Vitamin C supplementation in very preterm infants: a randomised controlled trial

B A Darlow, H Buss, F McGill, L Fletcher, P Graham, C C Winterbourn

Objective: To determine whether regulating vitamin C (ascorbic acid: AA) intake to achieve higher or lower plasma concentrations was associated with improved clinical outcome.

Design: A double blind, randomised controlled trial.

Setting: Neonatal intensive care unit at Christchurch Women’s Hospital.

Patients: Infants with birth weight <1500 g or gestation <32 weeks, admitted to the unit within 48 hours of birth.

Intervention: Infants were randomised to one of three protocols with regard to AA supplementation for the first 28 days of life: group LL received low supplementation throughout; group LH received low until day 10 and then high; group HH received high throughout.

Main outcome measures: Primary outcome measures were oxygen requirement at 28 days and 36 weeks postmenstrual age, total days supplemental oxygen, and retinopathy of prematurity. AA concentrations were measured at study entry (day 2), and days 10, 21, and 28.

Results: A total of 119 infants were enrolled over 24 months (mean gestation 28.4 weeks; birth weight 1161 g). Six infants died, and these had significantly higher AA concentrations before randomisation than surviving infants (116 μmol/l (95% confidence interval 90 to 142) vs 51 μmol/l (45 to 58), p < 0.0001). There were no significant differences in primary outcomes between the groups. However, the proportion of surviving infants with an oxygen requirement at 36 weeks postmenstrual age in group HH (19%) was half that in group LL (41%) (p = 0.06).

Conclusions: In a randomised controlled trial, no significant benefits or harmful effects were associated with treatment allocation to higher or lower AA supplementation throughout the first 28 days of life.

Vitamin C (ascorbic acid: AA) is an important aqueous phase antioxidant in cells and plasma. However, at least in vitro, AA also has pro-oxidant activity, principally by reducing ferric iron to the ferrous form, which converts hydrogen peroxide into the more toxic hydroxyl radical (Fenton reaction). AA has a number of important metabolic functions and is actively transported across the placenta. AA concentrations in cord plasma are higher than in the mother’s and, in term infants, plasma concentrations fall considerably over the first 24 hours of life. Preterm infants generally have higher cord AA concentrations than term infants, and concentrations then decline over a few days.

Most preterm infants receive AA as part of a multivitamin supplement, but there are few data on which to base optimum concentrations. One recommendation derives from the concentrations found in healthy breast fed term infants, with an adequate AA concentration stated as ≥34 μmol/l. Breast milk contains 3.5–5.5 mg AA per 100 ml, so that an average infant having 150 ml/kg/day of milk will receive 5.2–8 mg/kg/day. The alternative view is that preterm infants should receive higher doses of AA, 25–31 mg/kg/day, to achieve concentrations closer to those in utero in the third trimester.

There have been few studies of the relation between AA concentration and morbidity in very preterm infants. Silvers et al. reported that plasma AA concentrations within 2 hours of birth were significantly higher in infants who died compared with survivors. These researchers also observed that higher AA concentrations on day 2 were associated with a greater risk of developing bronchopulmonary dysplasia. In contrast, Moison et al. reported lower plasma AA concentrations on day 10 in preterm infants who developed bronchopulmonary dysplasia compared with those who did not.

In this study, we aimed to determine in a randomised, controlled trial whether regulating AA intake to achieve higher or lower plasma concentrations is associated with improved clinical outcome. We hypothesised that maintaining a lower plasma AA concentration (target 35–50 μmol/l) in the first week of life and a higher concentration (target 90 μmol/l) in weeks 3–4 would be accompanied by least morbidity.

METHODS

Infants with birth weight <1500 g or gestation <32 weeks admitted to the neonatal intensive care unit at Christchurch Women’s Hospital were eligible for study. Signed, informed parental consent was obtained within 72 hours of birth. The study was approved by the Canterbury Ethics Committee.

