Published in final edited form as:

JPEN J Parenter Enteral Nutr. 2015 May ; 39(4): 401–409. doi:10.1177/0148607114529994.

Glutamine and Antioxidants in the Critically Ill Patient: A Post Hoc Analysis of a Large-Scale Randomized Trial

Daren K. Heyland, MD[1], **Gunnar Elke, MD**[2], **Deborah Cook, MD**[3], **Mette M. Berger, MD, PhD**[4], **Paul E. Wischmeyer, MD**[5], **Martin Albert, MD**[6], **John Muscedere, MD**[1], **Gwynne Jones, MD**[7], **Andrew G. Day, MSc**[1] **Canadian Critical Care Trials Group**

[1]Kingston General Hospital, Kingston, Ontario, Canada [2]University Medical Center Schleswig-Holstein, Campus Kiel, Kiel, Germany [3]St Joseph's Healthcare, Hamilton, Ontario, Canada [4]University Hospital CHUV, Lausanne, Switzerland [5]University of Colorado School of Medicine, Aurora [6]Hôpital du Sacré-Coeur de Montreal, Quebec, Canada [7]Ottawa Hospital, General Campus, Ottawa, Ontario, Canada

Abstract

Background—The recent large randomized controlled trial of glutamine and antioxidant supplementation suggested that high-dose glutamine is associated with increased mortality in critically ill patients with multiorgan failure. The objectives of the present analyses were to reevaluate the effect of supplementation after controlling for baseline covariates and to identify potentially important subgroup effects.

Materials and Methods—This study was a post hoc analysis of a prospective factorial 2 × 2 randomized trial conducted in 40 intensive care units in North America and Europe. In total, 1223 mechanically ventilated adult patients with multiorgan failure were randomized to receive glutamine, antioxidants, both glutamine and antioxidants, or placebo administered separate from artificial nutrition. We compared each of the 3 active treatment arms (glutamine alone, antioxidants alone, and glutamine + antioxidants) with placebo on 28-day mortality. Post hoc, treatment effects were examined within subgroups defined by baseline patient characteristics. Logistic regression was used to estimate treatment effects within subgroups after adjustment for baseline covariates and to identify treatment-by-subgroup interactions (effect modification).

Results—The 28-day mortality rates in the placebo, glutamine, antioxidant, and combination arms were 25%, 32%, 29%, and 33%, respectively. After adjusting for prespecified baseline covariates, the adjusted odds ratio of 28-day mortality vs placebo was 1.5 (95% confidence interval, 1.0–2.1, $P = .05$), 1.2 (0.8–1.8, $P = .40$), and 1.4 (0.9–2.0, $P = .09$) for glutamine, antioxidant, and glutamine plus antioxidant arms, respectively. In the post hoc subgroup analysis, both glutamine and antioxidants appeared most harmful in patients with baseline renal dysfunction. No subgroups suggested reduced mortality with supplements.

Corresponding Author: Daren K. Heyland, MD, Kingston General Hospital, Angada 4, Kingston, ON K7L 2V7, Canada. dkh2@queensu.ca.

Supplementary material for this article is available on the *Journal of Parenteral and Enteral Nutrition* website at http://jpen.sagepub.com/supplemental.

Conclusions—After adjustment for baseline covariates, early provision of high-dose glutamine administered separately from artificial nutrition was not beneficial and may be associated with increased mortality in critically ill patients with multiorgan failure. For both glutamine and antioxidants, the greatest potential for harm was observed in patients with multiorgan failure that included renal dysfunction upon study enrollment.

Keywords

randomized clinical trials; glutamine; antioxidants; post hoc analysis; critical care

Introduction

Patients admitted to the intensive care unit (ICU) with multiorgan failure are often deficient in key nutrients responsible for responding to oxidant stress and cellular injury.[1–6] Multiple studies have documented that these patients also have low plasma levels of key nutrients involved in antioxidant and cell defense mechanisms.[2,7] Specifically, glutamine depletion has been associated with immune dysfunction[8] and increased mortality.[9,10] Finally, meta-analyses of randomized trials suggest glutamine and antioxidants supplementation in critically ill patients may be associated with a survival advantage.[11,12]

Building on this rationale, we conducted a large randomized trial powered to evaluate the effect of high-dose glutamine and antioxidant supplementation on mortality in 1223 critically ill patients.[13] We elected to study patients with multiorgan failure given their high degree of oxidative stress, since they may be most likely to benefit from aggressive supplementation of antioxidants and glutamine. Contrary to our hypothesis, the primary analysis demonstrated no clinical benefit of our interventions and identified a trend toward increased mortality at 28 days among patients who received high-dose glutamine compared with those who did not receive glutamine (32.4% vs 27.2%; adjusted odds ratio [OR], 1.28; 95% confidence interval [CI], 1.00–1.64; $P = .049$ [P value of <.044 required to declare statistically significant because of adjustment for interim analyses]). We did not find an effect of antioxidants on 28-day mortality (30.8% vs. 28.8%; adjusted OR, 1.09; 95% CI, 0.86–1.40; $P = .48$).[13]

Given these unexpected findings and additional concerns about baseline imbalances, we conducted post hoc analyses to examine the treatment effects of high-dose glutamine and antioxidants alone or in combination after controlling for baseline patient characteristics and to examine whether the treatment effect varied in specific subgroups of patients. In contrast to our primary publication, herein we report the outcomes by each treatment group individually (glutamine alone, antioxidants alone, and glutamine + antioxidants) vs placebo.

