Adverse Gastrointestinal Effects of Arginine and Related Amino Acids1,2

George K. Grimble*

Department of Food Biosciences, University of Reading, Whiteknights, Reading RG6 6AP, UK

Abstract

Oral supplements of arginine and citrulline increase local nitric oxide (NO) production in the small intestine and this may be harmful under certain circumstances. Gastrointestinal toxicity was therefore reviewed with respect to the intestinal physiology of arginine, citrulline, ornithine, and cysteine (which shares the same transporter) and the many clinical trials of supplements of the dibasic amino acids or N-acetylcysteine (NAC). The human intestinal dibasic amino acid transport system has high affinity and low capacity. L-Arginine (but not lysine, ornithine, or D-arginine) induces water and electrolyte secretion that is mediated by NO, which acts as an absorbagogue at low levels and as a secretagogue at high levels. The action of many laxatives is NO mediated and there are reports of diarrhea following oral administration of arginine or ornithine. The clinical data cover a wide span of arginine intakes from 3 g/d to >100 g/d, but the standard of reporting adverse effects (e.g. nausea, vomiting, and diarrhea) was variable. Single doses of 3–6 g rarely provoked side effects and healthy athletes appeared to be more susceptible than diabetic patients to gastrointestinal symptoms at individual doses >9 g. This may relate to an effect of disease on gastrointestinal motility and pharmacokinetics. Most side effects of arginine and NAC occurred at single doses of >9 g in adults (>140 mg/kg) when part of a daily regime of ~30 g/d (>174 mmol/d). In the case of arginine, this compares with the laxative threshold of the nonabsorbed disaccharide alcohol, lactitol (74 g or 194 mmol). Adverse effects seemed dependent on the dosage regime and disappeared if divided doses were ingested (unlike lactitol). Large single doses of poorly absorbed amino acids seem to provoke diarrhea. More research is needed to refine dosage strategies that reduce this phenomenon. It is suggested that dipeptide forms of arginine may meet this criterion. J. Nutr. 137: 1693S–1701S, 2007.

This article reviews the possible gastrointestinal toxicities that might arise from ingesting supplements of arginine and related compounds. This relation can be defined in 2 ways. First, arginine, the other dibasic amino acids, ornithine, and cysteine all share a common intestinal transport system. In addition, arginine is metabolically related to ornithine, citrulline, homocitrulline, and the polyamines. None of these compounds is found in the diet in significant quantities, but they have all been used as therapeutic supplements. Gastrointestinal toxicity might therefore arise through direct effects on the gut or through production of a metabolic product such as nitric oxide (NO)3. Although glutamine and glutamate are intimately related to intestinal arginine metabolism, they will not be discussed and the reader is directed elsewhere (1,2).

A brief history of therapeutic oral use of arginine and related amino acids

Arginine has been widely used because of its pivotal role in synthesis of urea, NO, and creatine (3). This makes arginine (and related amino acids) useful in treating insufficiencies of the urea cycle [e.g. hyperammonemia in liver failure (4)], insufficiencies in NO production (5–9), and inborn errors of creatine synthesis involving defects in arginine:glycine amidinotransferase and guanidinoacetate methyltransferase (10).

Supplements of arginine, ornithine, or citrulline are commonly ingested as the Cl− salt or as the salt of other anions such as α-ketoglutaryl [i.e. ornithine α-ketoglutarate (OKG)], aspartate, pyroglutamate, or malate. The organic anion may exert synergistic effects, as appears to be the case with OKG (11).

1 Published in a supplement to The Journal of Nutrition. Presented at the conference “The Sixth Workshop on the Assessment of Adequate and Safe Intake of Dietary Amino Acids” held November 6–7, 2006 in Budapest. The conference was sponsored by the International Council on Amino Acid Science (ICAAS). The organizing committee for the workshop was David H. Baker, Dennis M. Bier, Luc A. Cynober, Yuzo Hayashi, Motoni Kadowaki, Sidney M. Morris, Jr., and Andrew G. Renwick. The Guest Editors for the supplement were David H. Baker, Dennis M. Bier, Luc A. Cynober, Motoni Kadowaki, Sidney M. Morris, Jr., and Andrew G. Renwick. Disclosures: all Editors and members of the organizing committee received travel support from ICAAS to attend the workshop and an honorarium for organizing the meeting.

