2022

Estimation of 24-hour Urinary Creatinine Excretion through the Development of a Model and Its Relationship to Outcomes in Hospitalized Critically Ill Veterans

Lynn Diane Hiller
University of North Florida, hillerld@verizon.net

Estimation of 24-hour Urinary Creatinine Excretion through the Development of a Model and Its Relationship to Outcomes in Hospitalized Critically Ill Veterans

By
Lynn D. Hiller

DCN DISSERTATION

Submitted in partial fulfillment of the requirements of
the degree of Doctorate in Clinical Nutrition
University of North Florida

DISSERTATION COMMITTEE

Lauri Wright, PhD, RDN, LD/N, FAND
Associate Professor, Chair, Department of Nutrition and Dietetics
University of North Florida

Philip Foulis, MD, MPH
Department of Laboratory and Pathology Service
James A. Haley Veterans Hospital
Tampa Florida

Susan Goldsmith MD
Department of Gastroenterology
James A. Haley Veterans Hospital
Tampa Florida

James Epps, PhD
Associate Professor, Department of Mental Health Law and Policy
University of South Florida
Tampa Florida

Acknowledgements

First, I would like to recognize Herta Spencer, MD, a pioneer in nutrition research whom I had the honor to work with early in my career. She still inspires me all these years later.

Second, I would like to thank Jeff Fabri, MD, PhD, who gave me his unending support, made me believe I could do anything I desired and answered every question I could dream up.

I am immensely grateful for my family, who has patiently supported me.

Lastly, I want to thank each of my committee members for their time and contributions to this project. They have provided wise guidance and made this project better.

Table of Contents

List of Tables

List of Figures

Abstract

Background: Muscle mass has been found to be highly correlated with patient outcomes. Techniques to identify patients with low muscularity include computed tomography (CT) and bioelectrical impendence analysis (BIA) however disadvantages of cost, exposure to radiation and limited access make these measurements unavailable to the average dietitian. Urinary creatinine excretion (UCE) and the subsequent estimation of creatinine height index (CHI) have been strongly associated with muscularity and outcomes, however, these require a 24-hour urine collection. The postulation that UCE may be predicted from patient variables, through mathematical modeling, would avoid the need for a 24-hour urine collection and may be clinically useful.

Methods: Input variables of age, height, weight, gender, plasma creatinine, urea nitrogen, glucose, sodium, potassium, chloride, carbon dioxide, and magnesium, from a deidentified data set of 967 patients who had UCE measured in the previous 5 years, were used to develop models to predict UCE. The model identified with the best predictive ability was validated using four-fold cross validation and using a separate data set not used to construct the model. Model predicted UCE and CHI were compared to measures of muscularity. The model was then retrospectively applied to a convenience sample of 120 critically ill veterans to examine degree of low muscle mass observed in the cohort and if UCE and CHI were associated with outcomes in hospitalized veterans.

Results: A model to estimate UCE was identified utilizing the input variables of plasma creatinine, plasma BUN, age and weight which was found to be highly correlated, moderately predictive of UCE and statistically significant. Model predicted UCE was found to be highly correlated with accepted measures of muscularity. Applying the model to a cohort of subjects identified that 44.2% of the subjects had CHI levels $\leq$ 60% and were considered to have severe sarcopenia. Subjects with model estimated CHI $\leq$ 60% were found to have significantly lower body weight, BMI, plasma creatinine, albumin and prealbumin levels. Subjects with CHI $\leq$ 60% were found to be 8.0 times more likely to be diagnosed with malnutrition and 2.6 times more likely to be readmitted in 6 months. Subjects with low CHI trended towards longer hospital and ICU LOS, however it did not meet statistical significance. The NUTRIC score was found to have no relationship with the presence of malnutrition.

Conclusion: The development of a model which predicts UCE and correlates with muscle mass offers a novel method for the RDN to readily identify patients with sarcopenia on hospital admission. This method could allow the RDN to quickly screen new admissions for potential sarcopenia without the use of CT or DEXA scans and without the inconvenience of a 24-hour urine collection by using readily available patient variables

Introduction

Consensus guidelines from the Society of Critical Care Medicine (SCCM) and the American Society of Parenteral and Enteral Nutrition (ASPEN) have recommended that measurement of actual energy expenditure, through the use of indirect calorimetry, be used to determine energy requirements.[1] Actual measurement allows for individualized and targeted energy goals for the patient.[1] Similarly, The European Society for Parenteral and Enteral Nutrition (ESPEN) recommends that energy intake be determined using indirect calorimetry if the patient is mechanically ventilated or, if calorimetry is not available, to use oxygen consumption, VO2, from a pulmonary arterial catheter or carbon dioxide output, VCO2, derived from a ventilator, to determine needs.[2]

In contrast, for protein intake, the SCCM/ASPEN guidelines acknowledge that although protein needs in the critically ill appear to be higher than previously thought, determining individual requirements is difficult. As a result, the current recommendations are to provide between 1.2 to 2.0 g protein/kg actual body weight per day.[1] The protein recommendation from ESPEN is that 1.3g protein/kg per day can be delivered progressively.[2] These stark differences between energy and protein recommendations underscore the dilemma facing the practicing clinician. Calorie recommendations are based off of actual measurement of patient specific parameters whereas protein recommendations are based off of weight without any compensation for metabolic state, severity of illness or underlying muscularity. The range of protein intake, as recommended in the SCCM/ASPEN guidelines, when calculated for an individual patient are so wide and non-specific that they are not clinically useful.

Previous studies attempted to identify protein requirements in the critically ill. Although protein intake, when provided above maintenance amounts, has been found to be beneficial,

studies have failed to consistently identify the protein intake most associated with improved outcomes.[3–8] Although positive retrospective or observational studies have identified associations between higher protein intake and improved outcomes, these findings have been unable to be confirmed in randomized control trials (RCT).[9–11] It has been suggested that lack of positive findings and failure to show effectiveness of nutrition therapy in these studies may be due to their failure to control for patient population type, the suitability of included patient types or metabolic state.[12,13] Several studies have observed that targeted nutrition therapy appears to be the most beneficial in only patient specific groups such as those at high nutritional risk, with malnutrition or with low muscularity.[14–18]