Methods

Details of the study methods and main results were published previously.[14] In brief, this trial was a randomized, controlled, investigator-initiated trial in 40 ICUs in Canada, the United States, and Europe. We obtained local jurisdictional and institutional research ethics board approval from all participating sites to conduct this study (see “List of Investigators and Participating Sites”) and written informed consent from patients or their legal representatives

before enrollment. Randomization was concealed and stratified by center using permuted blocks of random size using a secure central web-based system. We enrolled consecutive mechanically ventilated adults admitted to participating ICUs if they had 2 or more of the following organ failures related to their acute illness:

1. A PaO_2/FiO_2 ratio of ≤300
2. Clinical evidence of hypoperfusion defined as vasopressor agents for ≥2 hours
3. In patients without known renal disease, renal dysfunction defined as a serum creatinine ≥171 μmol/L or a urine output of <500 mL/past 24 hours (or 80 mL/past 4 hours if a 24-hour period of observation not available), and in patients

List of Investigators and Participating Sites
Number in parentheses refers to number of patients enrolled at each site.
Canada
Kingston General Hospital (133), Kingston, Ontario: John Muscedere, Charlene Hammond, Monica Meyers, Susan Fleury, Nicole O'Callaghan
St Joseph's Healthcare (91), Hamilton, Ontario: Deborah Cook, Ellen McDonald, France Clarke
Ottawa Hospital, General Campus (254), Ottawa, Ontario: Gwynne Jones, Irene Watpool, Tracy McArdle, Rebecca Porteous
Ottawa Hospital, Civic Campus (91), Ottawa, Ontario: Guiseppe Pagliarello, Tracy McArdle
Vancouver General Hospital (35), Vancouver, British Columbia: Dean Chittock, Maureen Gardner, Susie Logie, Denise Foster
Hôpital du Sacré-Coeur de Montréal (89), Montréal, Québec: Martin Albert, Patrice Deroy, Huber Simard
Hôpital Maisonneuve-Rosemont (21), Montreal, Quebec: Stephane Ahern, Johanne Harvey
Royal Victoria Hospital (15), Montreal, Quebec: Sheldon Magder, Laura Banici
Royal Alexandra Hospital (36), Edmonton, Alberta: Jim Kutsogiannis, Patrica Thompson, Kirby Scott, Reagan Bartel, Darlene Jossy, Christine Krawchuk
Grey Nun's Hospital (23), Edmonton, Alberta: Dan Stollery, Jennifer Barchard, Michael Krause
Victoria General Hospital (11), Victoria, British Columbia: Gordon Wood, Fiona Auld, Leslie Atkins
London Health Sciences Centre (21), London, Ontario: Claudio Martin, Eileen Campbell
Capital Health (20): Rick Hall, Lisa Julien
Montreal General Hospital (34), Montreal, Quebec: Kosar Khwaja, Laura Banici
Hôpital l-Enfant-Jesus (45), Quebec City, Quebec: Francois Lauzier, Chantal Gagne, Marie Thibodeault
Royal Jubilee (15), Victoria, British Columbia: Gordon Wood, Fiona Auld, Peggy Leonard, Leslie Atkins
Mount Sinai Hospital (50), Toronto, Ontario: Sangeeta Mehta, Maedean Brown
University Hospital, London Health Sciences Centre (13), London, Ontario: Tina Mele, Tracey Bentall
Institut universitaire de cardiologie et de pneumologie de Québec (25), Quebec City, Quebec: Francois Lellouche, Marie-Claude Ferland
Hamilton General Hospital (4), Hamilton, Ontario: Maureen Meade, Lori Hand
University of Ottawa Heart Institute (4), Ottawa, Ontario: Bernard McDonald, Denyse Winch
Sunnybrook Health Sciences Centre (2), Toronto, Ontario: Rob Fowler, Nicole Marinoff
St Paul's Hospital (6), Vancouver, British Columbia: Peter Dodek, Betty Jean Ashley
Health Sciences Centre Winnipeg (8), Winnipeg, Manitoba: Kim Wiebe, Wendy Janz
United States
University of Colorado at Denver and Health Sciences Center (22), Denver, Colorado: Paul Wischmeyer, Elizabeth Luzier, Angela Baer
Miami Valley Hospital (16), Dayton, Ohio: Mary McCarthy, Laurie Chowayou, Kimberly Garrett
Fletcher Allen (12), Burlington, Vermont: Renee Stapleton, Julie Martin, Bridget Shea
University of Louisville Hospital (24), Louisville, Kentucky: Mohamed Saad, Crissie DeSpirito
University of Texas Health Sciences Center (13), Houston, Texas: Rosemary Kosar, Jeanette Podbielski, Laura Vincent, Kristi Morin
Jewish Hospital (13), Louisville, Kentucky: Mohamed Saad, Crissie DeSpirito
Atlanticare (5), Atlantic City, New Jersey: Catherine Dudick, Jackie White
Pennsylvania State University and the Milton Hershey Medical Center (1), Hershey, Pennsylvania: Heidi Frankel, Lee Ann Smith
Intermountain (6), Salt Lake City, Utah: Tom White, Merin Kinikini, Ben Briggs
Mayo Clinic, (22), Phoenix, Arizona: Michael Murray, Andre Watkins
Europe
Switzerland. Centre hospitalier universitaire vaudois (CHUV) (12), Lausanne: Mette Berger, Frederik Delodder
Germany. University Medical Center Schleswig-Holstein, Campus Kiel, Department of Anesthesiology and Intensive Care Medicine, Kiel (7): Gunnar Elke, Norbert Weiler, Nina Schulz-Ruthenberg, Stefanie D'Aria
Ernst-Moritz-Arndt University Greifswald, Department of Anesthesiology and Intensive Care Medicine, Greifswald (8): Matthias Gründling, Sven-Olaf Kuhn, Liane Guderian
Asklepios Clinics Hamburg-Altona, Department of Anesthesiology and Intensive Care Medicine, Hamburg (6): Hanswerner Bause, Philip Gabriel, Axel Prause, Cornelia Wolf
Belgium. UZ Brussel, Brussels (2): Herbert Spapen, Godelieve Opdenacker
University Hospital of Liège, Liège (8): Jean Charles Preiser, Joelle Lefranq

with acute-on-chronic renal failure (predialysis), an absolute increase of ≥80 μmol/L from baseline or a urine output of <500 mL/past 24 hours (or 80 mL/past 4 hours)

4. A platelet count of $\leq 50 \times 10^9$/L

Patients were excluded for 1 or more of the following criteria: (1) >24 hours from admission to ICU, (2) moribund patients (not expected to be in ICU for more than 48 hours due to imminent death), (3) a lack of commitment to full aggressive care (anticipated withholding or withdrawing treatments in the first week), (4) absolute contraindication to enteral nutrients (eg, gastrointestinal [GI] perforation, obstruction, or no GI tract access for any reason), (5) patients with severe acquired brain injury, (6) seizure disorder requiring anticonvulsant medication, (7) cirrhosis (Child's class C liver disease), (8) metastatic cancer or stage IV lymphoma with life expectancy <6 months, (9) routine elective cardiac surgery, (10) patients with primary admission diagnosis of burns (≥30% body surface area), (11) weight <50 kg or >200 kg, (12) pregnant patients or lactating with the intent to breastfeed, (13) previous randomization in this study, and (14) enrollment in a related ICU interventional study.

