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Review of Copper Provision in the Parenteral Nutrition of Adults

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Abstract

The essential trace element copper (Cu) is required for a range of physiologic processes, including wound healing and functioning of the immune system. The correct amount of Cu must be provided in parenteral nutrition (PN) if deficiency and toxicity are to be avoided. While provision in line with the standard recommendations should suffice for most patients, Cu requirements may be higher in patients with increased gastrointestinal losses and severe burns and lower in those with cholestasis. The tests of Cu status that are currently available for clinical use are unreliable. Serum Cu concentration is the most commonly ordered test but is insensitive to Cu deficiency and toxicity and is misleadingly increased during the acute phase response. These limitations make it difficult for prescribers to assess Cu status and to decide how much Cu to provide. There is a need for better tests of Cu status to be developed to decrease uncertainty and improve individualization of Cu dosing. More information is needed on Cu requirements in disease and Cu contamination of PN components and other intravenous fluids. New multi–trace element products should be developed that provide Cu doses in line with the 2012 American Society for Parenteral and Enteral Nutrition recommendations. This article discusses the evaluation and treatment of Cu deficiency and toxicity in patients treated with PN. (*Nutr Clin Pract*. XXXX;xx:xx-xx)

Keywords

copper; copper deficiency; copper toxicity; parenteral nutrition; nutritional support

Copper (Cu) is an essential trace element (TE) required for metabolism in all living cells. It has been known to be an essential component of parenteral nutrition (PN) since 1972.[1] Provision of the correct amount of Cu is necessary, not only to avoid deficiency and toxicity, but also to promote optimal recovery.[2] This article covers the relevant physiology of Cu. It then discusses Cu deficiency and toxicity and examines the evidence behind the recommendations on Cu provision in PN. It provides practical guidance on assessment of Cu status and Cu requirements. The article focuses on adults because provision of Cu in pediatric PN has recently been covered elsewhere,[3] but pediatric studies have been cited where considered appropriate.

Physiology

Cu is among the 3 most abundant transition metals in biological systems, the other 2 being iron (Fe) and zinc (Zn). The human body contains around 100 mg of Cu, more than half of which is in bone and muscle. The highest concentrations are in liver, kidney, and brain, reflecting the high metabolic activity of these organs.[2] Only 5% is in blood, about 95% of this being bound to ceruloplasmin (Cp) and the remainder to albumin and amino acids.[4] The serum Cu concentration therefore largely reflects the Cp concentration and is affected by factors influencing Cp. Intracellular Cu is usually bound to chaperones and other proteins because free Cu is potentially harmful to cells.

Cu, which functions as a component of cuproproteins, has a variety of physiologic roles. For example, it is required for humoral immunity and production of inflammatory cytokines.[5,6] It was recently suggested that the immune system uses Cu intoxication as a means of intracellular bacterial killing.[7,8] Cu is required for the physiologic response to low Fe stores. It is needed for absorption of dietary Fe, release of Fe from hepatocytes and production of hemoglobin.[9] Cp oxidizes Fe from the ferrous to ferric state, enabling its transport by transferrin and subsequent use in erythropoiesis. Superoxide dismutase is an antioxidant cuproenzyme that catalyzes the conversion of superoxide radicals to hydrogen peroxide, which is then reduced to water.[10] Cu is also necessary for wound healing because it is required for the synthesis of collagen.

The average daily oral intake of Cu is 1.0–1.6 mg, which exceeds the RDA of 0.9 mg.[11] Dietary Cu is absorbed mainly in the stomach and upper small intestine.[2,12] About 55%–75% of dietary Cu is absorbed, which is a high proportion as compared

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with other TEs.[2] The amount of Cu absorbed depends on dietary intake, Cu status, and the effects of other nutrients. Absorption is decreased by vitamin C, Fe, and Zn.[2] Zn inhibits Cu transport directly and by inducing metallothioneins that bind Cu in intestinal mucosal cells. Short-chain fructo-oligosaccharides[13] and Fe deficiency[9] increase Cu absorption. Once absorbed, Cu is bound to albumin and transcuprein and transported to the liver, where it is stored, released into the systemic circulation, or excreted into bile. Normally, about 80% of Cu excreted from the body is in bile and gastrointestinal (GI) secretions, and about 20% is in urine.[14,15] About 10%–15% of biliary Cu is reabsorbed, returning to the liver in an enterohepatic circulation. External fluid losses from fistulae, bile leaks, or enterostomies predispose to Cu deficiency by decreasing the amount of Cu reabsorbed.

Cu Deficiency

Deficiency occurs when Cu intake is consistently below requirements. This section focuses on Cu deficiency encountered among patients receiving nutrition support. Factors affecting individual susceptibility to deficiency during PN include the size of hepatic Cu stores, the extent of GI losses, and the amount of Cu provided. Cu provision includes supplementation of PN but also the amount delivered by concurrent oral or enteral nutrition and as a contaminant of PN and other intravenous (IV) fluids.

Causes of Cu Deficiency

Overt Cu deficiency is uncommon during short-term PN but has occurred during long-term PN when Cu has been provided below requirements.[16-19] A recent retrospective review of the Cu status of hospitalized pediatric patients treated with PN observed that, after 14 days of treatment, hypocupremia was present in 71% of patients receiving PN unsupplemented with Cu, as compared with 50% of those receiving supplemented PN.[20] Deficiency has also occurred when supplemental Cu has been withheld either because of concern about accumulation during cholestasis[21,22] or because of shortages of multi-TE (MTE) products.[23-25] There is much concern about the clinical implications of such shortages, especially for infants, for whom micronutrient deficiency can have irreversible consequences.[26] The prevalence of marginal Cu deficiency among patients treated with PN is unknown but could be anticipated to be higher than that in the general population, given the higher prevalence of risk factors for deficiency in hospitalized patients.