Chapter 1: Significance/Literature Review

Importance of skeletal muscle

It is well recognized that skeletal muscle has significant purpose beyond that of its structural functions. Skeletal muscle is an important metabolically active and homeostatic organ, accounts for approximately 50% of all body protein and plays key roles in immune function, glucose metabolism and protein synthesis.[19,20] Oft forgotten, muscle plays a pivotal role in whole-body protein metabolism, which is of particular importance during acute stress or illness.[21] All organs and tissues undergo protein turnover in which the rate of protein breakdown is balanced with the synthesis of new proteins. In the fed state, amino acids from food stuffs supply the needed precursors for new protein synthesis. Under normal conditions, ingested amino acids are incorporated into muscle to replace the amino acids which were lost during fasting, with the overall result being that the gains achieved are balanced with the losses. When nutrient intake is insufficient, muscle protein becomes the principal reservoir of needed amino

acids for all other organs and tissues.[21] Normal protein turnover and maintenance of other essential organs and tissues can continue provided adequate muscle mass is available for cannibalism.[21]

In the setting of acute illness, requirements for amino acids, intermediate metabolites and minerals from skeletal muscle increases due to increased synthesis of acute phase proteins, synthesis of protein components of the immune system and synthesis of proteins necessary for wound healing.[12,20–22] Conversely, the anabolic response to feeding is impaired and loss of lean body mass may not be reversible by nutrition support.[23,24] Mediators of this process are not entirely clear, however the counter-regulatory hormones, glucagon, catecholamines and glucocorticoids are generally elevated in the critically ill and stimulate amino acid catabolism.[23] In critically ill patients, directly correlated with the severity of the injury, increases in proinflammatory cytokines, glucocorticoids and oxidative stress reinforce the effect of catabolic hormones and contribute to muscle wasting.[24] Critical illness thus initiates a cascade of events that lead to accelerated protein degradation, decreased rate of synthesis of selected proteins and increased catabolism and nitrogen loss.[23]

The overall consequence of this rapid body protein remodeling is metabolic imbalance in which "net muscle protein loss greatly exceeds the gain of protein elsewhere and whole-body nitrogen balance becomes strongly negative".[22] Additionally, in the acute care setting, when the calorie provision is insufficient, amino acids from skeletal muscle are broken down to provide the body with energy by way of gluconeogenesis.[19]

Thus, acute illness results in a large protein loss from the body over a short period of hospitalization. Studies have further shown that this is largely muscle mass and clinically significant losses have been quantified.[13,25] Furthermore, immobility associated with

hospitalization causes atrophy of skeletal muscle and additionally contributes to protein loss.[23] These net losses ultimately impair the immune response and may increase morbidity and mortality of the patient.[23] It has been suggested that poor outcomes may occur when the increased amino acid uptake, to respond to the injury, is limited by both the lack of adequate exogenous protein provision and inadequate underlying muscle mass available to release amino acids.[22] Experts have proposed that providing sufficient exogenous amino acids could improve outcomes by "increasing central protein synthesis, optimizing the inflammatory response, mitigating the loss of muscle protein, and mitigating muscle atrophy".[22]

As studies have observed that targeted protein intake appears to be the most beneficial in only patient specific groups such as those at high nutritional risk, with malnutrition or with low muscularity, it appears that nutrition risk is related to outcomes in the acutely ill.[14–18] These findings, coupled with the recognition of the large protein losses and protein redistribution that occurs in acute illness, suggest that for personalized nutrition therapy to be beneficial, the patient with low muscularity or with malnutrition needs to be identified.[13,15,16,26] The aims of nutritional therapy in the critically or acutely ill patient are thus to blunt the loss of lean body mass (LBM) or restore body protein mass and to provide adequate protein and energy.[4,24]

As low muscle mass has been found to be predictive of outcomes, it may also be an important determinant of protein requirements.[27] Measurement and assessment of LBM may thus provide important insights into patient's protein needs as those who have low muscularity will have less to cannibalize and will require greater exogenous provision.[28] Monitoring muscle mass may additionally provide a method to evaluate the effect and timing of the prescribed nutrition interventions. Unfortunately, practitioners generally rely on readily available measures, such as weight or body mass index (BMI), which do not relate to underlying muscularity, and may

ultimately prevent the clinician from directing interventions towards preserving or restoring muscle mass.[29]

Widely used nutrition screening criteria and earlier diagnostic criteria for malnutrition have not included evaluation of muscle within their definition and instead focus on BMI and weight loss.[30] This results in failure to identify low muscle mass and leads to emphasis on energy provision and weight maintenance rather than protein intake and targeting muscle.[19] In contrast, both the AND/ASPEN and GLIM malnutrition criteria include identification of reduced muscle as a key diagnostic criteria.[31,32]

Recent recommendations have been made in an attempt to shift the focus of nutrition into managing and reducing the loss of muscle mass to improve patient clinical outcomes.[19] These recommendations propose that muscle mass should be at the core of nutrition management strategies. Patients should be screened for low muscle mass, and tools and techniques should be used to directly assess muscle mass within the nutritional assessment. Another key recommendation is that nutrition intervention should be optimized to focus on muscle rather than weight. To achieve these goals, the authors suggest that a change in focus and practice is required in how nutrition clinicians screen, assess and treat patients.[19]

Studies which have examined the relationship between muscularity and outcomes

Low muscle mass, as measured by a variety of methods, has been found to be associated with adverse outcomes in the critically ill and other hospitalized inpatient populations, Table 1. Studies have observed significant relationships between low muscle mass, or loss of muscle mass and in-hospital mortality, 30-day, 60-day, or 6-month mortality, higher disease severity scores,

hospital length of stay (LOS), pneumonia, respiratory failure, need for re-intubation and organ failure.[17,25,28,33–42]

A recent retrospective study of 279 intensive care unit (ICU) patients who had computed tomography (CT) scans was conducted to examine if low muscle mass, as determined from CT scans, was associated with outcomes.[36] They observed that 68% of the population was sarcopenic and that low muscle mass was significantly associated with older age, $p < .001$, more comorbidities, $p = .009$, and longer hospital LOS, $p = .043$. Importantly, on logistic regression, 30-day mortality was found to be strongly associated with low muscle area (OR 0.98, $p = .004$).

Loosen et al[37] conducted an exploratory observational study of 155 patients admitted to the medical ICU who had CT scans on admission. They calculated skeletal muscle area to identify sarcopenia and mean skeletal muscle attenuation (MMA) to determine muscular fat deposition, myosteatosis, and then explored if these parameters were related to outcomes. They identified no differences in short term outcome, of ICU survival, between those with or without sarcopenia or between those with or without myosteatosis. However, they did observe significant differences in 6-month and 1-year survival. Patients with low skeletal muscle or high amounts of fatty infiltration of muscle were more likely to have died at 6-months or 1-year time points. Binary logistic regression analysis revealed both low muscle mass (OR, 0.979, $p = .025$) and fatty infiltration (OR, 0.964, $p = .014$) as prognostic of 6-month mortality.