Eligible patients were randomized to a standard dose of intravenous (IV) glutamine supplementation (0.35 g/kg/d parenterally provided as 0.50 g/kg/d of the dipeptide alanyl-glutamine [Dipeptiven, Fresenius Kabi, Bad Homburg, Germany] and an additional 30 g/d of glutamine enterally, provided as 42.5 g alanyl-glutamine and glycine-glutamine dipeptides) or placebo. We used the Broca formula to estimate the "normal weight" (normal weight [kg] = height [cm] – 100 cm) for dosing the IV glutamine.[15] Using a 2 × 2 factorial design, patients were also randomized to receive antioxidants (500 μg of selenium parenterally and a specially prepared enteral antioxidant micronutrient solution [selenium, zinc, vitamins E and C, and β-carotene]) or placebo. For the parenteral study intervention, we used a saline placebo. For the enteral placebo, we used a readymade solution that contained small amounts of β-carotene (0.4 mg/100 mL) and vitamin C (<10 mg/100 mL) that was identical in volume, color, smell, and consistency to the active enteral intervention to maintain blinding. Thus, patients in each group received both an enteral and parenteral component to the intervention, preserving blinding of the clinical teams and research staff. Study supplements were started as soon as possible after randomization and administered continuously over 24 hours for a maximum of 28 days or until death or discharge from the ICU. Study supplements were administered separately from nutrition; all patients were fed according to the Canadian clinical practice guidelines for nutrition in the ICU.[16] Other management decisions were at the discretion of the clinical team.

Statistical Analysis

In accordance with the intention-to-treat principle, we included all patients in the group to which they were randomized. In contrast to our primary publication,[13] we have significantly expanded our analysis and compared each of the 3 active treatment arms (glutamine alone, antioxidants alone, and glutamine + antioxidants) with placebo. However, some pooled analyses comparing the 2 glutamine arms with the 2 nonglutamine arms and the 2 antioxidant arms with the 2 nonantioxidant arms are provided to allow comparison with our

primary study.[13] The Cox proportional hazards model, log-rank test, and Kaplan-Meier curve were used to assess the effect of treatment on mortality over the entire 6-month follow-up period. Actuarial life tables are provided to assess the timing of the effect of treatment on mortality.[17] Logistic regression was used to estimate the effect of treatment on 28-day mortality expressed as odds ratios and to produce Wald tests to test for treatment-by-subgroup interactions. We prespecified baseline characteristics that we believed were potential confounders or effect modifiers (see Suppl. Table S2) but were not necessarily identified in our initial published primary analysis plan. We hypothesized that older, sicker (as judged by higher Acute Physiology and Chronic Health Evaluation II [APACHE II] score, greater comorbidities, and greater degree of organ failures), and malnourished patients (as defined by body mass index [BMI]) were likely to be more deficient in these key substrates and benefit more from the therapeutic interventions. In addition, since most prior glutamine supplementation studies that were positive were conducted in surgical patients, mostly with cancer diagnoses, we explored whether there was a positive treatment effect in these subgroups. Finally, given differences in selenium concentration in the soil[18] and different practice patterns across countries, we explored differences in treatment effect by geographic region. These variables were used to define the subgroups and were all included as covariates for adjustment (control) in the multivariable Cox and logistic models (see Suppl. Table S1). We did not consider variables that occurred after randomization (such as amount of nutrition received) since this can lead to invalid inference.[19] However, we did explore the apparent subgroup effect of baseline renal dysfunction according to postbaseline dialysis use.

These exploratory analyses were considered post hoc and hypothesis generating. No adjustment was made for multiplicity of statistical tests. All analyses were performed using SAS version 9.3 (SAS, Inc, Cary, NC).

Results

As with the primary analysis, these post hoc analyses included all 1218 patients from the intention-to-treat analysis. The detailed flow diagram and patients' characteristics are reported by arm in our primary publication.[13] As reported in this primary publication, study patients on average received 45% of their prescribed protein and energy requirements. The total daily nitrogen intake in the glutamine treated group was 20.4 ± 7.0 g compared with 6.9 ± 4.4 g in the no-glutamine group.

The 6-month Kaplan-Meier survival curves for each of the 4 treatment arms are shown in Figure 1. The differences in survival curves tested across all 4 treatment arms were not statistically significant ($P = .11$). However, the 2 groups receiving glutamine had higher 6-month mortality rates compared with the 2 groups not receiving glutamine (unadjusted hazard ratio [HR], 1.24; 95% CI, 1.04–1.48; $P = .019$) and after controlling for all prespecified baseline covariates (adjusted HR, 1.23; 95% CI, 1.03–1.48; $P = .024$). There was no significant difference between patients receiving antioxidants vs not receiving antioxidants before or after controlling for all prespecified baseline covariates (data not shown). Of the 477 deaths reported over the 6-month follow-up period, 363 (76%) occurred within 28 days from randomization and 195 within the first 7 days (41%) (Figure 1 and

Table 1). The remainder of this analysis focuses on 28-day mortality, which was the prespecified primary outcome for this trial.