Short bowel syndrome. Patients with short bowel syndrome (SBS) are at risk of Cu deficiency even after weaning onto oral diet. A recent study investigated 22 adults with SBS following intestinal resection, of whom half had been weaned onto oral diet and half remained dependent on PN.[27] Patients in the PN group had a small intestine with a median length of 25 cm (range, 10–100), and 6 patients had a colon. Patients in the orally fed group had a small intestine with a median length of 110 cm (range, 40–210), and 9 patients had a colon. Serum Cu concentrations were significantly lower for the patients treated with PN (69 ± 24 μg/L, $P < .05$) and patients taking oral diet (72 ± 26 μg/L, $P < .05$), as compared with a control group (109 ± 16 μg/L; reference range, 70–140 μg/L). Similarly, Cu deficiency can occur during transition from PN to enteral nutrition (EN). A study of transition to EN for pediatric patients reported Cu deficiency as the most common among micronutrients, affecting 56% of patients.[28] After full EN was established, deficiency of Cu was less prevalent (22%) than vitamin D (68%), Zn (67%), or Fe (32%). These findings emphasize the importance of monitoring Cu status among patients with SBS, irrespective of the type of nutrition support.

Teduglutide—an analogue of glucagon-like peptide 2, which is an intestinal growth factor—has been used in patients with SBS to improve absorption of dietary nutrients and decrease dependence on PN. A recent case series reported on adverse events during weaning of patients treated with teduglutide.[29] One patient developed overt Cu deficiency, despite Cu supplementation, which did not respond to oral supplementation and necessitated recommencement of PN. In this patient, it appears that Cu absorption did not improve in response to teduglutide. Glucagon-like peptide 2 has recently been observed to improve bile flow in an animal model of cholestasis,[30] but its effect on Cu balance in humans is unknown.

Enteral tube feeding. Patients receiving long-term EN, especially those fed through a jejunostomy tube, are at risk of developing Cu deficiency. For example, one case series compared 23 patients fed through percutaneous endoscopic jejunostomy and 36 patients fed via percutaneous endoscopic gastrostomy.[31] After 6 months of EN, serum Cu was significantly lower in the percutaneous endoscopic jejunostomy group ($P < .001$). Six patients in this group had severe Cu deficiency with hematologic features. There have also been reports of severe Cu deficiency in patients on home EN via percutaneous endoscopic gastrostomy.[32] Enterally fed patients appear to be at risk of Cu deficiency because of decreased bioavailability rather than inadequate intake. The risk is higher in jejunostomy-fed patients because the main sites of Cu absorption are bypassed. In addition, interindividual variation in Cu absorption is high. Furthermore, absorption may be impaired by high enteral intake of Zn or Fe.

Bariatric surgery. Cu deficiency can occur after bariatric surgery, more commonly after biliopancreatic diversion than after Roux-en-Y gastric bypass. A 5-year follow-up study observed hypocupremia postoperatively in 30.3% of patients with biliopancreatic diversion, compared with 3.8% of patients with Roux-en-Y gastric bypass.[33] None of the patients had hematologic or neurologic features of deficiency, which suggests that, for overt deficiency to occur, deficiency must be sustained

long-term. Cu deficiency after bariatric surgery has recently been reviewed.[34,35] Clinical Cu deficiency has been described but is unusual for patients receiving adequate supplementation. The American Association of Clinical Endocrinologists' guidelines for perioperative support of the patient undergoing bariatric surgery recommend that Cu does not need to be routinely measured postoperatively but should be measured if there are hematologic or neurologic features consistent with deficiency or if there is impaired wound healing.[36] Clinicians should monitor patients carefully for these features.

Zn excess. Zn-induced Cu deficiency (ZICD) has been caused by excessive ingestion of Zn, but there is limited awareness among clinicians of this potential side effect of Zn supplementation.[37] It tends to occur with oral Zn doses >850 mg/d for 1 year, but negative Cu balance has resulted from doses of 18.5 mg/d for 2 weeks.[38] Misdiagnosis of Zn deficiency during an acute phase response (APR) can result in inappropriate Zn supplementation. ZICD should therefore be considered for patients treated with PN, many of whom have an ongoing APR and high Zn requirements. For patients treated with PN, the source of excess Zn may not necessarily be parenteral. For example, excessive intake of Zn from a denture adhesive was recently reported to cause ZICD for a patient on long-term PN containing standard Cu provision.[39] The patient presented with mild pancytopenia and perioral paraesthesia, which did not respond to additional Cu provision but resolved after the adhesive was switched to a Zn-free product. To facilitate diagnosis of ZICD, it has been recommended that, following the finding of hypocupremia, TE laboratories should automatically measure the Zn of the specimen.[40] Similarly, the finding of hyperzincemia should prompt the measurement of Cu to exclude ZICD. To avoid ZICD, it has been suggested that, to treat Zn deficiency, 1 mg of Cu should be given for every 8–15 mg of Zn (elemental doses).[36] Zn and Cu should be taken at least 2 hours apart to maximize the absorption of Cu.[41]

Other causes of Cu deficiency. Cutaneous exudative Cu losses amounting to 37 mg/wk have been reported for patients with severe burns.[42] There is evidence that supplementation of Cu in combination with other micronutrients improves the clinical outcome for patients with burns[43] or pressure ulcers.[44] The results of 2 randomized controlled trials were combined in which patients with burns received placebo or a combination of Cu, selenium, and Zn at doses up to 4 mg, 500 μg, and 40 mg, respectively, by IV infusions not associated with PN use.[45] In total, 41 patients were investigated for up to 21 days. There was a significant reduction in nosocomial pneumonia in patients supplemented with TE. The extent to which this outcome was attributable to Cu is unknown.