Jaitovitch et al[38] conducted a prospective, observational study of 423 subjects who had been admitted to the medical ICU and who had a CT scan within the first 24-hours of admission. They sought to determine if muscle mass and fat mass, as identified on CT scan, were associated with survival and disability at hospital discharge. Disability was defined as discharged to a facility or home with assistance compared to discharged home without assistance. They observed

that larger muscle mass was significantly associated with decreased odds of mortality at 6-months (OR 0.96 per cm^2 increase in mass, CI 0.94-0.97, $p < .001$) and with decreased odds of disability (OR 0.98 per cm^2 increase in mass, CI 0.96-0.99, $p = .012$). There was no statistically significant association between fat mass and survival, or disability observed. Larger muscle area was the only clinical parameter that remained significantly associated with survival after multivariable adjustments.

Looijard et al[17] conducted a retrospective database study of adult patients admitted to a mixed medical surgical ICU who were on ventilators and receiving enteral tube feedings. They examined data from 739 patients who had CT scans shorty after admission which they used to identify low muscle mass. Patients were divided into 3 groups: normal skeletal muscle area (SMA), low SMA and combined low SMA with low skeletal muscle density (SMD) based on CT scans. Patients were additionally stratified based on whether they received <1.2 or >1.2g protein/kg/day. The researchers sought to examine whether muscle mass and quantity of protein intake is associated with outcomes of 60-day and 6-month mortality. They observed a high prevalence of both low SMA and low SMD, 445 (60%) were found with low SMA, of those 200 (45% of the low SMA group) were found to have low SMA and low SMD. Patients with low SMA were older, $p < .001$, lower weight, $p < .001$, and had higher APACHE II scores, $p < .001$ than those with normal SMA. Low SMA group and Low SMA and SMD groups had higher 60-day and 6-month mortality, all $p < .001$, compared to normal SMA. In the combined low SMA and Low SMD group, 60-day and 6-month mortality were significantly lower in those that received protein intake >1.2g/kg/day, $p < .001$. The authors concluded that optimal nutrition strategies may differ between patients with certain sub-groups benefitting while others do not.

Their study suggests that patients with low SMA and low SMD may benefit from early higher protein intake.

Imamura et al[39] retrospectively examined a group of adults who underwent emergency surgery for colonic perforations who had CT scans the day of surgery. They further stratified patients by age into <75 years or ≥75 years old. They examined if there was a correlation between psoas muscle mass and LOS. A total of 46 subjects met criteria. The older group had significantly lower psoas muscle area, 961mm^2 vs 1622mm^2, $p < .001$, compared to the younger group. A significant negative correlation, $r = .23$, $p = .02$, was observed between LOS and psoas area in the younger group but not in the older group. They suggest that the older group may have generalized sarcopenia however many elderly transferred to other facilities for post-op care thus true LOS may be underestimated.

Fuchs et al[40] conducted a prospective observational study of 231 patients in medical and surgical ICUs who were recently extubated. They examined the effect of low skeletal muscle as determined from the skeletal muscle index, SMI, measured from CT scans on outcomes at 30 days after extubation. They observed that patients with low SMI had significantly greater rates of pneumonia, $p < .001$, 30-day mortality, $p = .004$, reintubation within 72 hours, $p < .02$, and respiratory failure within 30-days of extubation, $p < .02$ when compared to those with normal SMI. Low skeletal muscle was found to be a strong predictor of pneumonia, OR 0.96, $p < .002$, and 30-day mortality, OR 0.94, $p < .03$.

Abramowitz et al[41] conducted a retrospective data base analysis of NHANES data of 11,687 adult subjects who had dual energy x-ray absorptiometry (DEXA) scan results. They included subjects with BMIs between 18.5 and 40. They sought to determine if there was a

Table 1. Studies Examining Relationship of Muscle Mass on Outcomes

Study	Subject Type and Number	Design	Observation or Intervention and Duration	Results	Notes
Joyce 2020	Mixed med surg ICU; 279 subjects	Retrospective chart review	Identified low muscle mass based on CT scans. Examined whether low muscle mass was associated with outcomes.	189/279 (68%) were identified as being sarcopenic. Those with sarcopenia were significantly older, $p<0.001$, had more comorbidities, $p<0.009$, and hospital LOS, $p<0.04$. Using logistic regression, 30-day mortality was associated with low muscle area (OR 0.98, $p = 0.004$).	Baseline sarcopenia was highly prevalent.
Loosen 2020	Medical ICU; 155 subjects	Exploratory observational study	Examined CT scans on ICU admission and calculated skeletal muscle area to determine sarcopenia and mean skeletal muscle attenuation (MMA) to determine muscular fat deposition. Explored if these parameters were related to outcomes.	No differences in ICU survival between those with or without sarcopenia or between those with or without myosteatosis. Observed patients with low skeletal muscle or high amounts of fatty infiltration of muscle were more likely to have died at 6-months or 1-year time points. Binary logistic regression analysis revealed both low muscle mass (OR, 0.979, $p=0.025$) and fatty infiltration (OR, 0.0.964, $p=0.014$) as prognostic of 6-month mortality.	Unclear role of muscle quality versus muscle quantity. They may play equally important roles.
Jaitovich 2020	Medical ICU; 423 patients	Prospective observational study	Evaluated CT scans obtained within the first 24-hours of ICU admission. Determined if muscle mass, fat mass and bone density, as identified on CT scan, were associated with survival and disability at hospital discharge	Larger muscle mass was significantly associated with decreased odds of mortality at 6-months (OR 0.96 per cm^2 increase in mass, CI 0.94-0.97, $p< 0.001$) and with decreased odds of disability (OR 0.98 per cm^2 increase in mass, CI 0.96-0.99, $p=0.012$). There was no statistically significant association between fat mass and survival, or disability observed.	Muscle area was the only clinical parameter that remained significantly associated with survival after multivariable adjustments.
Looijard 2020	Mixed med-surg ICU; on ventilators; 739 subjects	Retrospective Database search	Patients were divided into 3 groups: normal skeletal muscle area (SMA), low SMA and combine low SMA with low skeletal muscle density (SMD) based on CT scans. Patients were stratified based on whether they	445 (60%) were found with low SMA, of those 200 (45% of the low SMA group) were found to have low SMA and low SMD. Patients with low SMA were older, $p<.001$, lower weight, $p<.001$, and had higher APACHE II scores, $p<.001$ than those with normal SMA. Low SMA group and Low SMA and SMD groups had higher 60-day and 6-month mortality, all $p<.001$, compared to	Lower muscle mass was associated with worse outcomes. Higher protein intake was associated with lower mortality in those with low muscle mass but not those with normal muscle mass.