The 28-day mortality rates in the placebo, glutamine, antioxidant, and combination arms were 25%, 32%, 29%, and 33%, respectively. Compared with placebo, the unadjusted OR (95% CI) of 28-day mortality was 1.4 (1.0–2.0, $P = .063$), 1.2 (0.8–1.7, $P = .31$), and 1.4 (1.0–2.0, $P = .049$) in the glutamine, antioxidant, and combined arms, respectively. After adjusting for all prespecified baseline characteristics, the corresponding adjusted ORs remained virtually unchanged at 1.5 (1.0–2.1, $P = .051$), 1.2 (0.8–1.8, $P = .40$), and 1.4 (0.9–2.0, $P = .092$) (see Suppl. Table S1 for details of the multivariable model). According to our primary study, we also adjusted for all prespecified baseline covariates with respect to the pooled comparison of the 2 glutamine arms and nonglutamine arms. The adjusted OR (95% CI) of 28-day mortality still was 1.30 (1.00–1.67, $P = .049$). Comparing the pooled antioxidant arms with the pooled nonantioxidant arms, the adjusted OR (95% CI) was 1.05 (0.81–1.36, $P = .72$).

Table 2 provides overall and subgroup-specific comparisons of the 3 active treatment arms vs the placebo by region and baseline patient characteristics. The treatment effect was significantly different by the presence of baseline renal dysfunction (test of interaction $P = .035$). Among patients with renal dysfunction at baseline, the odds of 28-day mortality significantly increased in patients who received glutamine or antioxidant supplementation alone or in combination compared with placebo. There was no suggestion of a treatment effect, either positive or negative, in the 776 patients who did not have renal dysfunction at baseline (Table 2). There was no difference in treatment effect in patients with ≥2 organ failures at baseline. No other tests for treatment-by-subgroup interaction were statistically significant. These subgroup results remained virtually unchanged after controlling for all prespecified covariates (Suppl. Table S2). Table 3 estimates the unadjusted treatment effect according to baseline renal dysfunction and postbaseline dialysis. The negative effect of treatment among patients with baseline renal dysfunction appears to be attenuated by dialysis initiated after randomization in the monotherapy arms but not in the combination arm. Again, these results did not change substantively after adjustment for all prespecified covariates (Suppl. Table S3).

Discussion

In a recent large prospective randomized trial, early provision of high-dose glutamine or antioxidants did not improve clinical outcomes, and glutamine was associated with a trend toward an increase in mortality among critically ill patients with multiorgan failure.[13] To further explore these unexpected findings in specific subgroups of patients and to adjust our assessment of treatment effect by potentially confounding covariates, we conducted extensive post hoc analyses. Importantly, an increased mortality in the glutamine arms persisted after adjustment for all prespecified baseline covariates. In unadjusted subgroup analyses, the treatment effect of glutamine and antioxidant supplementation on 28-day mortality differed significantly with regard to the presence or absence of baseline renal dysfunction. This effect remained in the adjusted multivariable model. Both glutamine and antioxidants were associated with increased mortality in the presence of baseline renal

dysfunction while neither intervention was associated with harm or benefit in patients without baseline renal dysfunction. Too few patients were enrolled from Europe (n = 43) to allow reliable inferences regarding potential effect differences between Europe and North America. We observed that the majority of the deaths and all of the apparent harmful effect of supplementation occurred within 28 days of randomization.

Our preliminary work[20] and nascent pathophysiologic understanding led us to hypothesize that patients with multiorgan failure, including renal failure, would have greater depletion of these key nutrients and would thus benefit the most from a higher dose of these nutrients. Thus, we designed a trial to administer early, high doses of glutamine provided both enterally and parenterally and independent of nutrition. Unexpectedly, only 31% of patients presented with low baseline glutamine (<420 μmol/L) in a laboratory substudy of 66 patients as reported in the primary publication,[13] and most of these patients were not glutamine depleted. This observation was also seen in a recent publication examining baseline glutamine levels in critically ill patients, which demonstrated that a significant proportion of patients have normal to high plasma levels of glutamine and that there may be a U-shaped curve relating plasma levels to 6-month mortality.[21]

Our findings of increased risk associated with glutamine administration have persisted despite adjustment for random imbalances in baseline covariates. In both the pooled analysis the current adjusted analysis confirms an increased mortality at where both glutamine-receiving groups were combined or 28 days and an increase in 6-month mortality associated with whether considering the effect of glutamine alone vs placebo, glutamine administration. Thus, the potential risk of high-dose glutamine effect persisted independent of the number of organ failures. Moreover, our unadjusted subgroup analysis showed that the trend toward harm with glutamine existed among the 879 patients with ≤2 organ failures and among the 335 patients with 3 or 4 organ failures. Thus, the random imbalance in the number of organ failures across groups does not affect our main inference that high-dose glutamine supplementation was not beneficial, and perhaps harmful, in critically ill patients with multiorgan failure. Note that only 4 (0.3%) and 15 (1.2%) patients had 1 and 4 organ failures, respectively, so little information was lost by collapsing the number of organ failures into 2 groups.

We are unsure as to the exact mechanism of injury but note that the negative effect of glutamine appears to be attenuated in the presence of dialysis initiated after randomization, regardless of the presence or absence of baseline renal dysfunction. It is well recognized that dialysis, particularly continuous renal replacement therapy (CRRT), removes amino acids and small to middle molecules (< 20 kDa).[22] This would lead us to speculate that the harm may be caused by glutamine or a metabolite of glutamine that builds in renal insufficiency, which can be removed by dialysis, thereby preventing its accumulation. Support for this assertion comes from the observation that elevated urea levels (>50 mmol/L) occurred much more frequently in the glutamine-treated groups compared with the no-glutamine groups. In contrast, as shown in Table 3, when combined with antioxidants, the protective effect of dialysis on glutamine administration seems negated. We are unclear as to the mechanism of this effect or even if this effect is real given the lack of statistical precision and the risk of a type I error. Unfortunately, we cannot comment on the dose, duration, timing, and intensity

of dialysis and its relationship to outcome as these data were not available. We do not believe this harmful effect was due to protein per se given that patients only received on average less than 50% of their protein prescription.[13]