As with other micronutrients, Cu is readily dialyzed. Continuous renal replacement therapy can result in significant Cu losses, especially if prolonged. If Cu provision is insufficient to replace losses, this will lead to negative Cu balance. Effluent losses of 0.41 mg/d have been reported,[46] an amount similar to the recommended Cu provision in PN. In the same study, Cu was undetectable in replacement solutions. Hemodialysis and peritoneal dialysis have not been associated with Cu deficiency in human subjects, but a recent study in an animal model concluded that hemofiltration may necessitate Cu replacement at doses exceeding standard provision.[47]

Chemotherapy with cisplatin should be considered a risk factor for Cu deficiency. Patients with esophageal cancer treated with cisplatin and PN had significantly lower serum Cu concentrations postchemotherapy ($P = .015$).[48] This change was prevented by additional TE supplementation. Iatrogenic Cu deficiency has also been observed following overtreatment with chelating agents, such as penicillamine, trientine, and tetrathiomolybdate, but this has not been reported in the context of PN.[49]

Features of Cu Deficiency

When intake is below requirements, Cu is initially replenished from hepatic stores. As a result, features of deficiency may occur long after the causal insult. When deficiency has occurred after omission of Cu from PN, clinical features have taken between 6 weeks and 15 months to appear.[21-23,25,50,51] In a series of 26 cases of neurologic complications occurring after bariatric surgery, overt Cu deficiency presented as late as 9 years postoperatively.[52]

The clinical features of Cu deficiency are mainly hematologic and neurologic, reflecting the requirement for Cu in erythropoiesis and synthesis of myelin.[53] The most common features are a microcytic or normocytic anemia, unresponsive to Fe supplementation, and neutropenia.[54] The mechanism whereby anemia develops is uncertain, but decreased Cp may result in impaired mobilization of Fe stores. Cu deficiency is also implicated in increased oxidative stress and deterioration in cognitive function for patients with Alzheimer's disease.[55]

The most common neurologic feature of Cu deficiency is myelopathy, but peripheral neuropathy and demyelination have also been reported.[56] The deficits of myelopathy resemble those of subacute combined degeneration of the cord resulting from vitamin B_{12} deficiency.[57] Patients typically present with a disordered gait and sensory ataxia. The mechanism of neurologic damage is unknown, but cuproenzymes such as cytochrome C oxidase have vital roles in the nervous system, the impairment of which would be expected to have adverse effects. When neurologic features consistent with Cu deficiency are present, the patient should be investigated by measurement of serum Cu and by spinal magnetic resonance imaging. In a case series of 25 patients with Cu deficiency myelopathy, abnormalities on magnetic resonance imaging were found in 44%, the most common being an increased T2 signal of the dorsal column in the cervical and thoracic cord.[58] In this case series, the duration of patients' symptoms prior to diagnosis of Cu deficiency ranged

from 2 months to 10 years. Treatment with Cu supplementation may arrest the neurologic deficits and result in improvement in sensory symptoms, but most patients have residual deficits resulting from irreversible neurologic injury. This emphasizes the importance of early diagnosis. Several months of Cu supplementation are likely to be required before the features improve. A study of 12 patients with Cu deficiency, all of whom had neurologic features, observed significant improvements in functional activities of daily living ($P = .007$) over 12 months of Cu supplementation.[59]

Nephrotic syndrome was recently reported as a feature of Cu deficiency for a patient treated with PN following bowel resection.[60] The patient also had anemia, neutropenia, and deteriorating kidney function, which responded to Cu supplementation. The urinary protein loss was attributed to loss of the protective effect of Cp on glomeruli. While this is the only published case report of nephrotic syndrome resulting from Cu deficiency, urinary protein loss has been reported to correlate with urine Cu for patients with nephrotic syndrome.[61]

Cu deficiency should be considered when metabolic bone disease occurs for patients treated with PN. The earliest reported case of Cu deficiency for a patient treated with PN was an infant who presented with osteoporosis and growth delay.[1] Osteoporosis resulting from Cu deficiency was also reported in 2 preterm infants with SBS treated with long-term PN.[62] It was diagnosed at 5 months of age following investigation for musculoskeletal discomfort. Both patients had severe hypocupremia and responded to IV supplementation of Cu. Pseudoscurvy has been reported as a feature of Cu deficiency,[63] as demonstrated in a 4-month-old female infant treated with PN from which TEs had been omitted.

There is evidence from numerous sources that Cu deficiency impairs the activity of the immune system, thereby predisposing to bacterial infection.[5,64] In vitro studies have observed decreased neutrophil function, decreased secretion of interleukin 2 from lymphocytes, and decreased cytotoxic activity of natural killer cells. Mortality from infection is higher in Cu-deficient animals.[65] For humans, it has long been known that Cu deficiency impairs phagocytosis[66] and increases mortality from infection.[67] Pulmonary and urinary tract infections are more common for patients with Menkes's disease, an inherited disorder of severe Cu deficiency.[68] The results of supplementation studies for patients with burns, discussed above, also suggest that Cu deficiency predisposes to pneumonia.[45] The conclusions that can be drawn from clinical studies regarding the role of Cu deficiency in infection are limited, given that micronutrient deficiencies do not occur in isolation; but, when taken together, the evidence is compelling to suggest that Cu deficiency impairs the immune system.

The clinical consequences of marginal Cu deficiency are unknown but may include neurologic,[57] cardiac,[69] and immune dysfunction.[5,70] A low-Cu diet has resulted in decreased proliferation of mononuclear cells.[71] Given the extensive role of Cu in Fe metabolism, it could be anticipated that marginal Cu deficiency may compromise utilization of Fe. These observations suggest that marginal Cu deficiency could be detrimental for patients treated with PN, but this awaits investigation.

