rapid increase in urinary oxalate; up to 5% of the original ascorbic acid intake may be metabolised in this way. Excess urinary oxalate has been associated with the formation of kidney stones.

Mengel and Greene (1976) showed that the rate of haemolysis (breakdown of red blood cells) was increased in healthy volunteers after consumption of 5g of ascorbic acid.

It has been concluded (Anon, 1976) that excessive doses of anything should be viewed with suspicion until evidence of the utility and safety is firmly established.

HUMAN REQUIREMENTS

From the above discussion, it is obvious that many of the functions of ascorbic acid remain to be elucidated. It is therefore not surprising that there is controversy about the optimal daily intake.

The primary known nutritional role of ascorbic acid is the prevention or cure of scurvy. This is achieved by relatively small quantities of ascorbic acid—less than 10 mg per day will prevent scur plan most individuals (Davidson & Passmore, 1971). However, the amount required to prevent scurvy may not be sufficient to promote optimal health, in view of the many proposed metabolic functions of ascorbic acid, some of which are discussed above.

One school of thought (De & Chakravorty, 1948) is that optimal intake is such that the tissues are saturated with ascorbic acid (i.e. can dissolve no more even if more is ingested). Saturation of the tissues (about 1.0 mg/100ml of blood plasma) requires about ten times the amount of ascorbic acid needed to prevent scurvy, i.e. 60 - 100 mg/day.

Although it is not clear that tissue saturation with ascorbic acid is the most desirable state (Irwin & Hutchins, 1976), this would indicate the upper limit to the amount of ascorbic acid that can be used by the human adult under normal conditions of health and activity. Therefore, optimal intake is likely to be somewhere between 10 mg and 100 mg per day.

The recommended daily intake for Australians is set by the National Health and Medical Research Council which recommends an intake of 30 mg per day (Thomas & Corden, 1977). However, the apparent per capita daily intake of ascorbic acid is about 65 mg as calculated from Australian Bureau of Statistics (1980).

The American authorities recommend an intake of 45 mg per day (Food and Nutrition Board, 1974) down from 60 mg in 1969 (Food & Nutrition Board, 1969). The World Health Organisation recommends 30 mg per day (Passmore, Nicol, Rao, Beaton, & De Maeyer, 1974).

By fortifying several popular items, the operational packs (1977/78 packing program) used by Army provided ascorbic acid in the range 35-170 mg per man per day (James and Forbes-Ewan, 1981). The average ascorbic acid level was 78 mg per man, comparing favourably with the apparent average Australian intake of 65 mg. Allowing for loss of ascorbic acid on storage and that due to wastage, actual intake will be less than 78 mg, but it is unlikely to drop below 30 mg, provided Army rations are consumed before their expiry date.

DEFICIENCY SYMPTOMS

The classical symptoms of scurvy are well known: indolence and weakness, muscular asthenia (lacking strength), pains in the joints followed by swelling of the gums, loosening of the teeth, bone fragility and anaemia (Johnson, 1979). However, scurvy represents an extreme case of ascorbic acid deprivation and it will not appear until months or even years after the ascorbic acid level of the diet has become deficient (Crandon, 1940).

Of more significance to Defence feeding is "subclinical" deficiency, leading to lassitude, fatigue and even mental dysfunction long before scurvy appears (Johnson, 1979).

SOURCES

Many animals, e.g. dogs and rats, synthesize their own ascorbic acid requirement. Man and guinea pigs are among the few that do not. Thus, all man's requirement for ascorbic acid must be derived from his food (Johnson, 1979).

Citrus fruits, leafy green vegetables and potatoes are the main sources of ascorbic acid in Western diets (Davidson and Passmore, 1971). The remainder comes from other fruits and vegetables, fresh milk and a small amount from offal. No significant contribution is made by meat, fish, eggs, bread or other cereal products (Thomas & Corden, 1977).

Our laboratory has recently analysed 60 "bush" or "survival" foods from the Northern Territory. Three foods were found to have very significant amounts of ascorbic acid. Table 2 compares the ascorbic acid levels of these survival foods with some conventionally high sources of ascorbic acid.

The most significant result was the extraordinary ascorbic acid level found in a fruit called "salty plum", a plant of the genus *Terminalia*. The pulp of this fruit contained over 400 mg/100g ascorbic acid, higher than mature red peppers, the best source of vitamin C in Thomas & Corden's (1977) tables.

Table 2 ASCORBIC ACID IN COMMON AND SURVIVAL FOODS

Food (conventional)*	Ascorbic acid (mg/100g)
Potato	14
Tomato	22
Orange	50
Mature red pepper	370
Parsiey	178
Brussel Sprouts	97
Survival Food†	
Salty Plum (Terminalia)	400
Myristica insipida	160
Green/Purple Fruit (unidentified)	100

^{*} Thomas & Corden (1977).

[†] Unpublished results, Armed Forces Food Science Establishment.

CONCLUSION

Ascorbic acid is essential for the prevention of scurvy. Although scurvy does not appear for many months after ascorbic acid deprivation, subclinical effects including lassitude and mental dysfunction can occur earlier.

Also of special significance to the Army is the importance of adequate daily intake to ensure that resistance to infection and wound healing are optimal. Therefore ascorbic acid retention is monitored in operational ration packs and selected items are fortified to ensure an adequate intake of ascorbic acid by ration users. Analysis in our laboratory is currently by titration with dichloroindophenol (AOAC, 1980). This method suffers from being time consuming and inaccurate at low ascorbic acid levels.

We have recently acquired a High Pressure Liquid Chromatography apparatus which should improve both the accuracy and output of ascorbic acid determinations.

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