2 Author disclosures: G. K. Grimble’s travel expenses to attend the meeting paid by ICAAS.

3 Abbreviations used: ASC, small aliphatic amino acid transporter; β6 AT, dibasic amino acid transporter; GSH, glutathione reduced; NAC, N-acetylcysteine; NO, nitric oxide; NOS, nitric oxide synthase; OKG, ornithine α-ketoglutarate; PEPT1, di- and tri-peptide transporter.
Many of the studies cited in this article use the Cl⁻ or the basic amino acid and this may, itself, be deleterious, because arginine hydrochloride contains 4.8 mequivalents Cl⁻/g and may provoke a hyperchloremic acidosis if taken acutely in excess (12).

Arginine and ornithine were first used to treat hyperammonemia by providing a sufficient supply of urea cycle intermediates. This was a serendipitous finding that arose from the switch from protein hydrolysates for i.v. nutrition to the potentially more flexible l-amino acid mixtures in the mid 1980s. The hydrolysates were generally successful and safe (13,14), but the l-amino acid mixtures caused problems. Sufficient arginine should be added to prevent hyperammonemia, which could be fatal (15). It was therefore a short step to suggest that arginine and ornithine supplements could successfully treat hepatic encephalopathy, of which 1 causal factor is hyperammonemia. In this therapy, arginine has been administered as the chloride salt (16) or as the aspartate salt, whereas ornithine has been administered as the hydrochloride salt (17), or the α-ketoglutarate salt (18). The amounts of l-arginine that were initially given were high [100–150 g/d; (16)] and Molimard (18,19) reported that i.v. infusions of 50 g of OKG corrected blood biochemistry and improved electroencephalogram scores.

The ability of arginine and ornithine to stimulate the pancreatic and pituitary axis has been used to alleviate growth retardation in children (20) and to promote growth hormone release in endurance athletes or body builders, with the hope of also relieving the hyperammonemia of intense exercise (21–23). Additionally, oral arginine or OKG have been used to promote wound healing in elderly people with pressure sores (6,24) and in scar healing of surgical scars (25,26) or burn scars (26,27).

However, the most intense research has been performed on the ability of oral arginine supplements to increase NO production. This has been investigated in progressive renal failure (28), atherosclerosis (29), pulmonary hypertension after congenital heart surgery (30), hypertension (31), pressure sores (24), erectile dysfunction (9), and necrotizing enterocolitis in premature infants (32).

Cysteine has been widely used since the early 1960s as a mucolytic agent in treatment of pulmonary symptoms of asthma and chronic obstructive pulmonary diseases such as cystic fibrosis (33). It is also thought to be the limiting amino acid in glutathione synthesis (34) and oral supplementation is an effective means to augment glutathione pools that have been depleted by oxidative damage (35,36). Paracetamol (acetaminophen) overdose causes depletion of liver glutathione pools and subsequent liver failure and supplemental N-acetylcysteine (NAC) is effective if given orally or i.v. within a few hours of overdose (37).

General considerations on gastrointestinal toxicity

Because the gastrointestinal tract experiences the first effects of orally administered arginine, its function might be a sensitive barometer of toxicity. Oral ingestion of arginine should, theoretically, increase local production of NO via intestinal constitutive NO synthase (NOS) or NOS-1. NO is involved in nearly every aspect of intestinal function and controls water and electrolyte transport (38), vasomotor function and, hence, vascular perfusion (39,40), motility at all levels of the intestine (40–42), and modulation of the inflammatory response of the intestine (43). In addition, excess NO production may be deleterious if intestinal disease is present. For example, in patients with cirrhosis and portal hypertension, increased local NO production may exacerbate vasodilatation (44). Likewise, in patients with collagenous colitis, luminal arginine perfusion exacerbated the preexisting secretory state (45).

A second effect might arise if large oral amino acid supplements bypass the normal mechanisms that regulate gastric emptying so that a large and damaging hypertonic load would be presented to the small bowel mucosa.

How can intestinal toxicity be defined?