Treatment of Cu Deficiency

The underlying cause of deficiency should be sought and treated. For patients already supplemented with Cu, the dose should be reviewed and possible factors decreasing bioavailability considered. Ideally, supplementation should be delivered orally or enterally, to enable absorption according to requirements and to avoid bypassing homeostatic mechanisms. However, for patients treated with PN, enteral tolerance or bioavailability of Cu may be limited by intestinal failure. In addition, the need to ensure delivery of Cu for patients with severe deficiency may necessitate IV administration. Supplementation should continue until normal serum Cu concentrations are restored and clinical features resolve. This should be followed by long-term supplementation sufficient to prevent recurrence of deficiency. When Cu deficiency has occurred for patients treated with PN, serum Cu concentrations have normalized, and clinical features have improved within 6 weeks of supplementation.[21,22,72] The hematologic features resolve within about 4 weeks, but neurologic features improve relatively slowly and may be in part irreversible. The rate of resolution of Cu deficiency is likely to depend on the severity of deficiency, route of supplementation, and Cu dosage. If features of deficiency do not respond to increased Cu provision, ZICD should be ruled out.

Cu dosing should be individualized according to the severity of hypocupremia and clinical features. Cu is available as MTE products, which typically provide 0.4–1.0 mg/mL, oral tablets (2 and 5 mg), and injection (0.4–2 mg/mL).[73] Severe Cu deficiency in adults can be treated with IV Cu (2–4 mg/d) for 6 days, followed by oral Cu sulfate or gluconate (3–8 mg/d).[36] In a reported case of severe Cu deficiency, it was possible, following IV Cu supplementation, to maintain the serum Cu concentration in the reference range by high-dose oral supplementation (8 mg/d), despite loss of absorptive surface area.[74] This suggests that sufficient absorption may be achieved by supersaturation of the remaining Cu transport capacity.[75]

When Cu deficiency occurs during EN, there are various treatment options. Enteral Cu provision can be increased by giving either pharmaceutic Cu products or cocoa powder, of which 100 g contains 3.61–3.79 mg of Cu.[76] In practice, daily doses of 10–40 g of cocoa powder have been used[31] that, in this study, were reported to deliver Cu doses of 1.36–2.56 mg/d. Clinicians should be aware that Cu may be poorly absorbed when given through a jejunostomy tube.[77] If necessary, IV Cu can be given. It is advisable to monitor Cu status for patients on long-term EN.

Cu Toxicity

Patients with acute Cu toxicity present with vomiting, diarrhea, and abdominal pain and, if more severe, hepatic necrosis, renal failure, encephalopathy, and death.[78] Acute toxicity has been reported following consumption of Cu-contaminated water but would be unlikely to occur during PN, unless an error resulted in overdose. For patients treated with long-term PN, there is concern about hepatotoxicity resulting from chronic hepatic accumulation of Cu in PN-associated liver disease (PNALD). Accumulation may occur if biliary excretion of Cu is impaired, either because of immaturity of excretory mechanisms or because of cholestasis. Overload may be exacerbated by excessive provision of Cu resulting from inappropriately formulated MTE products, contamination of PN, or additional intake from other sources.

There is extensive evidence that hepatic Cu accumulation occurs in PNALD. This is usually at concentrations below the diagnostic threshold for Wilson disease (WD)—that is, <250 μg/g, dry weight (reference range, <35 μg/g).[79,80] In 2005, Blaszyk et al measured hepatic Cu concentrations for patients treated with long-term PN who had abnormal liver enzymes and for control subjects who had drug-induced cholestasis.[81] In 89% of PN patients, hepatic Cu was >35 μg/g and in 29%, >250 μg/g (range, 10–2248). Hepatic Cu was also increased in the control subjects. It did not correlate with the serum Cu concentration or duration of PN. These findings suggest that cholestasis is the main causal factor in hepatic Cu accumulation for patients treated with PN, but they do not exclude the possibility that excessive provision is harmful. Given that about 30% of adult patients receiving long-term PN develop PNALD, the results from this small study, if typical of the PN population, suggest that about 10% of adults on long-term PN have significant hepatic Cu accumulation. An autopsy study of tissue TE concentrations was carried out in 8 adults with short bowel treated with long-term PN, compared with 45 control subjects on oral diet who had not suffered from GI disease.[82] Cu dosing was in accordance with 1979 recommendations,[83] the mean daily dose being 1.4 mg for 14 years. Cu concentrations were increased in liver and kidney specimens from patients receiving PN, being highest (>250 μg/g) in 2 who died of liver failure.

While the clinical outcome in PNALD appears to be poor for patients with severe hepatic Cu accumulation, it is unclear whether the Cu is directly hepatotoxic. Various observations suggest that Cu accumulation may be harmful. Supraphysiologic concentrations of Cu are known to be pro-oxidant, generating reactive oxygen species that can cause oxidative damage to macromolecules.[78] In addition, Cu accumulation in WD is hepatotoxic, neurotoxic, and nephrotoxic.[79,84] However, PNALD and WD are not directly comparable, because they have different causes and clinical features and the extent to which Cu causes harmful oxidative effects in vivo in PNALD is unknown, as are its macromolecular targets.[85] Moreover, the observation that neonates tolerate high hepatic Cu concentrations without adverse effects suggests that Cu accumulation per se is not necessarily harmful.[86] Whether Cu is hepatotoxic may depend on factors other than its total hepatic concentration, including its subcellular location and extent of protein binding. It has also been suggested that Zn decreases the hepatotoxicity of Cu in WD,[87] but whether it does so in PNALD is unknown. Until the results of further research clarify whether Cu is hepatotoxic in PNALD, Cu should be considered potentially harmful for patients treated with long-term PN. Chronic Cu toxicity is also implicated in atherosclerosis and neurodegenerative diseases, including Alzheimer's disease and Parkinson's disease.[88]