			received <1.2 or >1.2g protein/kg/day. Examined whether muscle mass and quantity of protein intake is associated with mortality.	normal SMA. In the combined low SMA and Low SMD group, 60-day and 6-month mortality were significantly lower in those that received protein intake >1.2g/kg/day, p<.001.	
Imamura 2019	Adults who underwent emergency surgery for perforated colons; 46 subjects	Retrospective chart review	Patients had CT scans on day of emergency surgery; stratified patients by age <75 years or >/=75 years old. Examined if there was a correlation between psoas muscle mass and mortality.	46 subjects met criteria. The older group had significantly lower psoas muscle area, p<.0001. A significant negative correlation, r=0.23, p=.02, was observed between LOS and psoas area in the younger group but not in the older group.	Longer LOS associated with low muscle mass only in younger age group. Many elderly transferred to other facility for post-op care thus true LOS may be underestimated.
Fuchs 2018	Adults; mixed med-surg ICU; recently extubated; 231 subjects	Prospective observational	Examined the effect of skeletal muscle measurements from CT scans on outcomes at 30 days after extubation	Pts with low skeletal muscle index had significantly greater rates of pneumonia, p<.0001, 30-day mortality, p<.004, reintubation within 72 hours, p<.02, and respiratory failure within 30-days of extubation, p<.02. Low skeletal muscle was a predictor of pneumonia, OR 0.96, p<.002, and 30-day mortality, OR 0.94, p<.03.	Skeletal muscle is a strong predictor of outcomes
Abramowitz 2018	Adults with BMIs between 18.4 and 40; 11,687 subjects	Retrospective data analysis	Examined data from NHANES to determine if there was a relationship between low muscle mass, as measured by DEXA, and risk of death. Muscle mass was estimated from the appendicular skeletal muscle mass index.	14% of all subject were found to have low muscle mass. Low muscle mass was not limited to those with low percent total body fat (%TBF). During their time frame of interest, 1,819 subjects died. When separated by muscle mass status, within each BMI category the hazard ratio for death was higher for subjects with low muscle mass, p<.001. When examining subjects with preserved muscle mass alone, there was a significantly increased risk of death among those who were obese compared with overweight, HR 1.23, 95% CI 1.04-1.47.	Skeletal muscle mass is a significant mediator and modifier of the relationship of BMI and mortality.
Shibahashi 2017	Adults with sepsis admitted to the ICU, 150 subjects	Retrospective	Lean skeletal muscle mass was estimated from the cross-sectional area of the psoas and paraspinal muscles at L3 vertebrae. Patients were stratified to survived and	Patients which survived had significantly larger muscle mass than those who did not, 43.3 cm^2 vs 36.8 cm^2, p<.001. Skeletal muscle area was found to be an independent predictor of in-hospital mortality, OR 0.94, 95% CI 0.90-0.97, p <.001.	Low skeletal muscle mass is associated with higher in-hospital mortality

			deceased and into 60-80 years and >80 years of age.	When age groups were examined, skeletal muscle area remained significant regardless of age.	
Thibault 2016	Adults admitted to mixed Med-surg ICU, 931 subjects	Multi-site Prospective observational	Obtained BIA measurements on day 1 of ICU admission to estimated fat-free mass. Examined if there was a relationship between BIA measurements and 28-day mortality and disease severity	BIA measures of fat-free mass were significantly lower in non-survivors than survivors, 4.10 vs 4.59, p<.00. Disease severity scores were also significantly worse in those patients with lower fat-free mass, 21.8 vs 17.7, p<.001. Variables significantly associated with 28-day mortality were admitting BIA measurement of fat-free mass, APACHE II score, surgical diagnosis and increasing age. BMI was not found to be a significant variable.	Future studies should be designed to assess whether nutrition interventions could change outcomes
Weijs 2014	Adults admitted to mixed Med-Surg ICU, who had a CT scan within the first 4 days, 240 subjects	Retrospective chart review	Examined the relationship between muscle mass, as determined from CT scans, and BMI on hospital mortality	Low muscle mass was found in 63% of all subjects. Patients with low muscle area had higher hospital mortality, 38.2% vs 12.5%, p<.001. Regression analysis demonstrated that muscle area, sex and APACHE II score where independent predictors of mortality while BMI and diagnosis were not.	Low skeletal muscle mass is an independent predictor of mortality in ventilated patients. BMI appeared to have no impact on mortality.
Moisey 2013	Adult trauma patients with CT scans on admission, 148 subjects	Retrospective chart review	CT scans were used to identify both skeletal muscle and adipose tissue cross-sectional areas. They sought to determine if low muscularity or low adiposity adversely affected outcomes.	Based on BMI alone, 57% were overweight or obese and 7% were underweight. Based on CT scan measurements, 71% of all subjects were sarcopenic. Significantly more patients with sarcopenia died compared to those not sarcopenic, 32% vs 14%, p=.018. After controlling for age, sex and injury severity, low muscle index but not BMI or low albumin, was associated with hospital mortality, OR =0.93, 95% CI 0.87-0.99, p =.025. Sarcopenic patients had significantly lower ventilator-free, p = .004, and ICU-free days, p=.004. No differences were noted in visceral adipose tissue mass between sarcopenic and non-sarcopenic patients. Adipose	BMI poorly identifies low muscle mass and that low muscularity has significant implications on outcomes.

				tissue was not associated with mortality, ventilator-free or ICU-free days.	
Puthucheary 2013	Adults, mixed med-surg ICU, on vents longer than 48 hours and ICU stays longer than 7 days; 63 subjects	Prospective observational study	Performed serial ultrasound measurements of rectus femoris and muscle biopsies on ICU days 1,3,7 and 10. Muscle protein synthesis, breakdown and balance was determined by leucine incorporation using leucine infusions on days 1 and 7	Muscle cross-sectional area decreased from baseline by 17.7%, $p < .001$, by ICU day 10 and the ratio of protein to DNA by 29.5%, $p < .001$. Leg protein breakdown was elevated compared to synthesis, $p = .05$, resulting in a net catabolic balance. Increasing organ failure score was correlated with change in muscle cross sectional area, $r = 0.23$, $p < .001$.	Skeletal muscle wasting occurred early and rapidly in critically ill patients and appeared to be due to both decreased synthesis and increased muscle breakdown.
Gruther 2008	Adults, mixed med-surg ICU; 125 subjects	Prospective observational study	Conducted ultrasound measurements of the rectus femoris and vastus intermedius muscles and estimated muscle layer thickness. Interested in identifying muscle wasting in ICU patients and whether there was a relationship between muscle and length of stay (LOS).	Observed a significant negative correlation between admission muscle layer thickness and LOS, $p = .005$	Muscle wasting was best described by using a logarithmic function

ICU = intensive care unit; Med = medical; Surg = surgical; LOS = length of stay; CT = computed tomography; BMI = body mass index

relationship between low muscle mass, as measured by DEXA, and risk of death at a mean time point of 9 years. Muscle mass was estimated from the appendicular skeletal muscle mass index. They observed that 14.1% of all subjects had low muscle mass and that low muscle mass was not limited to those with low percent total body fat (%TBF). During their time frame of interest, 1,819 subjects died. When separated by muscle mass status, within each BMI category the hazard ration for death was higher for subjects with low muscle mass, $p < .001$. When examining subjects with preserved muscle mass alone, there was a significantly increased risk of death among those who were obese compared with overweight, HR 1.23, 95% CI 1.04-1.47. Skeletal muscle mass was observed to be a significant mediator and modifier of the relationship of BMI and mortality and that muscle mass altered the relationship of BMI with %TBF.