Diarrhea is the simplest gauge of intestinal ill health, because it reflects an impairment of the ability of the intestine to conserve water and electrolytes, which is as important as assimilation of nutrients. Phillips (46) proposed that diarrhea was a hallmark of intestinal failure for this reason and other symptoms such as flatus, distension, pain, and borborygmi add further refinement because the definition of “diarrhea” is problematic (47). We and others have used this simple approach to determine oral tolerance to nonabsorbed but fermentable sugars, sugar alcohols, and fructose oligomers (48–51), which sweep water and electrolytes into the large bowel and, at a high enough dose, can temporarily overwhelm its salvage capacity, causing osmotic diarrhea (46,52).

Do arginine, ornithine, and citrulline cause diarrhea?

Using these criteria, the literature was searched for all trials in which oral arginine, ornithine, and citrulline had been given to healthy volunteers or patients (Table 1). The writer was struck both by the wide indications for these compounds and by the lack of consensus on dosage regimes (Table 1). Reporting of adverse effects was often inconsistent or nonexistent and, even where placebo and treatment had been compared, the incidence of gastrointestinal side effects varied greatly (Table 1). In some studies, authors commented on tolerance to the oral supplement; several studies reported no side effects even when large doses were administered. Side effects did not appear to be related to the concentration of plasma l-arginine that was achieved and would provide the substrate for systemic NO synthesis.

Adverse effects were observed [e.g. (22,55)] at single doses >10 g and comprised nausea or vomiting or diarrhea. In other studies where similar dosages were given [e.g. (71,73,74)] few, if any, side effects were reported.

The differences between tolerance in healthy people and patients are noteworthy and could be explained in several ways. First, the dosage regime used might be important and smaller divided doses might lead to fewer side effects. Second, the preexisting proabsorptive or prosecretory state of the intestine may be important and, last, the difference in insulin resistance between healthy young subjects and patients may markedly alter gastric and small bowel motility, thus mitigating secretory effects.

We observed the importance of these factors several years ago when investigating the metabolic effects of oral OKG administered by bolus or by continuous infusion during a 12-h enteral diet infusion in healthy volunteers (75). During some of the infusions, subjects experienced explosive diarrhea ~120–180 min after the start of the diet infusion. As we had already observed that l-ornithine tended to reduce water absorption at higher perfused concentrations (76), it seemed that ornithine was the causative factor per se. Certainly, it occurred most frequently following the 20-g bolus, not the 10-g bolus or during the 10-g infusion. However, subsequent studies showed that nasogastric enteral feeding per se caused diarrhea in healthy volunteers, because it establishes a secretory state in the fasted ascending colon (77,78). Thus, in the OKG study, the combination of a secretory state and the extra stimulus provided by a 20-g bolus of OKG may have overwhelmed the reserve absorptive capacity of the colon (52). This example makes it clear that there are several factors that affect gastrointestinal toxicity.
TABLE 1  Gastroenterological side effects resulting from ingestion of L-arginine, L-ornithine, or L-citrulline