Assessment of Cu Status

Serum Cu

All currently available biomarkers of Cu status are unreliable. The serum Cu concentration is the most useful and most frequently ordered test, but its limitations need to be considered.[89] When interpreting individual results, the clinician should first consider how reliably this can be done through the population reference range. Second, when interpreting serial results, the clinician must decide whether a change between consecutive results is significant, possibly requiring clinical intervention, or can be accounted for by a combination of biological variation and analytic imprecision. Regarding use of the reference range, serum Cu has an index of individuality of 0.41, which is low (ie, a ratio of intraindividual variation to interindividual variation).[90] Consequently, results within the reference range do not exclude the possibility that there has been a disease-related change in concentration that is highly significant for the individual. Unless a previous result is available for comparison, the clinician will be unaware of the significance of the result. Clearly, this low index of individuality decreases the value of the population reference range for interpreting individual serum Cu results. Regarding serial results, critical difference values can help clinicians interpret the significance of changes. Serum Cu has been reported to have a critical difference of 2.3 μmol/L (14.6 μg/L), suggesting that a relatively small change between consecutive results is likely to be significant.[90] Critical differences should ideally be determined by all clinical laboratories because the values are influenced by analytic imprecision, which varies among laboratories.[91]

The serum Cu concentration is insensitive to deficiency, tending to remain within the reference range except in severe deficiency. A normal or increased serum Cu result does not therefore rule out deficiency.[15] It is also insensitive to hepatic Cu accumulation, tending to plateau once requirements are met and correlating poorly with tissue accumulation.[14,81,82] The lack of correlation with Cu status makes serum Cu an unreliable test for guiding supplementation of PN, except at extremes.[92] Hypocupremia can occasionally occur in the absence of deficiency (eg, for patients with WD).[93] When the

cause of hypocupremia is uncertain, WD can be ruled out by the finding of a 24-hour urinary Cu ≤0.6 μmol.[94]

Confounding factors cause serum Cu to increase in the absence of Cu excess, the most common being the APR, during which proinflammatory cytokines stimulate the synthesis of Cp irrespective of Cu status. Indeed, hypercupremia is to be expected in hospitalized patients with trauma or infection or in those who are postsurgical.[95] A recent retrospective review of Cu status in hospitalized patients treated with PN observed that serum Cu correlated with C-reactive protein concentrations >4 mg/dL ($P = .03$).[20] This APR-associated increase in serum Cu concentrations can mask deficiency[2] or cause unnecessary concern about toxicity, either of which could result in inappropriate clinical action. In addition, serum Cu concentrations measured for monitoring the treatment of deficiency should be interpreted with caution if there is a concurrent APR. Cp synthesis is also stimulated by estrogens, resulting in increased serum Cu concentrations among women who are pregnant or taking estrogens. In the presence of these confounding factors, serum Cu concentrations within the reference range do not exclude deficiency, but hypocupremia is consistent with a diagnosis of Cu deficiency.[14,15,96,97]

It is advantageous to measure the Cp concentration when measuring serum Cu. Both increase in parallel because their concentrations are approximately linearly related. Any increase in Cu caused by a confounding factor will then be readily apparent. To allow for changes in Cp caused by age, sex, or inflammation, authors have suggested routinely adjusting serum Cu for the Cp concentration[98] or calculating the Cu:Cp ratio.[99] Laboratories should determine their own adjustment equation or ratio because these depend on the methods used and population studied.[100]

Cuproproteins

Many cuproproteins other than Cp have been investigated as possible markers of Cu status, but none reliably detect early deficiency or toxicity.[5,101] Assays are unstandardized, necessitating that individual laboratories determine local reference ranges. These tests are also subject to high intraindividual variation. Lability of cuproenzymes may necessitate rapid specimen processing, confining analysis to hospitalized patients. There is also limited information available on the diagnostic sensitivity and specificity of these tests.

Superoxide dismutase in red blood cells is considered a relatively sensitive marker of Cu deficiency, decreasing in Cu deficiency and in subjects with low Cu intake, but the change occurs slowly because of slow turnover of red blood cells.[102] Plasma diamine oxidase decreases in Cu-deficient subjects but has limited use in diagnosis because it increases during tissue injury. Studies have also investigated platelet cytochrome C oxidase as a biomarker of Cu status, but it is limited by lability and high interindividual variation. Neither marker is routinely measured in clinical practice. These are discussed in detail elsewhere.[4,89]

Cu chaperone for superoxide dismutase (CCS) is the most promising potential biomarker of Cu status. In humans, mononuclear cell mRNA for CCS increases in malnourished Cu-deficient patients and decreases in response to Cu supplementation.[102,103] A recent study observed that neither CCS protein nor mRNA transcripts were influenced by inflammatory status, supporting their use as biomarkers of Cu status.[104]

Liver Cu

The most reliable indicator of Cu status is liver Cu concentration, but this has limitations.[97] First, underestimation may result from inhomogeneous distribution of Cu.[105] Second, liver biopsy may be unsafe or contraindicated for some patients and is not feasible to repeat frequently, because of its invasive nature. A recently established technique called *laser ablation–inductively coupled plasma–mass spectrometry* has been used to measure liver Cu concentrations.[106] It is more accurate, quicker, and cheaper than standard metal deposit measurement and can simultaneously measure Zn and selenium. When applied to liver specimens from patients with WD, it has confirmed that hepatic Cu is inhomogeneously distributed, but the technique has not yet been applied to PNALD. The prognostic value of this method is worthy of assessment, but ideally, hepatic Cu would be measured noninvasively. This may eventually be possible through imaging techniques.[101]