This was a randomised, double blinded, placebo controlled trial. Infants were randomised by pharmacy through sealed envelopes to one of three protocols for AA supplementation for the first 28 days of life: group LL received low AA supplements throughout; group LH received low AA supplements until day 10, then high supplements until day 28; group HH received high AA supplements throughout.

Abbreviations: AA, ascorbic acid; CRIB, clinical risk index for babies; ROP, retinopathy of prematurity
AA supplements

Standard parenteral multivitamin supplementation was 1 ml/kg/day Soluvit N (Kabi Pharmacia AB, Stockholm, Sweden) plus 4 ml/kg/day Vitlipid N (Kabi Pharmacia AB), both added to Intralipid (Kabi Pharmacia AB), which provided 10 mg/kg/day AA. While requiring parenteral nutrition, infants randomised to low AA received this standard multivitamin regimen, and infants randomised to a high regimen received the standard regimen plus an extra 20 mg/kg/day AA. The combination of Soluvit and Vitlipid* was chosen because this regimen delivers comparable amounts of all vitamins to that delivered by 2 ml/kg/day MVI-Pediatric (Rhone-Poulenc Rorer, Montreal, Canada),† except that the AA content is lower (table 1).

Our policy is to encourage enteral feeding with mother’s own breast milk (assumed to contain 4.0 mg/100 ml AA). Formula fed infants received S26 LBW formula (Wyeth, Auckland, New Zealand), which contains 11 mg/100 ml AA. S26 LBW powder (5 g/100 ml, containing 3.3 mg AA) was used to fortify breast milk when required. Orally fed infants also received Vitadol C (Nutricia, Auckland, New Zealand), which contains 33 mg AA in 10 drops (0.3 ml). Table 2 shows feeding regimens for infants fed 150 ml/kg/day. Infants randomised to high AA (group HH and group LH from day 11 onwards) also received an additional 20 mg/kg/day AA as a clear oral solution (10 mg/ml). Infants randomised to low AA (group LL and group LH to day 10) received sterile water as placebo. Both AA solution and placebo were dispensed as 2 ml/kg from a separate named bottle for each infant, marked “vitamin C or placebo”.

Measurement of AA and protein carbonyls

Plasma AA concentrations were measured at study entry (usually day 2) and at day 10 (range 8–12), 21, and 28, on heparinised blood (0.4 ml) collected at the time of routine sampling. Samples were stored at 4°C for no more than 30 minutes before the plasma was separated and frozen at −80°C. AA, in the reduced form, was measured by high performance liquid chromatography (HPLC) using a C18 column with electrochemical detection. The detection limit was 1 μM, and the intra-assay coefficient of variation for plasma containing 60 μM AA was 3.9%. Protein carbonyls were measured on selected day 28 samples by an enzyme linked immunosorbent assay (ELISA) method that involves derivatising with 2,4-dinitrophenylhydrazine, using a commercial kit (Zenith Technology, Dunedin, New Zealand).

Outcome measures

Clinicians and laboratory staff remained blinded to treatment allocation throughout the trial. Perinatal and neonatal data, including the clinical risk index for babies (CRIB) score, were collected as part of an ongoing clinical audit. Primary outcomes were oxygen requirement at 28 days and 36 weeks postmenstrual age; total days supplemental oxygen, and ROP in infants eligible for screening. Suplemental oxygen was generally administered to infants unable to maintain a saturation of 95% or more in room air. ROP was assessed by an experienced paediatric ophthalmologist and reported using international criteria. Screening for ROP is routinely undertaken in New Zealand for infants of <1250 g birth weight or <31 weeks gestation, and outside these limits at physician discretion.

Power calculations and statistical analysis

Power calculations were conducted, using Monte Carlo simulation based on our preliminary study, for logistic regressions of major outcomes on AA concentrations, with adjustment for gestational age. They suggested that 120 patients would provide at least 80% power to detect a 40% reduction in risk for major outcomes—that is, a relative risk of 0.6—assuming significance testing at the 5% level.