One observation in this post hoc analysis that was not apparent in our primary analysis was that antioxidants were also associated with increased 28-day mortality in the presence of renal dysfunction. An earlier study suggested that selenium supplementation may actually reverse oxidant stress-related kidney injury, leading to reduced need for renal replacement therapy.[23] However, these early observations were not confirmed in subsequent trials.[24,25] We are not aware of studies describing the pharmacokinetic or pharmacodynamic profile of selenium or antioxidants in critically ill patients in renal failure, although vitamin C may have negative effects in the presence of renal failure.[26] Given our observations and the lack of understanding of the pharmacology of selenium or antioxidants in renal failure, we suggest that selenium and antioxidants not be administered to critically ill patients with renal failure until its safety and efficacy can be confirmed in subsequent randomized trials, particularly in the selenium-replete North American population.[17]

Our greatly expanded post hoc analyses are informative not only for the significant relationship between treatment, renal dysfunction, and outcome but also for the lack of any subgroup associated with a positive treatment effect. A priori, we hypothesized that sicker patients (as judged by higher APACHE II scores, greater degree of acute organ dysfunction, or more comorbidity) would benefit the most from supplementation. Given that older patients were more likely to be micronutrient deficient when becoming critically ill, perhaps they would have benefited more from high-dose supplementation. Finally, most of the other randomized trials of glutamine supplementation occurred in the context of patients undergoing GI surgery for cancer and requiring parenteral nutrition[27]; thus, we expected to see greater treatment effect in patients undergoing surgery or having an admission diagnosis of cancer. While we conclude that high-dose supplemental glutamine (both intravenously and enterally) should not be provided to critically ill patients with multiorgan failure, we suggest caution in extrapolating our findings to the use of glutamine, in lower doses, to other ICU patients. The most recent, updated meta-analysis of IV glutamine trials in heterogeneous ICU patients still suggests a reduction in infection, length of stay, and mortality.[26]

The strength of the current analyses is the thorough examination of several different subgroups and treatment effects in a large randomized trial database adjusting for potential confounders. However, these analyses are post hoc, exploratory, and hypothesis generating and therefore should be interpreted with caution as there is a high likelihood of both type I and type II error.

Conclusions

We confirmed that early provision of supplemental high-dose glutamine administered separate from artificial nutrition was not beneficial and appears to be associated with increased mortality in critically ill patients with multiorgan failure. We suggest that it should not be administered to these patients in this context. Much of the increased risk of high-dose

glutamine administration appears to be related to the presence of renal dysfunction upon study enrollment. Antioxidant supplementation was also associated with increased mortality only among patients with baseline renal dysfunction. Until further safety and efficacy data are available from adequately powered randomized trials, we suggest that glutamine and antioxidants not be administered to critically ill patients with multiorgan failure, particularly those with concomitant acute renal failure.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments

Financial disclosure: Funded by the Canadian Institutes of Health Research; ClinicalTrials.gov number, . Fresenius Kabi provided the glutamine supplements and an unrestricted grant-in-aid. Biosyn provided the intravenous selenium to all participating European sites.

Conflicts of interest: D.K.H. has received research grants and a speaker honorarium from Fresenius Kabi, Baxter, and Biosyn. G.E. has received speaker's honoraria from Abbott, B. Braun, and Fresenius Kabi. D.C. is a Canada Research Chair of the Canadian Institutes of Health Research. P.E.W. has received research grants and a speaker honorarium from Fresenius Kabi, Baxter, and Abbott. M.M.B. received a speaker honorarium from Fresenius Kabi, Nestlé, and Baxter.

References

1. Motoyama T, Okamoto K, Kukita I, Hamaguchi M, Kinoshita Y, Ogawa H. Possible role of increased oxidant stress in multiple organ failure after systemic inflammatory response syndrome. Crit Care Med. 2003;31: 1048–1052. [PubMed: 12682471]
2. Alonso de Vega JM, Díaz J, Serrano E, Carbonell LF. Oxidative stress in critically ill patients with systemic inflammatory response syndrome. Crit Care Med. 2002;30:1782–1786. [PubMed: 12163793]
3. Goode HF, Cowley HC, Walker BE, Howdle PD, Webster NR. Decreased antioxidant status and increased lipid peroxidation in patients with septic shock and secondary organ dysfunction. Crit Care Med. 1995;23:646–651. [PubMed: 7712754]
4. Brealey D, Brand M, Hargreaves I, et al. Association between mitochondrial dysfunction and severity and outcome of septic shock. Lancet. 2002;360:219–223. [PubMed: 12133657]
5. Goodyear-Bruch C, Pierce JD. Oxidative stress in critically ill patients. Am J Crit Care. 2002;11:543–561. [PubMed: 12425405]
6. Planas M, Schwartz S, Arbos MA, Farriol M. Plasma glutamine levels in septic patients. JPEN J Parenter Enteral Nutr. 1993;17:299–300. [PubMed: 8505843]
7. Borrelli E, Roux-Lombard P, Grau GE, et al. Plasma concentration of cytokines, their soluble receptors, and antioxidant vitamins can predict the development of multiple organ failure in patients at risk. Crit Care Med. 1996;24:392–397. [PubMed: 8625625]
8. Oehler R, Pusch E, Dungel P, et al. Glutamine depletion impairs cellular stress response in human leucocytes. Br J Nutr. 2002;87:S17–S21. [PubMed: 11895151]
9. Roth E, Funovics J, Muhlbacher F, Schemper M, Mauritz W, Sporn P. Metabolic disorders in severe abdominal sepsis: glutamine deficiency in skeletal muscle. Clin Nutr. 1982;1:25–41. [PubMed: 16829366]
10. Oudemans-van Straaten HM, Bosman RJ, Treskes M, van der Spoel JHI, Zandstra DF. Plasma glutamine depletion and patient outcome in acute ICU admissions. Intensive Care Med. 2001;27:84–90. [PubMed: 11280678]
11. Novak F, Heyland DK, Avenell A, Novak F, Drover J, Su X. Glutamine supplementation in serious illness: a systematic review of the evidence. Crit Care Med. 2002;30:2022–2029. [PubMed: 12352035]