<table>
<thead>
<tr>
<th>Study</th>
<th>Test amino acid</th>
<th>Dosage</th>
<th>Plasma concentration achieved, μmol/L</th>
<th>Side effects</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Healthy volunteers: higher doses</td>
<td>L-Arginine</td>
<td>5, 9, and 13 g as single doses</td>
<td>Arginine not measured but GH concentration increased at 5- and 9-g but not 13-g doses</td>
<td>7/8 subjects had gastrointestinal distress on 13-g dose</td>
<td>(53)</td>
</tr>
<tr>
<td>Effects on growth hormone secretion (n = 8, 18–33 y)</td>
<td>L-Arginine</td>
<td>0.75 g or 3 g hourly for 10 h (total 30 g)</td>
<td>Arginine* 471 ± 87 vs. 188 ± 29 (alanine control)</td>
<td>None reported; 30% of volunteers failed to increase plasma citrulline</td>
<td>(54)</td>
</tr>
<tr>
<td>Effects on insulin secretion (n = 26, 43 ± 3 y)</td>
<td>L-Arginine</td>
<td>3, 9, 21, and 30 g as divided doses (3/d)</td>
<td>9 g arginine: 169 ± 22 vs. 101 ± 4.1 (zero control) 21 g arginine: 164 ± 17 vs. 101 ± 4.1 (zero control)</td>
<td>30 g: 9/10 diarrhea 21 g: 4/12 diarrhea 3 and 9 g/d: no diarrhea</td>
<td>(55)</td>
</tr>
<tr>
<td>Effect on anal pressure and blood flow (n = 8, 24–52 y)</td>
<td>L-Arginine</td>
<td>15 g/d for 7 d as divided doses (3/d)</td>
<td>Arginine: 284 ± 44 (d 3), 157 ± 19.6 (d 7) vs. 107 ± 8.6 (d 0)</td>
<td>None reported</td>
<td>(40)</td>
</tr>
<tr>
<td>Effect on immune responsiveness (n = 21)</td>
<td>L-Arginine</td>
<td>30 g/d</td>
<td>“Minimal side effects…such as nausea or diarrhea,… responded to lowering the dose ingested at 1 time.”</td>
<td>None observed</td>
<td>(56)</td>
</tr>
<tr>
<td>Comparison of oral and intravenous L-arginine loading (n = 10, 25–52 y)</td>
<td>L-Arginine</td>
<td>10 g as single oral dose</td>
<td>Arginine (peak): 287 ± 77 Baseline: 80</td>
<td>None observed</td>
<td>(57)</td>
</tr>
<tr>
<td>Effect of mixed amino acids on collagen growth using subcutaneous collection tube (n = 35)</td>
<td>β-Hydroxy-β-methylbutyrate (3 g/d) + glutamine (14 g/d) + arginine (14 g/d)</td>
<td>14 g arginine in 2-g doses vs. positive amino acid control</td>
<td>Arginine (d 14): 162 ± 14 vs. 72 ± 3 (control)</td>
<td>None reported</td>
<td>(58)</td>
</tr>
<tr>
<td>Effect of L-arginine on plasma lipid status in normal (n = 12) or hypercholesterolemic (n = 27) young men</td>
<td>L-Arginine</td>
<td>21 g arginine in 3 7-g doses vs. placebo powder</td>
<td>Healthy subjects: 303 ± 36 vs. 128 ± 12 Hypercholesterolemic men: 231 ± 125 vs. 115 ± 103</td>
<td>None reported</td>
<td>(59,60)</td>
</tr>
<tr>
<td>Effect of L-arginine on substrate metabolism in endurance athletes</td>
<td>L-Arginine L-Aspartate</td>
<td>5.7 g/d or 2.8 g/d arginine</td>
<td>N/A</td>
<td>None reported</td>
<td>(61)</td>
</tr>
<tr>
<td>Effect of L-arginine on endurance in long-distance runners</td>
<td>L-Arginine L-Aspartate</td>
<td>5.7 g/d as 2 doses</td>
<td>120 vs. 30 (placebo group)</td>
<td>None reported</td>
<td>(23)</td>
</tr>
<tr>
<td>Effect of ornithine on growth hormone in bodybuilders</td>
<td>L-Ornithine hydrochloride</td>
<td>40, 100, 170 mg/kg (i.e. 3–4 g, 7.4–10.4 g, and 12.6–17.7 g, depending on body weight)</td>
<td>48, 58, and 85 for the 3 doses</td>
<td>None observed</td>
<td>(30)</td>
</tr>
<tr>
<td>Healthy volunteers: lower doses</td>
<td>L-Arginine, L-ornithine, and L-lysine</td>
<td>2 g/d as 2 1-g doses</td>
<td>Not measured. No effect on plasma GH</td>
<td>None reported</td>
<td>(62)</td>
</tr>
<tr>
<td>Effects on growth hormone secretion (n = 11, 19–35 y)</td>
<td>L-Ornithine-L-aspartate</td>
<td>3 g Omithine 189 ± 61 vs. 50 (at start)</td>
<td>Omithine 189 ± 61 vs. 50 (at start)</td>
<td>None reported</td>
<td>(63)</td>
</tr>
<tr>
<td>Bioavailability and pharmacokinetics (n = 12, 24.1 ± 2.5 y)</td>
<td>L-Citrulline</td>
<td>3.8 g·m⁻²·1·d⁻¹</td>
<td>12 h postoperative Arginine 36 ± 24 vs. 23 ± 13 (placebo) Citrulline 37 (18–83) vs. 20 (15–29) P &lt; 0.02</td>
<td>None reported</td>
<td>(30)</td>
</tr>
<tr>
<td>Patients</td>
<td>L-Citrulline</td>
<td>3.8 g·m⁻²·1·d⁻¹</td>
<td>12 h postoperative Arginine 36 ± 24 vs. 23 ± 13 (placebo) Citrulline 37 (18–83) vs. 20 (15–29) P &lt; 0.02</td>
<td>None reported</td>
<td>(30)</td>
</tr>
<tr>
<td>Acute myocardial infarction (n = 792)</td>
<td>L-Arginine vs. placebo</td>
<td>3 g/d as 3 doses</td>
<td>Not reported</td>
<td>9/385 vs. 10/387 (placebo)</td>
<td>(30,64)</td>
</tr>
<tr>
<td>Stable coronary artery disease (n = 31)</td>
<td>L-Arginine vs. vitamin C (500 mg) crossover</td>
<td>10 g/d (twice daily)</td>
<td>Not reported</td>
<td>Arginine: bowel disturbance 5/31 vitamin C: 2/31</td>
<td>(65)</td>
</tr>
<tr>
<td>Heart failure (n = 15)</td>
<td>L-Arginine vs. placebo crossover</td>
<td>5.6 g/d (2.8 g twice) or 12.6 g/d (4.2 g 3 times)</td>
<td>Arginine 98 ± 28 vs. 85 ± 21 (placebo)</td>
<td>Arginine: no diarrhea Placebo: 1/15 diarrhea</td>
<td>(67)</td>
</tr>
</tbody>
</table>

