Stability of intravenous vitamin C solutions: a technical report

Anitra Carr, Christina Wohlrab, Paul Young and Rinaldo Bellomo

ABSTRACT

Background: There has recently been a surge of interest in intravenous (IV) vitamin C as a potential therapy in intensive care unit (ICU) patients, particularly in those with septic shock. Establishing the safety and efficacy of IV vitamin C therapy through rigorously conducted randomised controlled trials is a priority. A key logistical issue for such trials is to establish the stability of IV vitamin C solutions prepared for infusion ahead of time. Accordingly, we aimed to assess the stability of IV vitamin C solutions over time using doses of vitamin C from previous pilot trials.

Methods: We used spectrophotometry to measure the concentration of vitamin C remaining in solutions of 1.5 g per 50 mL of 0.9% saline and 2.5 g per 50 mL of dextrose 5% in water (D5W) at 0, 1, 3, 6, 9, 24, 48, 72 and 96 hours after preparation. The concentration of vitamin C in these solutions over time was assessed at 4°C in the dark and at ambient temperature and light.

Results: The concentration of vitamin C in diluted solutions was essentially unchanged over a period of 24 hours, and decreased less than 10% by 96 hours both at 4°C in the dark and at ambient temperature and light.

Conclusions: Our findings suggest that vitamin C solutions of 1.5 g per 50 mL of 0.9% saline and 2.5 g per 50 mL of D5W remain stable for up to 96 hours and do not need to be protected from light.

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Vitamin C (ascorbic acid) is an essential nutrient for humans as, unlike most other animal species, we lack the ability to synthesise the molecule. Vitamin C functions as a cofactor for a family of metalloenzymes, with numerous regulatory and biosynthetic roles in the body, including synthesis of the vasopressors noradrenaline and vasopressin.[1] Critically ill patients have a high incidence of hypovitaminosis C,[2,3] but while a number of preliminary studies have assessed the pharmacokinetics and therapeutic effects of intravenous (IV) vitamin C in critically ill patients,[4-7] further trials are needed to establish the role of IV vitamin C in clinical practice.

One important logistical issue for the conduct of such clinical trials is to ensure the stability of vitamin C solutions prepared for infusion. This issue is important because unblinded research staff, who typically only work office hours, may wish to prepare masked study medication ahead of time to allow blinding of treating clinicians for overnight and weekend administration. In previous studies, IV vitamin C was diluted in dextrose 5% in water (D5W) or 0.9% saline and administered in four equal doses totalling 6 g/day or up to 200 mg/kg/day.[4-6] The stability of vitamin C solutions prepared in this way, however, has not previously been reported.

We hypothesised that vitamin C solutions prepared for infusion would retain a stable concentration of vitamin C over a sufficiently long period so that study medication could be prepared during office hours for use overnight and at weekends.

Methods

Stability of vitamin C solutions

The ASCOR L 500 stock solution (500 mg/mL ascorbic acid; McGuff Pharmaceuticals, Santa Ana, USA) was stored in the dark at 4°C, and a 0.2 μm air filter needle was used to keep the solution sterile during withdrawal of aliquots. Aliquots of the ASCOR L 500 were diluted to 1.5 g per 50 mL of sterile 0.9% saline or 2.5 g per 50 mL of D5W (Baxter, Christchurch, New Zealand) in line with previously published protocols.[4-6] The solutions were stored at 4°C in the dark or at ambient temperature and light. Duplicate aliquots were withdrawn at 0, 1, 3, 6, 9, 24, 48, 72 and 96 hours. All aliquots were serially diluted to 50 μmol/L in 77 mmol/L perchloric acid containing the metal chelator diethylenetriaminepentaacetic acid (100 μmol/L), and the ascorbic acid absorbance measured spectrophotometrically at 245 nm as previously described.[8] This experiment was repeated and the data averaged. Within-sample variance (duplicate readings) was 0.003% and between-sample variance (duplicate samples) was 0.1%. The stability of the ASCOR L 500 stock solution was also tested daily following serial dilution and spectrophotometric measurement as described above.

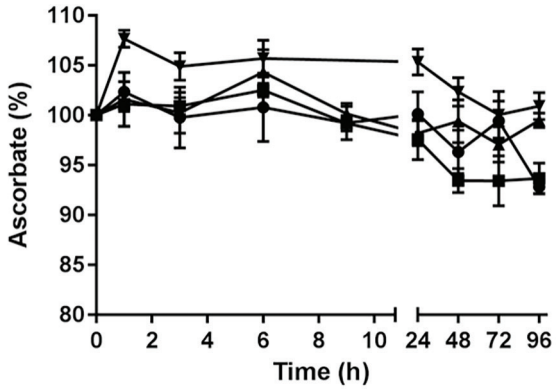
Results

Under all four conditions tested (ie, diluted in 0.9% saline or D5W and stored in the dark at 4°C or in the light at ambient temperature), the concentration of the vitamin C solutions was essentially unchanged over a period of 24 hours and decreased by less than 10% over 96 hours (Figure 1). The vitamin C stock solution (ASCOR L 500) was stable for 24 hours at 4°C and less than 10% vitamin C loss was observed after 48 hours.