Metabolomics and Transcriptomics

Clinically useful biomarkers of micronutrient status may be identified by using *-omics* techniques to detect changes in response to supplementation.[107] As yet, few such approaches have been described in relation to Cu status. A recent study of proteins correlating with micronutrient status in undernutrition unexpectedly identified a Ras protein that explained variation in plasma Cu concentration additional to that explained by Cp. This protein merits further investigation as a biomarker of Cu status.[108] A network of proteins representing the Cu interactome identified ATPases 7A and 7B as proteins worthy of further assessment as markers of Cu status.[102]

There is a clinical need for biomarkers, ideally measurable in peripheral blood, that can detect Cu accumulation before the onset of clinical features. Studies in animals have shown that *-omics* approaches can sensitively detect Cu toxicity by observing genetic and metabolic changes. A transcriptomics approach observed downregulation of genes associated with cholesterol synthesis, as in a mouse model of WD, and upregulation of metallothionein and catalase.[109] A metabolomics approach identified a metabolic signature of Cu exposure.[110] These techniques have not yet been applied to the study of Cu exposure in humans. At present it is difficult to predict which patients are

Table 1. Parenteral Copper Requirements in Adults.

Condition: Requirement, mg/d	Year	Reference
Stable		
0.5–1.5	1979	83
0.3–0.5	2002	112
317–518[a]	2014	113
Diarrhea: 0.4–0.5	1981	92
Cholestasis: 0.15	1981	92

[a]Values in µg/d.

susceptible to Cu accumulation, but the ability to do this could guide Cu provision. Gene testing may have predictive value because it is likely that genetic factors contribute to interindividual variation in the effects of Cu exposure. For example, individuals who are heterozygous for ATP7B mutations for WD may be predisposed to liver disease when exposed to excess Cu during long-term PN. The carrier frequency for these genes is relatively common at 1 in 90.[111] These are all key areas for future research.

Practical Considerations

Standard Requirements

In 1979 the American Medical Association published recommendations on Cu supplementation of PN based on knowledge of oral intake and estimated absorption of Cu from a normal diet.[83] The amount recommended for adults was 0.5–1.5 mg/d. Subsequently, the results of Cu balance studies suggested that the dose should be lower.[92] In response to this, the American Society for Parenteral and Enteral Nutrition (ASPEN) changed the standard recommendation to 0.3–0.5 mg/d, which has remained unchanged since 2002[112] (Table 1). The European Society for Clinical Nutrition and Metabolism (ESPEN) made the same recommendation in its guidance on perioperative PN.[114] These recommendations were supported by a recently published systematic review of TE supplementation in PN.[115]

Individualization of Cu Provision

Standard recommendations should be considered a starting point for estimating individual requirements and adjusting Cu provision accordingly. However, studies suggest that in practice this is poorly done. A 2013 Canadian review of 135 patients treated with long-term PN observed that Cu supplementation was 0.64 ± 0.35 mg/d, exceeding the standard recommendation.[116] Supplementation did not appear to be influenced by factors such as the GI anatomy of individual patients or indication for PN. This failure to adjust Cu provision risks causing deficiency or toxicity. Similarly, a retrospective observational study of TE status and dosing among 26 adult patients treated with long-term PN reported that 95.5% of Cu doses delivered exceeded the standard recommendation.[117] Cu doses of 1 mg/d resulted in hypercupremia in 22.5% of the tests performed. The excessive Cu dosing observed in these studies is in part a consequence of inappropriately formulated MTE products.

In practice, prescribers have insufficient information to enable them to fully individualize Cu doses. This would require knowledge of the patient's Cu status, the disease-specific requirements, and the amount of bioavailable Cu already present in the PN and other sources. Nevertheless, individual clinical circumstances should be carefully assessed and Cu provision adjusted if necessary. In what follows, situations are considered in which individual requirements may differ from the standard recommendation. The discussion is confined to Cu, but in practice all micronutrients should be considered together.

Increased requirements. Cu requirements increase for patients with prolonged, increased GI losses or persistent malabsorption.[16,118] In this situation, it may be appropriate to give higher doses of Cu. Balance studies have suggested that patients with persistent diarrhea (GI secretions >300 g/d) require 0.4–0.5 mg/d to maintain balance (ie, doses at the upper end of the standard recommendation).[92] This study investigated patients receiving total PN. In practice, however, many patients with short bowel take some oral diet, the effect of which on parenteral Cu requirement is difficult to predict. Oral diet resulting in net Cu absorption will decrease the requirement. However, oral diet also stimulates the production of Cu-containing secretions, potentially resulting in net Cu loss and an increased requirement. In practice the net effect of oral diet on parenteral Cu requirement is unknown because it is not feasible to carry out balance studies with individual patients.

The observations of high exudative Cu losses for patients with severe burns suggest that Cu requirements are likely to increase in these patients. ASPEN has recommended higher Cu provision in this situation.[73] ESPEN has recommended increasing provision 5-fold (3.0–3.5 mg/d) especially while wounds remain open.[119] Higher Cu doses may also be required to replace dialysate losses for patients treated with continuous renal replacement therapy.[46] Increased Cu provision should also be considered for patients treated with cisplatin.[48] Clearly, higher doses are required for patients with Cu deficiency. If deficiency is suspected, it may be appropriate to increase Cu supplementation to provide at least 1.0 mg/d in PN.[16]

Decreased requirements. Various publications suggest that for critically ill patients the amount of Cu that would be required in PN is decreased. ESPEN recommends that, except for patients with severe burns, parenteral Cu requirements are below the amount provided by currently available MTE products.[119] Elsewhere, authors have recommended against delivery of Cu to

Table 2. Practical Assessment of Cu Status During Parenteral Nutrition.