(Continued)
**Possible causes of dibasic amino acid-induced diarrhea**

The secretory effect of high perfused concentrations of L-arginine (79) was shown to be unique to L-arginine (not D-arginine) and it seemed that the effect was mediated by NO synthesis (80). However, during intestinal perfusion, L-arginine perfusion could not only reverse the prosecretory effects of NOS inhibition (81) but also caused secretion (as did the NOS inhibitor L-NAME). In contrast, luminal or systemic inhibition of NOS or i.v. arginine infusion reversed the prosecretory state induced by choleratoxin (82). This is clearly a complicated picture, because it implies that NO can be both a secretagogue at low concentration and an antisecretory absorbagogue at high concentrations. In other words, NO production is an important means of maintaining the appropriate secretory/absorptive state and may form a feedback loop that controls water and electrolyte balance in the intestine (83). Most stimulant laxatives require production of intestinal NO to exert their secretory effects. This can generally be blocked by NOS inhibitors whose action can be reversed by L-arginine (38).

**Intestinal assimilation and metabolism of arginine**

*From protein meals.* Pancreatic trypsin and carboxypeptidase B preferentially release lysine and arginine as free amino acids from dietary protein (93) so that after a protein meal, the free L-arginine concentration in the succus entericus increases from 0.36 mmol/L arginine (94) to 1.86 mmol/L with an increment of 0.16 mmol/L in peptide-bound arginine (95). Amino acid or protein meals almost exclusively increase the luminal concentration of 0.16 mmol/L in peptide-bound arginine (95). Amino acid or protein meals almost exclusively increase the luminal concentration of arginine (95).

**Intestinal assimilation and metabolism of arginine**

*From protein meals.* Pancreatic trypsin and carboxypeptidase B preferentially release lysine and arginine as free amino acids from dietary protein (93) so that after a protein meal, the free L-arginine concentration in the succus entericus increases from 0.36 mmol/L arginine (94) to 1.86 mmol/L with an increment of 0.16 mmol/L in peptide-bound arginine (95). Amino acid or protein meals almost exclusively increase the luminal concentration of arginine (95).

**Intestinal assimilation and metabolism of arginine**

*From protein meals.* Pancreatic trypsin and carboxypeptidase B preferentially release lysine and arginine as free amino acids from dietary protein (93) so that after a protein meal, the free L-arginine concentration in the succus entericus increases from 0.36 mmol/L arginine (94) to 1.86 mmol/L with an increment of 0.16 mmol/L in peptide-bound arginine (95). Amino acid or protein meals almost exclusively increase the luminal concentration of arginine (95).