12. Heyland DK, Dhaliwal R, Drover JW, Gramlich L, Dodek P, for the Guidelines Committee. Canadian clinical practice guidelines for nutrition support in the adult critically ill patient. JPEN J Parenter Enteral Nutr. 2003;27:355–373. See www.criticalnutrition.com/clinicalpracticeguidelines for the most current version of the glutamine meta-analysis Accessed August 1, 2013. [PubMed: 12971736]
13. Heyland DK, Muscedere J, Wischmeyer P, et al., for the Canadian Critical Care Trials Group. A randomized trial of glutamine and antioxidants in critically ill patients. N Engl J Med. 2013;368(16):1489–1497. [PubMed: 23594003]
14. Heyland DK, Dhaliwal R, Day A, et al., for the Canadian Critical Care Trials Group. REducing Deaths due to OXidative Stress: The REDOXS© Study: rationale and study design for a randomized trial of glutamine and antioxidant supplementation in critically ill patients. Proc Nutr Soc. 2006;65(3):250–263. [PubMed: 16923310]
15. Hamwi GJ. Therapy: changing dietary concepts In: Danowski TS, ed. Diabetes Mellitus: Diagnosis and Treatment. Vol 1 New York, NY: American Diabetes Association; 1964:73–78.
16. Heyland DK, Dhaliwal R, Drover JW, Gramlich L, Dodek P, for the Guidelines Committee. Canadian clinical practice guidelines for nutrition support in the adult critically ill patient. JPEN J Parenter Enteral Nutr. 2003;27:355–373. [PubMed: 12971736]
17. Kalbfleisch JD, Prentice RL. The Statistical Analysis of Failure Time Data. New York, NY: John Wiley; 1980.
18. Johnson CC, Fordyce FM, Rayman MP. Symposium on 'Geographical and geological influences on nutrition': factors controlling the distribution of selenium in the environment and their impact on health and nutrition. Proc Nutr Soc. 2010;69:119–132. [PubMed: 19968907]
19. Sun X, Briel M, Walter SD, Guyatt GH. Is a subgroup effect believable? Updating criteria to evaluate the credibility of subgroup analyses. BMJ. 2010;340:c117. [PubMed: 20354011]
20. Heyland DK, Dhaliwal R, Day A, Drover J, Cote H, Wischmeyer P. Optimizing the dose of glutamine dipeptides and antioxidants in critically ill patients: a phase I dose finding study. JPEN J Parenter Enteral Nutr. 2007;31(2):109–118. [PubMed: 17308251]
21. Rodas PC, Rooyackers O, Hebert C, Norberg A, Wernerman J. Glutamine and glutathione at ICU admission in relation to outcome. Clin Sci. 2012;122:591–597. [PubMed: 22248298]
22. Brunet S, Leblanc M, Geadah D, Parent D, Courteau S, Cardinal J. Diffusive and convective solute clearances during continuous renal replacement therapy at various dialysate and ultrafiltration flow rates. Am J Kidney Dis. 1999;34:486–492. [PubMed: 10469859]
23. Angstwurm MW, Schottdorf J, Schopohl J, Gaertner R. Selenium replacement in patients with severe systemic inflammatory response syndrome improves clinical outcome. Crit Care Med. 1999;27:1807–1813. [PubMed: 10507602]
24. Angstwurm MW, Engelmann L, Zimmermann T, et al. Selenium in Intensive Care (SIC): results of a prospective randomized, placebo-controlled, multiple-center study in patients with severe systemic inflammatory response syndrome, sepsis, and septic shock. Crit Care Med. 2007;35(1): 118–126. [PubMed: 17095947]
25. Mishra V, Baines M, Perry SE, et al. Effect of selenium supplementation on biochemical markers and outcome in critically ill patients. Clin Nutr. 2007;26(1):41–50. [PubMed: 17174015]
26. Mashour S, Turner JF Jr, Merrell R. Acute renal failure, oxalosis, and vitamin C supplementation: a case report and review of the literature. Chest. 2000;118:561–563. [PubMed: 10936161]
27. Bollhalder L, Pfeil AM, Tomonaga Y, Schwenkglenks M. A systematic literature review and meta-analysis of randomized clinical trials of parenteral glutamine supplementation. Clin Nutr. 2013;32:213–223. [PubMed: 23196117]

Clinical Relevancy Statement

In this post hoc analysis, we demonstrate that glutamine administration, compared with placebo, was not associated with any clinical benefit and may be associated with increased mortality. The negative treatment effect of glutamine persisted after adjustment for baseline covariates. The greatest signal of harm for both glutamine and antioxidants occurred in patients with renal dysfunction at baseline. We could not identify any subgroup of patients with multiorgan failure who benefited from the high-dose glutamine and antioxidants.