Shibahashi et al[42] conducted a retrospective study of 150 adults > 60 years of age who were admitted to the ICU with sepsis and had a CT scan performed the day of ICU admission. They were interested if whether decreased skeletal muscle mass is associated with mortality. Lean skeletal muscle mass was estimated from the cross-sectional area of the psoas and paraspinal muscles at L3 vertebrae. Patients were stratified into survived and deceased groups and into 60-80 years and >80 years of age groups for analysis. Patients which survived had significantly larger muscle mass than those who did not, 43.3 cm^2 vs 36.8 cm^2, $p < .001$. Skeletal muscle area was found to be an independent predictor of in-hospital mortality, OR 0.94, 95% CI 0.90-0.97, $p < .001$. When age groups were examined, skeletal muscle area remained significant regardless of age. The authors propose that identifying patients with low muscularity may allow for earlier intervention.

Thibault et al[33] conducted a large multi-site prospective observational study of 931patients admitted to mixed medical surgical ICUs who underwent BIA measurements to

assess fat-free mass on day of ICU admission. They were interested in examining if there was a relationship between BIA measurements and 28-day mortality and disease severity, APACHE II score. BIA measures of fat-free mass were significantly lower in non-survivors than survivors, 4.10 vs 4.59, $p < .001$. Disease severity scores were also significantly worse in those patients with lower fat-free mass, 21.8 vs 17.7, $p < .001$. Variables significantly associated with 28-day mortality were admitting BIA measurement of fat-free mass, APACHE II score, surgical diagnosis and increasing age. BMI was not found to be a significant variable. They concluded that fat-free mass measured on ICU admission by BIA is associated with 28-day mortality.

Weijs et al[28] conducted a retrospective chart review of 249 patients who were admitted to a mixed medical-surgical ICU, were on ventilators and who had a CT scan within the first 4 days. They were interested if there was a relationship between muscle mass, as determined by the CT scan, BMI and outcomes. They observed that 63% of all patients had low muscle mass. Patients with low muscle area had higher hospital mortality, 38.2% vs 12.5%, $p < .001$. Regression analysis demonstrated that muscle area, sex and APACHE II score where independent predictors of mortality while BMI and diagnosis were not. When muscle area was examined as a continuous variable and not as a sex related cut-off category, sex disappeared as an independent predictor of mortality.

Moisey et al[34] conducted a retrospective study of 148 adult patients admitted to their trauma unit who had a CT scan on day on admission. CT scans were used to identify both skeletal muscle and adipose tissue cross-sectional areas. They sought to determine if low muscularity or low adiposity adversely affected outcomes. On the basis of BMI alone, 57% were overweight or obese and 7% were underweight. In stark contrast, based on CT scan measurements, 71% of all subjects were sarcopenic. Significantly more patients with sarcopenia

died compared to those not sarcopenic, 32% vs 14%, $p = .018$. After controlling for age, sex and injury severity, low muscle index but not BMI or low albumin, was associated with hospital mortality, OR =0.93, 95% CI 0.87-0.99, $p = .025$. Sarcopenic patients had significantly lower ventilator-free, $p = .004$, and ICU-free days, $p = .004$. No differences were noted in visceral adipose tissue mass between sarcopenic and non-sarcopenic patients. Adipose tissue was not associated with mortality, ventilator-free or ICU-free days. The authors conclude that BMI poorly identifies low muscle mass and that low muscularity has significant implications on outcomes.

Puthucheary et al[25] conducted a prospective observational study on 63 subjects admitted to a mixed medical-surgical ICU who were anticipated to be on a ventilator longer than 48 hours and require ICU stays of greater than 7 days. They were interested in characterizing the time course and pathophysiology of muscle loss in the critically ill. They performed serial ultrasound measurements of rectus femoris and muscle biopsies on ICU days 1,3,7 and 10. Additionally, muscle protein synthesis, breakdown and balance was determined by leucine incorporation using leucine infusions on days 1 and 7. They observed that muscle cross-sectional are decreased from baseline by 17.7%, $p < .001$, by ICU day 10 and the ratio of protein to DNA by 29.5%, $p < .001$. Leg protein breakdown was elevated compared to synthesis, $p = .05$, resulting in a net catabolic balance. Increasing organ failure score was correlated with change in muscle cross sectional area, $r = .23$, $p < .001$. The authors concluded that skeletal muscle wasting occurred early and rapidly in critically ill patients and appeared to be due to both decreased synthesis and increased muscle breakdown and was associated with organ failure.

Gruther et al[35] conducted a prospective observational study on 125 ICU patients. They performed ultrasound measurements of the rectus femoris and vastus intermedius muscles and

estimated muscle layer thickness (MLT). They were interested in identifying muscle wasting in ICU patients and whether there was a relationship between muscle and length of stay (LOS). They observed a significant negative correlation between admission MLT and LOS, $p = .005$. Additionally, they were able to describe muscle wasting using a logarithmic function.

These studies lay the foundation for the growing understanding of the negative outcomes associated with low muscle mass. Outcomes of increased mortality, longer LOS, greater readmission rates and increased disease severity have all been demonstrated. Identifying low muscularity and changes in lean body mass are then essential for the provision of prompt, individualized nutritional therapy and mitigating adverse outcomes.[43] The key step for the practitioner thus lies in how to easily identify these at-risk patients with low muscularity in the hospital setting.