**Intestinal assimilation and metabolism of arginine**

*From protein meals.* Pancreatic trypsin and carboxypeptidase B preferentially release lysine and arginine as free amino acids from dietary protein (93) so that after a protein meal, the free L-arginine concentration in the succus entericus increases from 0.36 mmol/L arginine (94) to 1.86 mmol/L with an increment of 0.16 mmol/L in peptide-bound arginine (95). Amino acid or protein meals almost exclusively increase the luminal concentration of arginine (95).

**Intestinal assimilation and metabolism of arginine**

*From protein meals.* Pancreatic trypsin and carboxypeptidase B preferentially release lysine and arginine as free amino acids from dietary protein (93) so that after a protein meal, the free L-arginine concentration in the succus entericus increases from 0.36 mmol/L arginine (94) to 1.86 mmol/L with an increment of 0.16 mmol/L in peptide-bound arginine (95). Amino acid or protein meals almost exclusively increase the luminal concentration of arginine (95).
of absorbed arginine (97) but that uptake of arginine was considerably faster from the peptide meal and more was taken up by peripheral tissues (98).

During intestinal perfusion. Human perfusion studies have characterized the transporter. When Hellier and colleagues (99) used the human intestinal perfusion technique to study arginine and lysine absorption, they observed that at "concentrations >100 mM/L, absorption of amino acid ceased and excessive secretion of water and electrolytes into the lumen occurred. It seems probable that this was a nonspecific toxic reaction of arginine upon the mucosa" (79). Perfusion at lower concentrations showed saturable kinetics for both arginine (Km, 3.8 mM/L; Vmax, 46.3 μmol·min⁻¹·30 cm⁻¹) and lysine (Km, 4.9 mM/L; Vmax, 51.3 μmol·min⁻¹·30 cm⁻¹). We have used the same technique to investigate the intestinal uptake of both components of OKG singly or together. Ornithine uptake could be fitted to a 2-component model comprising saturable (Km, 2.33 mM/L; Vmax, 17.7 μmol·min⁻¹·25 cm⁻¹) and nonsaturable transport (Vmax, 2.01 μmol·min⁻¹·25 cm⁻¹·mM⁻¹·L⁻¹). α-Ketoglutarate increased the affinity (Km, 33.2 mM/L) but not the capacity of the saturable system (Vmax, 20.3 μmol·min⁻¹·25 cm⁻¹), whereas nonsaturable transport increased by 60% (Vmax, 3.23 μmol·min⁻¹·25 cm⁻¹·mM⁻¹·L⁻¹). α-Ketoglutarate uptake had low affinity (Vmax, 22.3 mM/L) and high capacity (Vmax, 124.9 μmol·min⁻¹·25 cm⁻¹), which was unaffected by ornithine concentration (76). The only in vitro kinetic study of cystine or cysteine uptake reported that the Km for cystine was 1.4 mM/L (90).

These data therefore indicate that during intestinal perfusion, the dibasic amino acid transport system has modest capacity but high affinity with a component of passive diffusion. Following a protein meal, luminal arginine concentrations match the Km. This transporter system comprises 4 different transport proteins. At the apical membrane, amino acid uptake via b0,+ AT is driven by the membrane potential and exchanges dibasic amino acids and l-cysteine for intracellular neutral amino acids that reenter via System B0,+ at the basolateral membrane, dibasic amino acids efflux via y⁻ LAT1 in exchange for extracellular neutral amino acids. l-Cysteine uptake is essentially “downhill,” because the concentration gradient is maintained by intracellular reduction of l-cysteine to l-cysteine (90). Cystinuria and lysinuric protein intolerance arise from defects in the apical and basolateral transporters, respectively (87,100,101). Although some neutral amino acids can trans-stimulate arginine uptake, dipeptide uptake via di- and tri-peptide transporter (PEPT1) is a much more efficient stimulatory couple to dibasic amino acid transport (102). Thus, if adverse effects arising from arginine intake were purely osmotic, then coingestion of a stimulatory neutral amino acid or dipeptide might increase arginine absorption and relieve the symptoms.