Assessment	Rationale
Clinical workup	
History	Elicit possible causes and consequences of Cu deficiency and toxicity.
Examination	Elicit anemia, poor wound healing, and neurologic abnormalities, which may occur in deficiency. Features of liver disease may occur in PNALD.
Biochemistry (serum)	
Cu	Hypocupremia is consistent with Cu deficiency.
Cp	Assists with interpretation of serum Cu results.
CRP	Assists with interpretation of serum Cu by quantifying APR.
Zn	Overprovision of enteral or oral Zn can cause Cu deficiency.
Fe status	Fe deficiency may coexist with Cu deficiency.
Vitamin B_{12}, D, E	Vitamin deficiency may coexist with Cu deficiency.
Full blood picture	Microcytic or normocytic anemia and neutropenia can occur in Cu deficiency.
Liver Cu	Confirmation of Cu accumulation.
Bone imaging	Osteoporosis can occur in Cu deficiency.

APR, acute phase response; Cp, ceruloplasmin; CRP, C-reactive protein; Cu, copper; Fe, iron; PNALD, parenteral nutrition–associated liver disease; Zn, zinc.

critically ill patients at doses >1.2 mg/d.[120] In a study of Cu provision for critically ill patients treated with PN, doses of 0.3 mg/d were sufficient to maintain constant serum Cu concentrations.[121] In consideration of these recommendations, it should be remembered that critical illness encompasses a diverse range of conditions and disease severities.

It may be necessary to decrease Cu provision for patients with cholestasis. A difficulty in practice is that cholestasis is difficult to quantitate and its severity varies widely. Ideally, it would be quantitated by direct measurement of bile flow, but this cannot be done in clinical practice. Clinicians therefore have to rely on surrogate measures and the presence of clinical features. Cholestasis can be considered to be present if there is direct (conjugated) hyperbilirubinemia with (1) direct bilirubin >1 mg/dL when total bilirubin is <5 mg/dL or (2) direct bilirubin >20% of total bilirubin when it is >5 mg/dL.[3]

Limited data are available on which to base guidance on parenteral Cu provision for patients with cholestasis. However, the observations of liver Cu accumulation for patients with cholestasis suggest that caution is necessary.[81,82] Howard et al recommended that Cu be withheld once liver aminotransferase and alkaline phosphatase levels increase to twice that of reference values and before serum bilirubin levels increase.[82] ASPEN has recommended decreasing or withholding Cu provision for patients with significant cholestasis or liver disease.[73] On the basis of balance studies, a dose of 0.15 mg/d has been suggested.[92] Doses below this, if continued indefinitely, risk the development of deficiency. This contention is supported by the reports of severe Cu deficiency occurring for patients with cholestasis after Cu has been withheld from PN.[21,22] The risk of deficiency would be expected to be higher if GI losses increase or if there is high Zn provision. To avoid the development of Cu deficiency for patients with cholestasis, it may be preferable to decrease Cu provision rather than to withhold it altogether. Whether decreased or withheld, Cu provision should be kept under close review because individual requirements may change.

Monitoring of Cu Status

Serum Cu should be measured regularly for patients treated with long-term PN and in any patient in whom Cu deficiency is suspected. There are limited data available to guide the frequency of measurements, the recommendations being based on expert opinion. ESPEN guidelines on home PN recommend measuring serum Cu every 6 months.[122] The frequency of measurements should be increased for patients who are clinically unstable.[123] A recent study of TE monitoring among critically ill patients observed that significant cost savings could be made by targeting the sickest patients for monitoring, as opposed to automatic testing of all patients.[124] For patients with cholestasis supplemented with Cu provided by a standard MTE product, 6-monthly monitoring of Cu should suffice,[16] but 3-monthly monitoring has been recommended for patients with increased total bilirubin attributed to liver disease.[125] For patients treated with PN from which supplemental Cu has been withheld, monthly monitoring has been recommended to facilitate early detection of Cu deficiency.[16,125]

Serum Cu results should be considered in the clinical context and along with the results of other investigations. Factors to consider in the practical assessment of Cu status are summarized in Table 2. Causes and features of Cu deficiency should be sought and C-reactive protein measured to assess the APR.[113] If Cu deficiency is suspected, a full blood picture should be ordered, to exclude hematologic features of deficiency, and a trial of supplementation considered.[22,72] Resolution of features in response to supplementation may help to confirm the diagnosis. Serum Zn should be measured during long-term PN to exclude Zn excess. Zn excess can also

be excluded by 24-hour urine Zn <19 μmol.[40] Vitamin B_{12} status should be assessed, especially after gastric surgery. Its deficiency may coexist with that of Cu, as can deficiencies of Fe and vitamins D and E. If Cu accumulation is suspected, features of cholestatic liver disease should be sought and liver function tests measured. Liver biopsy may be considered to measure the liver Cu concentration.

Cu Contamination

Contamination of PN with Cu may result in excessive delivery of Cu. One study that examined 8 component solutions observed that Cu was 1 of 12 TEs present in amounts >1 μg/L in every solution.[126] Cu was a minor contaminant present in PN at a final concentration of 82 μg/2L. Cu was present as a contaminant in the amino acid solutions and sterile water but not in potassium chloride, sodium chloride, and calcium gluconate solutions. More recently, Cu was reported as a contaminant from 5 of 14 PN components undeclared on the product label.[127] The actual amount of Cu was estimated to exceed the prescribed amount by 7%–426%. The total Cu contamination of a PN regimen depends on the volume of individual components added and has been reported to range from 0.1–0.4 mg/d.[15] Patients treated with PN may receive Cu in other IV fluids, causing the total amount delivered to greatly exceed the amount prescribed. Contamination is highest in blood products such as packed red blood cells and frozen plasma and in albumin solutions (0.5 mg/L) and crystalloids (0.14 mg/L).[128] Berger and Cavadini estimated that critically ill patients, with burns or trauma who were treated with large volumes of these fluids, received Cu doses 2.3 times the RDA.[128]

The inadvertent delivery of Cu raises safety concerns. Whether safety is compromised depends on the individual clinical circumstances and on the amount of Cu delivered. The concerns are greatest for patients with cholestasis who are treated with long-term PN. ASPEN has recommended that Cu contamination of composite PN regimens delivered to adults should not exceed 0.1 mg/d.[73] To achieve this target, all components of PN should be considered.