Methods of measuring muscularity in hospitalized patients

Relying on body weight or body mass index (BMI) will fail to identify those with altered body composition, particularly in those who are obese with low muscularity.[43] The assessment of muscle loss is now considered to be a key component of nutrition status.[31,44] Although the clinician may attempt to identify loss of muscle mass through the use of a nutrition focused physical exam or anthropometric measurements, it remains a subjective assessment.[31,45] Lean tissue loss may occur prior to weight loss and may be difficult to discern with visual techniques when there are concomitant changes in fat mass or extracellular water.[44] More sensitive body composition technologies and techniques are available to objectively measure or estimate lean body mass.[43,44,46,47] These techniques are gaining in acceptance and becoming more readily available for use in the hospitalized patient. The most commonly used methods to evaluate body

composition and muscle mass include computed tomography (CT), dual energy x-ray absorptiometry (DEXA), bioelectrical impendence analysis (BIA) and ultrasound (US).[43,44,46,47] Recent guidelines are available to aid the clinician in understanding the validity of some of these various measurements.[46]

Studies which have examined the relationship between muscle mass and outcomes have generally relied on CT scans to identify patients with low muscularity.[43,44,47] CT scans use high-dose radiation to produce cross-sectional images of organs, skeletal muscle, adipose tissue and bone. Several computer programs are available to then analyze the images and make determinants of body composition.[43,47] CT scan measurements at the mid-lumbar, L3, vertebral slice has been used to predict body fat and fat free mass. Cut-off points for low muscularity have been established and thus are considered to provide reference measurements of body composition.[43] Although utilizing CT scans to measure muscle mass is considered a gold standard, it is not always practical.[43,44,47] As CT scans expose the patient to a high dose of radiation, they are not ordered for the purpose of the nutrition assessment. Rather, their nutritional use is limited to those patients who underwent CT scans as part of their medical treatment. Short comings of relying on CT scans are that not all patients undergo CT scans early enough or frequently enough to be useful in identifying malnutrition, muscle loss or sarcopenia. These scans also require that the patient physically go to the radiology department which may be precluded in the unstable patient. Importantly, measurements of muscle mass are not routinely reported as part of CT scan results and these measurements require specialists trained in measuring muscle and body compartments.[43,44,47]

Alternative methods for identifying low muscularity exist, although these have limitations as well.[19,44,46–48] DEXA uses much lower doses of radiation and relies on measured

changes in x-ray attenuation to estimate body composition. As DEXA imaging is safer, it has been utilized to develop normal body composition data which has been extrapolated to develop reference values.[47] DEXA has been found to be a valid technology to assess fat mass in adults, however studies validating lean mass are lacking.[46] DEXA can provide a measure of appendicular lean mass, which is considered an indicator of muscle mass although hydration levels can impact soft tissue readings.[19,48] Currently there is a lack of studies using DEXA in the inpatient setting and it may not be readily accessible in a variety of clinical settings.[19] Lastly, most DEXA imaging currently done only encompasses the lumbar or femoral regions and thus does not include areas needed for the assessment of lean tissue. Whole body DEXA imaging for body composition analysis will have additional costs associated and will require trained personnel to read and interpret the results.[46]

Bioelectrical impendence analysis (BIA) uses low amplitude electrical current to characterize the conductive and non-conductive fluids of the body. Fat and bone are poor conductors of electrical currents compared to water, muscle and blood which are excellent conductors. Electrodes placed on the body can measure the opposition to an electrical current through the body tissues which can then be used to estimate total body water and body composition.[44,46] Advantages to the use of BIA for analysis of body composition include that it is inexpensive, portable, non-invasive and the measurements can be performed relatively quickly. BIA has several drawbacks of which the clinician should be aware of. BIA does not directly measure body composition. However, it provides indirect estimates based on manufacturer specific regression models. These models rely on several key assumptions which include that the body is comprised of 5 cylinders of uniform cross-sectional area. This assumption may be violated in those who are obese, have had an amputation or have shorter or longer than average

limbs. BIA accuracy has also been found to be affected by adiposity, fluid and electrolyte status, skin temperature and ambient temperature.[43,44,46–48] Importantly, regression equations have been generally derived from healthy, normal weight ambulatory patients. The assumptions of these equations may not be true in the critically ill patient with alterations in intracellular and extracellular compartments or in disease states which are characterized by expansion of extracellular water.[43] BIA has also been found to be unreliable in patients receiving electrolyte containing intravenous fluids as the measurement appears to be affected by the ion content of the fluid.[49] It is currently unclear how long it takes to achieve ionic equilibrium after intravenous administration of fluids, thus recommendations for appropriate timing of BIA measurements in those hospitalized who require intravenous fluids cannot be made.[49] For BIA to become more reliable in the acute care setting, resolution of these issues and the development and validation of accurate algorithms in the critically ill or hospitalized patients will be required.[19,43,46,49]

Ultrasound imaging of muscles utilizes high frequency soundwaves to produce images based on the amplitude of the reflected soundwaves.[43,46,47,50] US has been used to assess body composition including visceral and subcutaneous adiposity and skeletal muscle. It has the advantage of examining individual muscle groups and examining both muscle quantity and quality through measures such as muscle thickness, cross sectional area and echogenicity. Muscle thickness can be measured at multiple sites and equations have been proposed to predict fat-free mass or skeletal muscle mass.[44] These measurements have been used to identify sarcopenia, malnutrition and muscle loss. The most commonly measured muscle in the critical care setting has been the quadriceps as it is readily accessible and has easy to identify fascial borders. US of the quadriceps has been used to predict whole body muscle mass and this technique has been shown to provide reasonable estimates of muscle mass compared to other

reference techniques.[47,51] Musculoskeletal US is gaining in popularity, as it is less expensive, less invasive and more portable compared to CT scans. These advantages allow the clinician to obtain repeat measures and follow changes in muscle mass. For US use to become more widely accepted, expert consensus on choice of muscle groups or anatomic site, degree of compression and use of muscle thickness or cross-sectional area as the measurement standard will need to be determined.[52,53] Lastly, although sarcopenic cut-offs have been proposed to identify low muscularity, further validation is required.[35,43,46,47,50]

Using creatinine to evaluate muscularity in hospitalized patients

Although these measurement techniques can provide valuable information, disadvantages of cost, exposure to radiation, lack of standardization of procedures, need to transport the patient to the radiology department, lack of mathematical models for specific patient populations, lack of trained personnel and limited access to techniques and equipment will likely make these measurements infrequently available to the average clinician.[54] A widely available and long-accepted method for evaluating muscularity is by use of plasma creatinine levels and the 24-hour urinary creatinine excretion method.[54–57] Plasma creatinine levels and measurement of 24-hour urinary creatinine excretion provide viable alternatives and avoid the disadvantages that occur with the other measurement techniques.