Discussion

We conducted a bench experiment using spectrophotometry to measure the concentration of vitamin C remaining over

Figure 1. Stability of diluted intravenous vitamin C solutions



Ascorbic acid was diluted in 0.9% saline and stored in the dark at 4°C (■) or in the light at ambient temperature (●), or diluted in dextrose 5% in water and stored in the dark at 4°C (▼) or in the light at ambient temperature (▲). Data represent mean of two independent experiments. Error bars represent the range. Note that glucose has enhanced viscosity at 4°C (▼); therefore, the vitamin C solution may not have had sufficient time to equilibrate prior to the time zero sampling.

time in vitamin C solutions prepared for infusion in 0.9% saline or D5W. We found that such solutions were stable at 4°C in the dark and at ambient temperature and light, with concentrations decreasing by less than 10% over 96 hours of storage. Although gram doses of IV vitamin C have been administered daily in a number of critical care studies,[4-7] to the best of our knowledge, there have been no previous reports on the stability of these solutions.

Because vitamin C is a potent electron donor and antioxidant, it is susceptible to oxidation.[9] Oxidation of vitamin C can be influenced by temperature, pH and the presence of transition metal ions such as copper and iron.[8] Vitamin C can exhibit instability in biological samples, such as plasma, particularly at ambient temperature;[8] however, our findings indicate that it is much more stable in 0.9% saline and D5W. We have tested only one of many commercially available vitamin C formulations for infusion (ASCOR L 500), but, as ASCOR L 500 contains EDTA, it might be expected to be less stable than formulations that do not contain EDTA because transition metal ion/EDTA complexes are redox active and can facilitate oxidation of vitamin C.[8]

Our data indicate that gram doses of vitamin C can be diluted in 0.9% saline or D5W and stored for up to 96 hours without loss of stability. The product information recommends that vitamin C is protected from light.[10] While this may be necessary to protect it from degradation over the longer term, our findings suggest that there is little loss of vitamin C with exposure to light over a period of 96 hours. Thus, it appears unnecessary to protect vitamin C from light during infusion where exposure is only likely to occur for an hour or two.

Conclusion

We have shown that diluted IV vitamin C solutions can be prepared up to 96 hours in advance of administration and do not need to be protected from ambient light, particularly during administration. These findings are important for the conduct of future trials because they should allow unblinded research or pharmacy staff to prepare masked solutions of study medication during office hours for use overnight and at weekends.

Competing interests

None declared.

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References

1 Carr AC, Shaw GM, Fowler AA, Natarajan R. Ascorbate-dependent vasopressor synthesis: a rationale for vitamin C administration in severe sepsis and septic shock? *Crit Care* 2015; 19: e418.
2 Carr AC, Rosengrave PC, Bayer S, et al. Hypovitaminosis C and vitamin C deficiency in critically ill patients despite recommended enteral and parenteral intakes. *Crit Care* 2017; 21: 300.
3 Story DA, Ronco C, Bellomo R. Trace element and vitamin concentrations and losses in critically ill patients treated with continuous venovenous hemofiltration. *Crit Care Med* 1999; 27: 220-3.
4 Fowler AA, Syed AA, Knowlson S, et al. Phase I safety trial of intravenous ascorbic acid in patients with severe sepsis. *J Transl Med* 2014; 12: 32.
5 Zabet MH, Mohammadi M, Ramezani M, Khalili H. Effect of high-dose Ascorbic acid on vasopressor's requirement in septic shock. *J Res Pharm Pract* 2016; 5: 94-100.
6 Marik PE, Khangoora V, Rivera R, et al. Hydrocortisone, vitamin C, and thiamine for the treatment of severe sepsis and septic shock: a retrospective before-after study. *Chest* 2017; 151: 1229-38.
7 de Grooth HJ, Manubulu-Choo WP, Zandvliet AS, et al. Vitamin-C pharmacokinetics in critically ill patients: a randomized trial of four intravenous regimens. *Chest* 2018; 153: 1368-77.
8 Pullar JM, Bayer S, Carr AC. Appropriate handling, processing and analysis of blood samples is essential to avoid oxidation of vitamin C to dehydroascorbic acid. *Antioxidants* 2018; 7: 29.
9 Carr A, Frei B. Does vitamin C act as a pro-oxidant under physiological conditions? *FASEB J* 1999; 13: 1007-24.
10 New Zealand Datasheet for ASCOR L 500. http://www.medsafe.govt.nz/profs/Datasheet/a/ascorlinj.pdf (viewed March 2018).