Data were analysed in two ways. Firstly, conventional between group analysis by intention to treat was performed using χ² tests, Fisher’s exact test, or non-parametric analysis of variance (Kruskal-Wallis) as appropriate. Adjustment for covariate imbalance between the three treatment groups was achieved using a logistic regression model. Secondly, the association between AA concentrations, treated as a continuous variable, and binary outcomes was investigated with logistic regression models. Logistic regression models were used to estimate relative risks of morbidity (ROP, oxygen requirement at 28 days, and at 36 weeks postmenstrual age) comparing risk at optimal AA concentrations at different time points with risk at higher or lower concentrations.

RESULTS

A total of 119 infants, 40 in both LL and LH groups and 39 in the HH group, and 90% of those eligible, were enrolled over 24 months. The mean (SD) gestational age was 28.4

Table 1

<table>
<thead>
<tr>
<th>Treatment</th>
<th>C (mg)</th>
<th>A (mg)</th>
<th>D (mg)</th>
<th>B₁ (mg)</th>
<th>B₂ (mg)</th>
<th>B₃ (mg)</th>
<th>Niacin (mg)</th>
<th>Dextanthal (mg)</th>
<th>E (mg)</th>
<th>Biotin (μg)</th>
<th>Folate (μg)</th>
<th>B₁₂ (μg)</th>
<th>K (μg)</th>
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<tr>
<td>EBM</td>
<td>22.5</td>
<td>0.47</td>
<td>6.6</td>
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<tr>
<td>S26</td>
<td>26.4</td>
<td>0.37</td>
<td>5.8</td>
<td>2.4</td>
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</tr>
<tr>
<td>EBM+5 g/100 ml</td>
<td>20.4</td>
<td>0.32</td>
<td>4.3</td>
<td>1.4</td>
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</tbody>
</table>

*Nutricia, Auckland, New Zealand.

EBM, expressed breast milk.

Table 2

<table>
<thead>
<tr>
<th>Treatment</th>
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<td>4.3</td>
<td>1.4</td>
</tr>
</tbody>
</table>

*Nutricia, Auckland, New Zealand.

EBM, expressed breast milk.
There was no difference in birth weight, gestation, or CRIB score between groups, which were also well matched in other respects (table 3).

Six infants (5%) died, one each on days 1, 3, 5, 7, 8, and 9. One infant was in group LL, two in group LH, and three in group HH. Their mean CRIB score was 5.2 (n = 5; missing data for one infant). These six infants had significantly higher AA concentrations before randomisation (day 2) than the 111 surviving infants (116 nmol/l (95% CI 90 to 142) v 51 nmol/l (95% CI 45 to 58), p = 0.0001). The odds ratio (OR) for risk of death corresponding to a 35 nmol/l (approximately 1 SD) increase in AA concentration before randomisation was 5.21 (95% CI 2.16 to 19.23), and this estimate changed little after adjustment for gestational age and CRIB score (OR 5.26 (95% CI 2.15 to 20.83)).

Table 3 shows the relations between group assignment, AA concentrations, and primary and other clinical outcomes. Based on logistic regression modelling, there was no significant difference between groups with respect to AA concentrations before randomisation (day 2), but differences on days 10, 21, and 28 were highly significant and in the direction predicted (fig 1). There were no significant differences in primary outcomes between the groups, although the proportion of surviving infants with an oxygen requirement at 36 weeks postmenstrual age in group HH (19%) was half that in group LL (41%). When the analysis was restricted to these two groups, this difference approached significance (p = 0.06). Adjusting data for gestational age and CRIB score, using logistic regression, did not alter the between group differences substantially (table 5), with the standardised proportion of surviving infants with an oxygen requirement at 36 weeks postmenstrual age in group HH (16%) remaining half that in group LL (36%) (p = 0.06). There were no significant differences between groups for other outcomes.