Following single or multiple oral amino acid doses. The bioavailability of single oral doses of arginine (3–30 g) varies from 21 to 68% depending on the experimental method (57,103,104), whereas only 6–10% of NAC is bioavailable (105,106). If included in a meal, the bioavailability of NAC is much higher (107). Citrulline is also being investigated as a supplement from which arginine can be synthesized. The intestine of preterm infants is unlikely to be able to synthesize arginine from citrulline because of the low activity of argininosuccinate synthase and argininosuccinate lyase (108). In contrast, these enzymes are expressed at a higher level in term infants as a result of the cortisol surge of later in trimester 3 (109,110). The intestine may therefore be a major site of arginine synthesis in early life. This is eventually overtaken by development of renal arginine synthesis. In one recent study in children undergoing cardiopulmonary by-pass, oral citrulline supplements markedly increased plasma citrulline and arginine concentration and improved NO production (30). Conversely, the ability of oral arginine to increase plasma arginine and citrulline has been repeatedly shown in adults [e.g. (57)]. However, the increase in plasma citrulline concentration does not always occur (24,111) and there appear to be “responders” who increase plasma citrulline concentration and reduce glucose production and “hyporesponders” who do not (54). This may be a result of insulin resistance, because diabetes diverts the balance away from intestinal citrulline production toward net whole-body arginine synthesis (112–115). Perhaps the variable efficacy of citrullinemia following oral arginine loading reflects the extent of insulin resistance in different subjects. This will be discussed later.

The special case of cysteine and cystine absorption and gastrointestinal toxicity
As discussed above, 4 clinical situations in which cysteine/cystine toxicity should be considered are cystinuria, chronic gastrointestinal toxicity of arginine and related amino acids 1697S

**FIGURE 1** Multiple routes for intestinal assimilation of arginine and cysteine from dietary protein and oral supplements. The interrelations between the 3 transporters for cysteine (Cys ASC), cystine and arginine (Cys and Arg b0,+ AT), and PEPT1 is shown. Left: Cysteine may arise from dietary protein digestion, from secretions, or from NAC hydrolyzed by brush-border N-acetyltransferases. It may either be absorbed via the ASC transporter or may reduce oxidized glutathione to GSH (122) and will yield cysteine, which will be absorbed via b0,+ AT. Right: Arginine released during protein digestion as di- or tripeptides will be absorbed either as intact peptides via PEPT1 or, after hydrolysis to free amino acids, as free arginine via b0,+ AT. Arginine released from dietary protein as the free amino acid will be absorbed directly by b0,+ AT (14).
obstructive pulmonary disease, paracetamol overdose, and critical illness (where glutathione pools may be depressed). Cystinuria is characterized by excessive deposition of insoluble cystine crystals and stones within the urinary system. Treatment involves oral bicarbonate therapy that alkalinizes urine and thus solubilizes cystine or α-mercapto-propionylglycine or 5-n penicillamine treatment that converts cysteine to more soluble mixed disulfides (100). In addition, oral mega-therapy with vitamin C has been used to reduce cystine to its more soluble cysteine form that can be reabsorbed via the renal tubular small aliphatic amino acid transporter (ASC) system. Results of this therapy have been both encouraging and inconclusive (116,117). In contrast, cyst(e)ine uptake from dietary protein is normal in cystinuric patients, because much is taken up either as cysteine (90) or in di- and tripeptide form (118) (Fig. 1).

Under physiological conditions, a large efflux of reduced glutathione (GSH) and cysteine into the intestinal lumen (119) forms a redox control system that stimulates epithelial cell proliferation (more reduced) or apoptosis (more oxidized) and, hence, the health of the gut (120,121). It has been proposed that luminal GSH that has been oxidized by factors such as mucosal peroxidized lipids can be regenerated through the action of secreted cysteine, which will then be absorbed as cystine, reduced within the enterocyte, and secreted again (122) (Fig. 1).

Oral NAC must be deacetylated by mucosal brush-border N-acylases (123) before cysteine can be absorbed (Fig. 1). As discussed above, it is unclear how much of the cysteine liberated from NAC is converted to cystine within the intestinal lumen.