Creatine is a nitrogenous organic compound which participates in cellular energy metabolism and is found primarily in muscle. Creatine production requires several steps. The first step involves the synthesis of guanidoacetate from glycine and arginine and requires the rate limiting enzyme glycine amidinotransferase in the kidney.[55,58] Then creatine is formed by the transfer of s-methyl group from *S*-adenosylmethionine to guanidoacetate in the liver. This step is

irreversible and not rate limiting. This synthesized creatine is then released into the circulation where the next step is active uptake against a concentration gradient by muscle. This active uptake of creatine results in the replacement of about 2% of the total amount of creatine in muscle each day. As creatine uptake by muscles is relatively complete, blood levels remain low. Within the muscle, creatine exists in two forms, creatine and creatine phosphate. Creatine is continuously dehydrated by a nonenzymatic reaction to creatinine at a constant turnover rate.[57,58] Once formed, creatinine then diffuses from the cell, is transported by the blood and ultimately appears in the urine after glomerular filtration. In the healthy state and on a stable diet, creatinine output is constant on a day to day basis for the individual.[55,58]

Creatinine is the sole metabolite of creatine and once formed undergoes renal excretion at a constant rate.[55] Since 98% of creatine is located in striated muscle, skeletal and cardiac muscle, the amount of creatinine produced by the body varies directly and proportionally with the muscle mass. It has been identified that 17.9g of muscle produces 1g of urinary creatinine every 24 hours, thus muscle mass can be predicted from 24-hour urinary creatinine excretion.[56] Additionally, since muscle creatinine concentration is 3-5g/kg of wet fat-free tissue, muscle mass can be reliably estimated.[57] Due to these well identified processes and relationships, evaluation of serum creatinine levels and urinary creatinine excretion can be used to estimate muscle mass by validated equations.[54,55] The ratio of total body muscle mass and 24-hour urinary creatinine excretion approximates a constant commonly referred to as the creatinine equivalence.[54]

Researchers have confirmed strong correlations between plasma creatinine levels or urinary excretion of creatinine and lean body mass. Schutte et al[56] observed that total plasma creatinine is a linear function of striated muscle mass and the two variables are strongly correlated. They observed that plasma creatinine and urinary creatinine excretion were also

strongly correlated, $r = .82$, $p < .001$, and that plasma creatinine was strongly correlated with lean body mass, $r = .72$, $p < .001$.[56] Urinary excretion of creatinine has also independently been found to be strongly correlated, $r = .98$, with LBM as measure by K^{40} counting.[59] Welle et al[60] also demonstrated that creatinine excretion was closely associated, $r = .93$, with LBM as determined by K^{40} counting and that the relationship was not affected by age or sex. Urinary creatinine excretion was also found to be highly correlated with fat-free mass (FFM) as measured by densitometry and deuterium dilution, $r = .89$, $p < .001$ and considered a good predictor of FFM.[61] Heymsfield et al[55] demonstrated that muscle mass, as estimated from mid-arm muscle area calculated from arm circumference and triceps skinfold, was strongly correlated with urinary creatinine excretion, $r = .94$, $p < .001$.

Proctor et al[62] compared DEXA and urinary creatinine excretion and compared those methods to known standards of underwater weighing and total body water as estimated with deuterium oxide dilution. They found that DEXA and urinary creatinine excretion did not detect similar differences in total body skeletal muscle mass as a function of age. DEXA was not as sensitive as urinary creatinine in identifying age related changes in muscle mass and that the disparity between the two methods was accounted for by differences in total body water. They observed that the creatinine excretion method provided a better estimate of age-related muscle loss than DEXA and that the decline in muscle strength observed with the older subjects corresponded with declines in muscle mass. Higher DEXA-based muscle mass estimates in older subjects appeared to be due to age-related increases in body water content which in turn was overestimated as lean tissue by DEXA.[62]

The clinician should be aware of certain metabolic or medical conditions that can alter creatinine output and in those instances, creatinine should not be used to estimate muscle mass.

Factors that can lead to elevated plasma creatinine concentration render the results unreliable and include muscle injury, intense exercise and kidney disease.[55,56] Renal disease, which results in declines in glomerular filtration rate, will impact creatinine excretion. As glomerular filtration falls, urinary creatinine excretion falls, and the creatinine output is no longer proportional to muscle mass.[55]

Lastly, nutritional intake may affect creatine production and creatinine excretion and may make results more difficult to interpret.[55] Urinary creatinine excretion is influenced by three dietary constituents: protein, creatine, and creatinine. Dietary protein is the main source of the amino acid precursors of creatine and the level of protein intake has been noted to have a small effect on urinary creatinine excretion. Providing nutrition supplements with increased amounts of arginine and glycine, the two dietary amino acid precursors of creatine, may enhance transamidinase activity and result in higher creatine production. Dietary creatine intake, from meat, increases the size of the creatine pool which then proportionally increases the output of creatinine in the urine. Therefore, when using creatinine excretion to assess muscle mass it is important to control as many of these dietary factors as possible.[55]

Due to the correlation between plasma creatinine levels, urinary creatinine excretion and muscle mass, low plasma creatinine levels and low urinary creatinine output can be considered a marker of diminished muscle mass.[20,56] This simple and inexpensive method to identify low muscularity allows assessment without relying on more complex techniques such as CT scans or DEXA.[54,60] Perhaps more importantly, creatinine excretion originates from metabolically active muscle cell mass which may have significant implications and advantages over other estimates of muscle mass.[62]

The relationship between creatinine and outcomes

Researchers have sought to examine the relationship between low plasma creatinine concentration or low urinary creatinine excretion and outcomes. Udy et al[63] conducted a large retrospective study of over 1 million patients admitted to 175 ICUs across Australian and New Zealand. They stratified patients based on their plasma creatinine concentration measured within the first 24 hours of admission. They observed the highest in-hospital mortality in those with the lowest creatinine levels, adjusted OR = 2.03, 95% CI 1.86-2.21 and that this was greater than for those with elevated creatinine levels, adjusted OR = 1.60, 95% CI 1.55-1.66. Additionally, they observed this relationship between in-hospital mortality and plasma creatinine levels across all BMI categories.

Cartin-Ceba et al[64] had previously conducted a retrospective study of 11,291 ICU patients and examined the association between baseline plasma creatinine measured on admission and hospital mortality. For purposes of analysis, they stratified patients into one of four plasma creatinine levels, very low creatinine $\leq$ 0.6mg/dL, low creatinine 0.6-0.8 mg/dl, normal 0.9-1.4mg/dL and high >1.4mg/dL. They observed a U-shaped curve with both low and high creatinine levels associated with unadjusted in-hospital mortality. A similar relationship was observed for ICU length of stay. After multivariate logistic regression analysis was used to control for BMI, age and gender, low baseline creatinine was associated with increased mortality in a dose response manner, OR = 2.59, 95% CI, 1.82-3.61, for creatinine $\leq$ 0.6mg/dL and OR = 1.28, 95% CI, 1.03-1.60, for creatinine 0.6-0.8mg/dL.