Logistic regression analyses at each time point for oxygen requirement at 28 days and 36 weeks, and ROP showed no significant relations with AA concentration after adjustment for gestational age and CRIB score (table 6).

To assess whether high AA concentrations could have a pro-oxidant effect, we measured protein carbonyls as an index of protein oxidation in a subset of plasma samples. We selected two groups of day 28 samples, those with AA concentrations ≥ 50 μmol/l (n = 30; mean (SD) 31 (12) μmol/l) and those with AA concentrations ≥ 80 μmol/l (n = 27; 103 (29) μmol/l). The low AA samples had a mean protein carbonyl concentration of 0.133 (0.071) nmol/mg compared with 0.126 (0.059) nmol/mg for the high AA samples; these values were not significantly different.

DISCUSSION

We hypothesised that having lower AA concentrations in the first week of life (target 35–50 μmol/l) and higher concentrations in weeks 3–4 (target 90 μmol/l) would decrease morbidity (chronic lung disease and ROP) in very low birthweight infants. Therefore we designed the study with three groups, one aiming to keep AA concentrations low throughout (LL), one with high concentrations throughout (HH), and a group crossing over from low initially to high at day 10 (LH). Although we achieved significantly different AA concentrations at days 10, 21, and 28 between the groups in the direction expected, there were no significant differences in primary outcomes. The results did show trends towards better respiratory outcome being associated with higher AA intake, and it is a possibility that by distributing infants in...
Table 4  Plasma ascorbic acid concentrations (μmol/l) and clinical outcomes by study group

<table>
<thead>
<tr>
<th></th>
<th>LL</th>
<th>LH</th>
<th>HH</th>
<th>p Value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N Mean (95% CI)</td>
<td>N Mean (95% CI)</td>
<td>N Mean (95% CI)</td>
<td></td>
</tr>
<tr>
<td>Ascorbic acid day 2</td>
<td>38 53.9 (44.1 to 63.4)</td>
<td>39 51.2 (40.2 to 62.1)</td>
<td>39 58.3 (45.1 to 71.6)</td>
<td>0.69</td>
</tr>
<tr>
<td>Ascorbic acid day 10</td>
<td>32 385 (28.9 to 48.1)</td>
<td>29 35.9 (28.4 to 43.4)</td>
<td>26 69.7 (49.6 to 89.8)</td>
<td>0.001</td>
</tr>
<tr>
<td>Ascorbic acid day 21</td>
<td>33 47.7 (39.5 to 55.8)</td>
<td>25 55.8 (44.7 to 66.9)</td>
<td>32 70.6 (57.5 to 83.7)</td>
<td>0.006</td>
</tr>
<tr>
<td>Ascorbic acid day 28</td>
<td>33 51.5 (42.0 to 61.0)</td>
<td>31 59.4 (50.0 to 68.9)</td>
<td>29 78.6 (63.4 to 93.8)</td>
<td>0.0003</td>
</tr>
<tr>
<td>Days of ventilation</td>
<td>31 10.3 (4.8 to 15.7)</td>
<td>27 9.2 (3.5 to 15.0)</td>
<td>25 7.5 (2.7 to 12.2)</td>
<td>0.98</td>
</tr>
<tr>
<td>Days of oxygen therapy</td>
<td>40 29 (18 to 40)</td>
<td>40 25 (12 to 38)</td>
<td>39 28 (14 to 41)</td>
<td>0.08</td>
</tr>
<tr>
<td>Days parenteral nutrition</td>
<td>32 11.7 (7.5 to 14.0)</td>
<td>29 9.9 (5.5 to 14.4)</td>
<td>30 7.7 (5.6 to 9.7)</td>
<td>0.32</td>
</tr>
<tr>
<td>Age (days) at full oral feeding</td>
<td>40 10.6 (8.2 to 13.0)</td>
<td>40 88.8 (58 to 11.8)</td>
<td>39 8.2 (6.2 to 10.2)</td>
<td>0.12</td>
</tr>
<tr>
<td>Oxygen at 28 days</td>
<td>39 51 (36 to 67)</td>
<td>38 37 (22 to 52)</td>
<td>36 36 (20 to 52)</td>
<td>0.32</td>
</tr>
<tr>
<td>Oxygen at 36 weeks PMA</td>
<td>39 41 (26 to 57)</td>
<td>38 29 (15 to 43)</td>
<td>36 19 (7 to 32)</td>
<td>0.12</td>
</tr>
<tr>
<td>Death or oxygen at 28 days</td>
<td>40 53 (37 to 68)</td>
<td>40 40 (25 to 55)</td>
<td>39 41 (26 to 57)</td>
<td>0.46</td>
</tr>
<tr>
<td>Death or oxygen at 36 weeks PMA</td>
<td>40 43 (27 to 58)</td>
<td>40 33 (18 to 47)</td>
<td>39 26 (12 to 39)</td>
<td>0.28</td>
</tr>
<tr>
<td>Postnatal steroids</td>
<td>40 15 (4 to 26)</td>
<td>40 10.0 (1 to 19)</td>
<td>39 5.1 (2 to 12)</td>
<td>0.39</td>
</tr>
<tr>
<td>ROP</td>
<td>31 39 (22 to 56)</td>
<td>30 37 (19 to 54)</td>
<td>25 40 (21 to 59)</td>
<td>0.97</td>
</tr>
<tr>
<td>Necrotising enterocolitis</td>
<td>40 2.5 (0 to 7)</td>
<td>40 7.5 (0 to 16)</td>
<td>39 2.6 (0 to 8)</td>
<td>0.62</td>
</tr>
</tbody>
</table>

