Complement component C1q unaltered by ascorbate supplementation in healthy men and women

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In the guinea pig, ascorbate administration elevated plasma C1q values 30 to 50% over control values. To determine the effects of vitamin supplementation on C1q in man, healthy men and women (n = 14) consumed placebo capsules for 4 weeks and ascorbate supplements, 1500 mg/d, for 4 weeks, in a double-blind, cross-over fashion. Subjects ate self-selected diets but were instructed to avoid vitamin C-rich foods during the 8-week experimental period. Fasting blood samples, collected at the start of the study and at the end of weeks 4 and 8, were analyzed for ascorbate and C1q, measured directly by immunodiffusion against anti-human C1q. Compared to placebo values, plasma ascorbate rose 92% following the 4-week supplementation period, and plasma C1q rose 18%. The rise in C1q following ascorbate administration was not significant (P = 0.12). Furthermore, plasma ascorbate was not correlated with C1q (r = 0.21, P = 0.14). Hence, in healthy men and women, ascorbate supplementation, at levels 25 times that which is recommended, did not significantly alter plasma C1q levels.

Keywords: ascorbic acid; complement component C1q

Introduction

Plasma protein C1q plays an integral role in disease resistance by identifying infectious, blood-borne agents and subsequently activating complement proteins, a process which destroys the foreign agents directly or facilitates their clearance from blood.[1] C1q contains a collagen-like segment rich in hydroxyproline.[2] Since ascorbic acid is the required reducing agent for the post-ribosomal hydroxylation of proline during the biosynthesis of collagen,[3] and since the levels of connective tissue collagen reflect tissue ascorbate,[4] ascorbate nutriture may also alter C1q levels in plasma. Indeed, several studies in the guinea pig have demonstrated clearly that C1q levels in plasma reflect ascorbic acid nutriture.[5,6]

Given the significant role of C1q in disease resistance, the present study was undertaken to examine the effect of dietary ascorbate on plasma C1q levels in healthy men and women.

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Subjects and methods

Healthy men (n = 3) and women (n = 11), ages 18–40, were screened by personal interview for recent history of medication or illness; tobacco use; unusual dietary habits or supplement use. Participants gave informed consent, and the study was conducted in accordance with the guidelines of the University Human Subjects Research Review Committee at Arizona State University. Subjects consumed self-selected diets during the eight-week experimental period. However, all subjects were provided with a list of 10 commonly consumed vitamin C-rich foods (foods which contained greater than 30 mg ascorbate per serving), and subjects were instructed to avoid these foods during the experimental period. Utilizing a double-blind, placebo-controlled, crossover design, subjects consumed 500 mg ascorbate (Twin Laboratories Inc., Ronkonkoma, NY), or placebo, 3 times daily with meals for four weeks. On day 29 the tablets were changed to placebo, or ascorbate, and the study continued for the remaining four weeks. All subjects reported to the test site weekly to receive a 7-day supply of tablets, to submit to a 24-hour diet recall, and to answer questions regarding study compliance and/or complications.

Fasting blood samples were collected at the start of the study and at the end of weeks 4 and 8. Plasma vitamin C was determined colorimetrically on all blood samples using the 2,4-dinitrophenylhydrazine method of Omaye et al.[7] Plasma C1q levels were measured directly by immunodiffusion against anti-human C1q (Cytotech, San Diego, CA) as described previously.[6] Mean plasma vitamin C and C1q levels following placebo administration were compared to values obtained after the supplementation period using paired t tests. Pearson's correlation was used to examine the relationship between the two variables. All statistical tests were calculated using a Statistical Package for the Social Sciences, SPSS/PC+,[8] and significance was set at $P < 0.05$.

Results and discussion

At the initiation of the eight-week study, plasma vitamin C ranged from 19.9 to 62.5 μmol/L (45.5 ± 3.6 μmol/L,mean ± SEM, $n = 14$). During the eight-week experimental period, mean daily vitamin consumption, estimated by diet recalls, was 68 ± 18 mg per day, a level near that which is recommended by the National Research Council.[9]

Although mean plasma vitamin C rose significantly ($P < 0.001$) following the 4 week supplementation period, compared to the mean value observed following placebo ingestion, mean plasma C1q rose only modestly (+18%, $P = 0.12$) (*Table 1*). Furthermore, plasma vitamin C and C1q values were not significantly correlated ($r = 0.21$, $P = 0.14$). A trend towards a positive association between vitamin status and plasma C1q, however, was apparent. Since the normal value for C1q is considered to be about 70 mg/L,[10,11] and, in the present study, plasma C1q ranged from 32.6 to 63.7 mg/L when vitamin consumption was near the recommended level, vitamin C-depletion may significantly impact C1q levels. Our results simply imply that supplemental vitamin intakes, at levels 25 times that which is recommended, do not significantly raise plasma C1q values above normal values.

In the guinea pig, a striking relationship between ascorbate supplementation and plasma C1q has been noted. C1q rose 30 to 50% in animals fed 25 times the recommended level of dietary ascorbate, and liver vitamin C was highly correlated to plasma C1q ($r = 0.60$, $P < 0.001$).[6] Although low levels of plasma C1q are associated with inadequate complement activation[12,13] and a depressed host defense response,[14,15] the benefits of elevated plasma C1q levels are not readily apparent. Data from the report of Bates et al.[16] imply that although plasma C1q values were significantly elevated in guinea pigs fed supplemental ascorbate compared to vitamin C-deficient animals (see ref. 5 for discussion), complement activation was not improved by vitamin administration. In man, elevated C1q levels are typically associated with inflammatory disease,[17,18] a deleterious condition in which the immune response is stimulated but unchecked.[19] Hence, significantly enhanced C1q production may occur only during activation of the immune system, and not in healthy, non-infected individuals. It remains to be determined, however, whether the modest changes noted in the present study (i.e., an 18% rise in C1q) might alter complement activation or disease resistance in man.

Table 1 Effect of placebo and ascorbate (1500 mg/d) ingestion on plasma ascorbate and C1q in healthy men and women ($n = 14$)

Measure	Pre-study	Placebo	Ascorbate
Plasma ascorbate, μmol/L	45.5 ± 3.6[a]	36.4 ± 3.4[a]	70.0 ± 3.8[a,b]
Plasma C1q, mg/L	60.3 ± 4.2[a]	52.4 ± 2.5[a]	61.6 ± 3.9[a]

[a] Values are the mean ± SEM.
[b] Significantly different from placebo and pre-study values, $P < 0.05$.

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