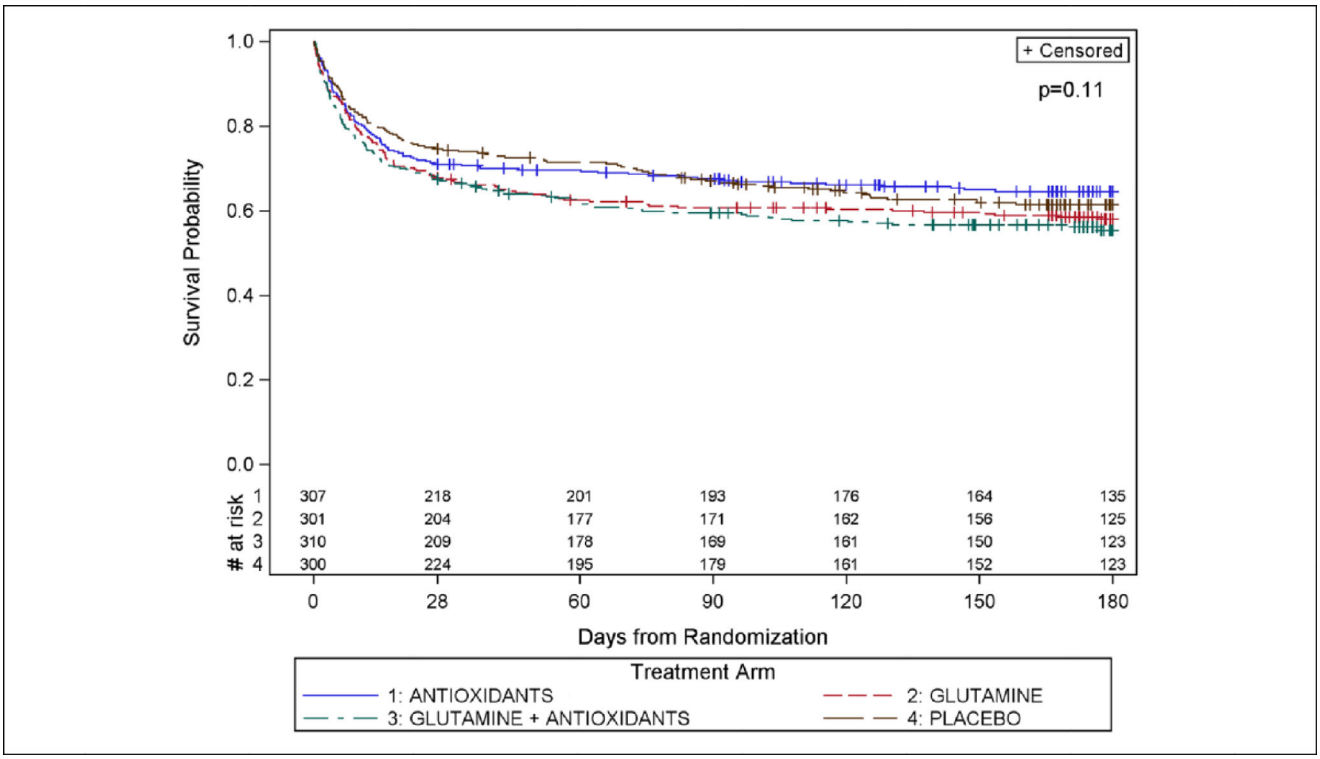

Figure 1.

Kaplan-Meier survival curve by treatment arm. Kaplan-Meier survival curves of the 4 different treatment groups: (1) antioxidants, (2) glutamine, (3) glutamine and antioxidants, and (4) placebo. In total, 128 (10.5%) of patients were lost to follow-up prior to the final 6-month assessment. The large amount of censoring after day 150 is not loss to follow-up but rather patients who had their final assessment within 4 weeks prior to day 180, as allowed by the study protocol.

Table 1.

Timing of Mortality Assessed by Actuarial Life Table.

Period	Deaths	Censored	Effective n	% Mortality (95% CI)
First 7 days				
Placebo	42	0	300	14 (10–18)
Glutamine	46	0	301	15 (11–19)
Antioxidants	45	0	307	15 (11–19)
GLN + AOX	62	0	310	20 (16–24)
Total	195	0	1218	16 (14–18)
Days 8–14				
Placebo	18	0	258	7 (4–10)
Glutamine	26	0	255	10 (6–14)
Antioxidants	24	0	262	9 (6–13)
GLN + AOX	23	0	248	9 (6–13)
Total	91	0	1023	9 (7–11)
Days 15–28				
Placebo	16	0	240	7 (4–10)
Glutamine	25	0	229	11 (7–15)
Antioxidants	20	0	238	8 (5–12)
GLN + AOX	16	0	225	7 (4–10)
Total	77	0	932	8 (6–10)
First 28 days				
Placebo	76	0	300	25 (20–30)
Glutamine	97	0	301	32 (27–38)
Antioxidants	89	0	307	29 (24–34)
GLN + AOX	101	0	310	33 (27–38)
Total	363	0	1218	30 (27–32)
Days 29–90				
Placebo	21	24	212	10 (6–14)
Glutamine	20	13	198	10 (6–14)
Antioxidants	10	15	211	5 (2–8)

JPEN J Parenter Enteral Nutr. Author manuscript; available in PMC 2019 October 18.

Period	Deaths	Censored	Effective n	% Mortality (95% CI)
GLN + AOX	23	17	201	11 (7–16)
Total	74	69	821	9 (7–11)
Days 90–180				
Placebo	14	42	158	9 (4–13)
Glutamine	7	39	152	5 (1–8)
Antioxidants	8	50	168	5 (2–8)
GLN + AOX	11	35	152	7 (3–11)
Total	40	166	629	6 (4–8)

AOX, antioxidants; CI, confidence interval; GLN, glutamine. In total, 128 (10.5%) of patients were lost to follow-up prior to the final assessment. However, as allowed by the study protocol, an additional 107 (8.8%) had their final assessment within 4 weeks prior to 180 days and were thus censored in the final month of follow-up. There were 44 patients who had improved and were discharged from hospital within 28 days but were not reached for further follow-up. In light of our follow-up procedures, we strongly believe these patients survived at least 28 days and thus we censored these patients at day 28. A sensitivity analysis has confirmed that excluding these 44 patients does not substantively change any results. In accordance with the life table method, the effective n is the number of patients alive at the beginning of the interval minus half the number censored during that interval, and the mortality rate is the number of deaths divided by the effective sample size. Effective n totals may be 1 lower than the sum of the 4 groups due to rounding up of half units.

Table 2.

Unadjusted Subgroup-Specific Treatment Effect.