Study groups: LL, received low ascorbic acid (AA) supplementation throughout; LH, received low AA until day 10 and then high; HH, received high AA throughout.

*Screening for retinopathy of prematurity (ROP) confined to infants < 31 weeks gestation or <1250 g birth weight.
†Kruskal-Wallis test.
‡χ² test with 2 degrees of freedom.
*Fisher’s exact test.
PMA, postmenstrual age.
three groups, the study lacked sufficient power to detect a difference. Comparing only the LL and HH groups, the mean requirement for oxygen at 36 weeks was twice as high in the low intake group. Most would consider this difference clinically significant and it approached statistical significance (p = 0.06), as did comparisons across the three groups with the data corrected for gestation and CRIB score (table 5, p = 0.06). We must be cautious in attributing any advantage to the high dose AA, as there was no relation between AA concentrations at any time point and outcome measures. However, there was no evidence that the high dose AA was harmful, and the results suggest that supplementing very preterm infants at this higher AA concentration may be beneficial.

One possible limitation is that AA concentrations in the high groups did not reach our target range. This target was based on our preliminary observational study where the mean AA concentration at 28 days for infants without bronchopulmonary dysplasia was 93 μmol/l. During that study, infants who were fed parenterally received 25 mg/kg/day AA in addition to the standard dose for 10 days raised plasma AA concentrations by 30 unit increases after adjustment for gestational age and CRIB score (table 5, p = 0.06), as did comparisons across the three groups with the data corrected for gestation and CRIB score (table 5, p = 0.06). We must be cautious in attributing any advantage to the high dose AA, as there was no relation between AA concentrations at any time point and outcome measures. However, there was no evidence that the high dose AA was harmful, and the results suggest that supplementing very preterm infants at this higher AA concentration may be beneficial.

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effects, we suggest it may be reasonable to provide at least this level of supplementation.

ACKNOWLEDGEMENTS
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Competing interests: none declared

REFERENCES
32 Berger TM, Frei B. Pro- or antioxidant activity of vitamin C in preterm infants? [Correspondence]. Arch Dis Child Fetal Neonatal Ed 1995;72:F211.