Stability Considerations

Unwanted interactions among components of PN can result in precipitation or degradation of micronutrients. Cu has been reported to interact with cysteine to form precipitates that are trapped by the filter, thereby decreasing the bioavailability of both nutrients. The mechanism of precipitation is uncertain, but spectroscopic examination of precipitates recently identified Cu and sulfur as the main elements.[129] Cu may react directly with cysteine to form Cu cysteinate. Alternatively, it may react with hydrogen sulfide, formed from cysteine by heat sterilization, to form Cu sulfide.[130] The probability of precipitation is highest at high concentrations of both nutrients, the Cu concentration in one reported case being 170 μg/L.[131] It may also be influenced by the timing of additions. When cysteine was added to PN immediately before infusion, no significant differences were observed between prefilter and postfilter concentrations of Cu and cysteine, nor was there visible precipitation.[132] The authors concluded that L-cysteine added to PN immediately before infusion is stable over 24 hours of infusion. This interaction can be prevented by omission of Cu from PN, but this is impractical. Instead, limiting the Cu concentration to 157 μg/L with the use of low-pH cysteine-containing amino acid solutions has been suggested.[133] When Cu requirements are high, it may be necessary to deliver some or all of the Cu by a separate IV infusion. The feasibility of providing Cu enterally should also be considered.

Ascorbic acid is the biologically active form of vitamin C. It is an unstable component of PN, being reversibly oxidized anaerobically to dehydroascorbic acid that, in the presence of oxygen and Cu, is irreversibly oxidized to inactive diketogulonic acid.[134] This is then further oxidized to oxalate. The oxygen for this reaction reaches PN either in component solutions or by permeation through the wall of the bag. Ascorbate can be protected during storage of PN by using multilayered bags that are less permeable to oxygen.[135,136] As new PN regimens are developed, the potential for interaction of components with Cu should be considered.

MTE Products

It is difficult for prescribers to comply with the current ASPEN recommendations on Cu provision in PN because of the formulation of most MTE products currently available in the United States and Europe. These products provide up to twice the recommended amount of Cu, which exceeds most patients' requirements, and a recent review concluded that they were potentially toxic.[73] Moreover, the formulation of these products is not conducive to individualization of Cu dosing. When prescribers wish to decrease or withhold Cu, the MTE product must be withheld and the PN regimen supplemented individually with the necessary doses of Cu, selenium, and Zn. When Cu provision is being increased, it is not safe to increase the MTE dosage, because this could result in excessive provision of manganese and other TEs. In this situation, the PN regimen can be supplemented with an individual Cu product, unless stability considerations demand that the Cu be delivered by a separate IV infusion. These approaches are costly and labor intensive and increase the risk of errors.

ASPEN has made a call to action to bring safer products to the market, recommending that Cu doses provided by adult parenteral MTE products are decreased to 0.3–0.5 mg/d.[137] Similarly, the Australasian Society for Parenteral and Enteral Nutrition has recommended that Cu dosing in MTE products, for the Australian and New Zealand market, be decreased to 315 μg/d.[113] In addition, there may be a place for low-Cu MTE products for use among patients whose requirements are below the standard recommendations. Ideally, a range of products should

be developed with doses of Cu and other TEs appropriate for situations commonly encountered in practice. The availability of such products would greatly facilitate individualization of Cu provision. There is also a need for pharmaceutic companies to routinely provide information on Cu contamination—in PN products and other IV fluids as well. The availability of this information would inform decisions on supplementation.

ASPEN has provided guidance on managing product shortages,[26,138] summarized briefly here. Most important, supplies should be reserved for the most vulnerable patients—namely, those with existing deficiency or at high risk of developing deficiency if micronutrients are withheld. TEs should be supplemented orally for patients with sufficient GI absorptive capacity. Rationing may be necessary—for example, by delivering standard doses 3 times weekly rather than daily or by providing daily delivery of half the standard dose. If rationing is necessary, clinicians should be alert to possible deficiencies and should monitor patients accordingly. Supplies should be sought for micronutrients, including Cu, for which deficiency is likely to occur if the micronutrient is withheld. Advice on strategic planning for future shortages is available elsewhere.[139]

Future Directions

Prescribers of micronutrients should aspire to individualize provision. However, much research and development will be necessary before true individualization of Cu provision is possible. More information is required on the Cu provision required to maintain optimal status in different diseases, especially critical illness. Sensitive and specific biomarkers of Cu status will need to be developed, enabling mild derangements in Cu status to be detected and provision to be adjusted before the onset of clinical features. It is likely that new tests will emerge from studies using *-omics* technologies or from studies of the effects of Cu deficiency on physiologic systems such as the immune system. The availability of better tests of Cu status will decrease the uncertainty that affects decisions on Cu provision in PN. It will also facilitate the study of marginal Cu deficiency in hospitalized patients and in the general population. There is an urgent need for a range of appropriately formulated MTE products to be developed, both to enable compliance with the 2012 ASPEN recommendations and to facilitate adjustment of parenteral Cu doses.

Statement of Authorship

C. Livingstone conceived and drafted this article, gave final approval, and agrees to be accountable for all aspects of work ensuring integrity and accuracy.

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