In the case of paracetamol (acetaminophen) overdose, rapid administration of NAC is most important if liver failure is to be prevented (124) and the regime requires a loading dose of 140 mg/kg with 17 following doses of 70 mg/kg every 4 h (total NAC, 1330 mg/kg over 72 h or 31 g/d for a 70-kg patient). The route of administration is still controversial. Oral treatment caused a high incidence of nausea, vomiting, and abdominal pain in 56% of patients in a large study (125) and 63% of patients in a small study (126). I.v. administration of NAC has fewer gastrointestinal side effects, but skin rash may be more common. The controversy is not over effectiveness but cost. I.v. treatment of renal disease. J Nutr. 2004;134:S2801–6.

In most clinical trials, however, in which NAC has been used as an antioxidant, side effects are rare at the typical dosage regime of 0.6–3.0 g/d in divided doses. In placebo-controlled clinical trials of NAC in patients with HIV/AIDS (129) or idiopathic pulmonary fibrosis (130) adverse gastrointestinal events occurred equally in NAC and placebo groups. A recent meta-analysis of meta-analyses (131) concluded that prophylactic NAC given to prevent contrast medium-associated nephropathy was associated with no evidence of toxicity or adverse effects in the 2284 patients enrolled in the trials.

To conclude, NAC ingested orally in small doses does not seem to cause any gastrointestinal symptoms but given at high doses has potent effects on the intestine. The etiology of these effects is unclear but may involve acute disturbances of luminal redox state. These conclusions were derived from a reasonable series of medium-sized clinical trials.

It is clear from the literature that arginine, ornithine, and NAC can produce adverse effects on the gastrointestinal tract when ingested at high doses but not when given as smaller divided doses. It is also clear that an effect of doses >10 g is not universally observed and in most studies, l-arginine was as well tolerated as l-citrulline and OKG (Table 1). There is clearly a need to establish the laxative threshold of these compounds, which is defined as the dosage that provokes symptoms of pre-determined severity in 50% of subjects. If we were, for the sake of argument, to postulate that 15 g of l-arginine provoked gastrointestinal symptoms in 50% of subjects, then this dosage contains 86 mmol of arginine, which compares with the laxative threshold of 74 g (194 mmol) (49) for lactitol. However, with lactitol, the effect of divided doses was cumulative, whereas the evidence suggests that by simply dividing the doses, symptoms could be eliminated at a daily intake of 30 g arginine (56). This simple maneuver might reduce the luminal point load presented to the small intestine below a critical threshold. By analogy with water instilled into the large bowel, slow infusion did not provoke diarrhea, whereas rapid infusion of 500 mL did (52).

At present, with respect to these amino acids, there is insufficient evidence to address this point. In one study, the rate of gastric emptying of a solution containing 12 g l-arginine was no different if 25 g of glucose was added (132). Although large oral amino acid loads have been shown to delay gastric emptying (107,133–135), an intriguing study recently showed that during continuous enteral nutrition, a 15-g bolus of arginine promoted earlier gastric relaxation (107,136), whereas high doses of 30 g induced diarrhea, as we had found with OKG (75). There is clearly a need to investigate this topic, because coingestion with other nutrients may be a way to improve compliance in healthy individuals by avoiding gastrointestinal side effects.

Patients appear to be more tolerant to the effects of single, large doses of l-arginine than healthy individuals with excellent insulin sensitivity. This might reflect the effect of insulin resistance on slowing gastric emptying, but this is not clear at present.

Similarly, there is insufficient data to show which form of arginine (free base, chloride, or aspartate salt) is best tolerated. There are also no data to show whether trans-stimulation of uptake by coingestion with a neutral amino acid might improve uptake. It may also be possible to improve pharmacokinetics by using dipeptides containing arginine because of their more rapid uptake. Unfortunately, arginine homopolymers, which are polyacations, are contra-indicated, because they damage the mucosa, as Smith and colleagues (137) discovered nearly 30 y ago, and are efficient cell-penetrating peptides that augment drug delivery (138).

Intolerance therefore seems to occur through a combination of simple osmotic action and excess NO production, but there are still several strategies that might reduce the likelihood of adverse effects and further improve the therapeutic usefulness of these amino acids.

**Literature Cited**