Subgroup	Deaths, No./Total No. (%)	OR (95% CI) vs Placebo			*P* Value[a]
		GLN Alone	AOX Alone	GLN + AOX	
Overall	363/1218 (30)	1.40 (0.98–2.00)	1.20 (0.84–1.72)	**1.42 (1.00–2.03)**	.18
Study setting: region					.37
Canada	303/1044 (29)	1.41 (0.96–2.07)	1.14 (0.77–1.67)	1.29 (0.88–1.89)	
United States	44/131 (34)	1.56 (0.51–4.81)	1.43 (0.47–4.38)	**3.43 (1.17–10.07)**	
Europe	16/43 (37)	0.86 (0.12–5.9)	2.40 (0.39–14.88)	0.89 (0.14–5.48)	
Baseline patient characteristics					
Age, y					.18
<55	62/303 (20)	1.77 (0.79–3.96)	1.07 (0.43–2.65)	1.98 (0.87–4.54)	
55–64	69/290 (24)	0.94 (0.44–2.04)	0.87 (0.41–1.82)	1.26 (0.59–2.66)	
65–74	107/333 (32)	1.56 (0.81–3.01)	0.76 (0.39–1.49)	1.21 (0.64–2.28)	
≥75	125/292 (43)	1.53 (0.76–3.08)	**2.49 (1.23–5.03)**	1.35 (0.68–2.70)	
BMI					.39
<25	128/374 (34)	1.10 (0.60–2.00)	0.71 (0.39–1.30)	0.97 (0.53–1.79)	
25–34.9	175/610 (29)	1.57 (0.93–2.66)	1.64 (0.97–2.77)	1.57 (0.93–2.64)	
≥35	60/234 (26)	1.69 (0.71–4.00)	1.35 (0.55–3.32)	**2.36 (1.04–5.36)**	
Admission category					.52
Surgical	59/255 (23)	2.16 (0.91–5.15)	1.94 (0.78–4.82)	1.58 (0.67–3.76)	
Medical	304/963 (32)	1.28 (0.87–1.89)	1.08 (0.73–1.60)	1.43 (0.97–2.12)	
APACHE II score					.70
<21	48/256 (19)	2.20 (0.86–5.60)	1.87 (0.72–4.86)	2.18 (0.82–5.79)	
21–26	92/328 (28)	1.17 (0.55–2.49)	1.61 (0.79–3.26)	1.92 (0.96–3.82)	
26–31	89/310 (29)	1.41 (0.70–2.82)	0.88 (0.44–1.75)	1.07 (0.53–2.16)	
>31	133/323 (41)	1.16 (0.62–2.18)	1.00 (0.51–1.97)	0.98 (0.52–1.84)	
Charlson comorbidity index					.92
0–1	154/662 (23)	1.52 (0.90–2.56)	1.28 (0.75–2.20)	1.61 (0.94–2.73)	
>1	209/556 (38)	1.41 (0.85–2.33)	1.14 (0.70–1.87)	1.26 (0.78–2.03)	
Patients with cancer					.74

Subgroup	Deaths, No./Total No. (%)	OR (95% CI) vs Placebo			*P* Value[a]
		GLN Alone	AOX Alone	GLN + AOX	
No	297/1048 (28)	**1.48 (1.01–2.18)**	1.15 (0.77–1.71)	1.42 (0.97–2.10)	
Yes	66/170 (39)	1.05 (0.41–2.73)	1.43 (0.60–3.40)	1.38 (0.58–3.27)	
Etiology of shock					.71
Cardiogenic	74/240 (31)	1.24 (0.56–2.79)	1.62 (0.75–3.51)	**2.19 (1.03–4.67)**	
Septic	256/826 (31)	1.43 (0.93–2.19)	1.06 (0.69–1.63)	1.21 (0.79–1.86)	
Other/unknown/none	33/152 (22)	1.45 (0.46–4.57)	1.45 (0.43–4.86)	1.83 (0.60–5.78)	
Vasopressors, mcg/min					.37
<15	162/595 (27)	1.58 (0.92–2.70)	1.66 (0.97–2.84)	1.50 (0.87–2.58)	
≥15	201/623 (32)	1.32 (0.82–2.13)	0.92 (0.57–1.51)	1.39 (0.87–2.22)	
No. of organ failures on presentation					
2	238/879 (27)	1.27 (0.83–1.95)	1.18 (0.77–1.79)	1.16 (0.76–1.78)	
>2	125/335 (37)	1.68 (0.88–3.20)	1.33 (0.66–2.67)	**2.06 (1.09–3.90)**	
Renal dysfunction					**.035**
No	216/776 (28)	0.93 (0.59–1.46)	0.90 (0.58–1.40)	1.14 (0.74–1.77)	
Yes	147/442 (33)	**2.75 (1.50–5.03)**	**2.16 (1.15–4.07)**	**2.15 (1.17–3.94)**	

AOX, antioxidants; APACHE II, Acute Physiology and Chronic Health Evaluation II; BMI, body mass index; CI, confidence interval; GLN, glutamine; OR, odds ratio. Odds ratios in bold indicate treatment arm had significantly higher 28-day mortality than placebo at $P < .05$.

[a] *P* values test for interaction (effect modification) between subgroups and treatment. Significant *P* values indicate that the effect of supplementation was statistically significantly different between subgroups.

JPEN J Parenter Enteral Nutr. Author manuscript; available in PMC 2019 October 18.

Table 3.

Unadjusted Examination of Treatment Effect on 28-Day Mortality by Baseline Renal Dysfunction and Postbaseline Dialysis.

Subgroup			OR (95% CI) vs Placebo Arm		
Renal Dysfunction	**Postbaseline Dialysis**	**Deaths, No./Total No. (%)**	**GLN Alone**	**AOX Alone**	**GLN + AOX**
No	No	158/634 (25%)	1.15 (0.68–1.93)	1.04 (0.63–1.73)	1.30 (0.77–2.17)
No	Yes	58/142 (41%)	0.44 (0.17–1.19)	0.48 (0.18–1.28)	0.63 (0.25–1.57)
Yes	No	76/240 (32%)	**3.91 (1.71–8.96)**	**3.39 (1.41–8.17)**	1.63 (0.71–3.76)
Yes	Yes	71/202 (35%)	1.82 (0.75–4.42)	1.38 (0.55–3.48)	**3.07 (1.24–7.59)**

AOX, antioxidants; CI, confidence interval; GLN, glutamine; OR, odds ratio. Bold numbers indicate treatment arm had significantly higher 28-day mortality than placebo at $P < .05$. Test for interaction (effect modification) between treatment and 4 subgroups, $P = .011$.