Thongprayoon et al[65] conducted a broader study of 73,994 patients admitted to the general hospital population to determine if creatinine level was associated with in-hospital mortality or 1-year mortality. For analysis, baseline plasma creatine was categorized into one of

7 groups. They observed the lowest mortality in those with baseline creatinine levels of 0.7-0.8mg/dL and a U-shaped distribution with the highest mortalities observed in the highest and lowest levels of creatinine. The risk associated with the lowest creatinine level, OR = 3.29, 95% CI, 2.08-5.00, was greater than the risk associated with the highest creatinine level, OR = 2.56, 95% CI, 2.07-3.17. This association remained statistically significant even after adjusting for BMI. Similar findings were observed for 1-year mortality, with patients with the lowest and highest creatinine levels, experiencing the greatest mortality. They concluded that low plasma creatinine value at hospital admission has important prognostic importance for patient outcomes and appears to be a better surrogate marker for low muscle mass than BMI.[65]

Hessels et al[66] examined the relationship between urinary creatinine excretion (UCE) and mortality in 6151 ICU patients who had 24-hour urine collections within the first 3 days of ICU admission. For purposes of analysis UCE was stratified into sex specific quintiles. They observed that in-hospital mortality decreased for sex-specific quintiles of UCE, from 31% in the lowest to 9% in the highest quintile, $p < .001$ of urinary creatinine excretion. In multivariable logistic regression analysis with sex specific quintiles of UCE, they observed a 2.4 times increase risk of in-hospital mortality in the lowest quintile compared to the highest, OR = 2.56, 95% CI 1.96-3.34, $p < .001$. In multivariable logistic regression analysis, UCE expressed as a continuous variable was found to be inversely associated with in-hospital mortality, with every 5mmol/24 hour decrease in UCE, OR= 1.18, 95% CI 1.66-1.97, $p < .001$. Creatinine excretion additionally had the advantage of not being affected by edema, common in the ICU setting, which confounds weight, BMI and anthropometric measures.[66]

Khan et al[67] utilized UCE to identify skeletal muscle loss in critically ill patients who required ICU readmission. Using UCE to estimate skeletal muscle mass, they identified a >47%

reduction in skeletal muscle mass from the first to second ICU stay. All patients in their series met definition for sarcopenia based on muscle mass and all unfortunately expired.

UCE has also been associated with poor outcomes in specific disease states. For example, ter Maaten et al[68] examined 120 clinically stable outpatients with congestive heart failure and sought to determine if a relationship existed between UCE and death, myocardial infarction or heart failure hospitalization. They observed that patients in the lowest tertile of UCE experienced the worst clinical outcomes and that low UCE was associated with more severe heart failure.

These studies demonstrate the strong relationship that has been identified between plasma creatinine levels or UCE and mortality and other outcomes. Plasma creatinine levels and UCE also appear to be strong predictors of poor outcomes independent of BMI. These represent relevant biomarkers to aid the clinician in risk stratification and identifying those with low muscularity who are at the greatest nutritional risk.

The Creatinine Height Index

Although advanced malnutrition or significant loss of muscle mass may be easy to identify via nutrition focused physical exam, for the hospitalized patient, the detection of less visually obvious or more subtle cases is key to prompt nutrition intervention and mitigation of poor outcomes and may single out those patients most likely to benefit from aggressive nutrition. The creatinine height index (CHI), initially developed for children and then later modified for use in adults, has been used for over 40 years. The CHI is a simple method to assess degree of muscle mass deficit and can be thought of as a combined anthropometric and biochemical measure.[69–72] The CHI utilizes the principles of the relationship between UCE and muscle mass and is defined as the observed 24-hour urinary creatinine excretion divided by the amount expected in normal subjects of the same sex and height expressed as a percentage.[69–71] Ideal body

weight, based on height and gender, when multiplied by the standard creatinine coefficient, milligrams of creatinine excreted per day per kilogram body weight, provides the expected amount of creatinine excretion per day. Normal values of UCE by gender and for various heights have been determined and published.[69]

The CHI is thus considered a marker of muscularity and used to detect protein depletion.[57,69–71] A CHI <60% of predicted is defined as severe loss of muscle and related to a midarm muscle circumference of <10th percentile, between 60-80% of predicted as moderate loss, 80-90% as mild loss and >90% as normal.[70,72] CHI has been observed to be more sensitive than other measures of nutrition status.[69] An advantage of CHI is that it is not affected by weight, edema or obesity and its value is not altered by the inflammatory response like albumin or prealbumin.[73] Although less commonly used today, CHI is considered a well-established standard in evaluating malnutrition and muscularity.[57,70–72,74–76]

The relationship between CHI and outcomes:

CHI, as a surrogate for LBM, has been examined in a variety of settings and correlated to outcomes. Datta et al[73] studied 167 patients on ventilators in a long-term acute care facility. They obtained CHI and a variety of other nutritional indices in these patients and then followed them to assess ability to wean from the ventilator. Patients were stratified by the CHI into normal to mild malnutrition, CHI >81%, moderate malnutrition, CHI 61-80%, severe malnutrition, CHI 41-60%, and very severe malnutrition, CHI <40%. They observed that total serum protein, hemoglobin and CHI were significant predictors for successful weaning, with CHI having a stronger statistically significant effect on successful weaning and survival than any other variable, $p = .0002$.

Schwebel et al[77] examined CHI and percent Ideal Body Weight (%IBW) in a group of patients awaiting lung transplantation. They observed that decreased LBM, as identified by CHI, was not always related to %IBW and that low CHI was strongly related to degree of hypoxemia and death while awaiting transplant, $p < .03$. In those that received a transplant, low CHI was also found to be related to prolonged time on vent, $p < .05$, and ICU LOS, $p < .001$.

Medhat et al[78] utilized CHI along with serum albumin level, mid-arm circumference, triceps skinfold, mid-arm muscle circumference, BMI and Subjective Global Assessment (SGA) to evaluate 103 patients with cirrhosis. The researchers observed that SGA identified malnutrition in 92.2% of patients, CHI identified malnutrition in 89.2%, and mid-Arm muscle circumference identified malnutrition in 86.4% of patients. CHI was noted to significantly fall with progression of cirrhosis from Child Pugh Score A, to B to C, $p < .001$. Of note, no patient was identified as being underweight by BMI, with 23% being normal weight and 77% being overweight or obese.

Apelgren et al[79] examined a variety of commonly available nutritional parameters, including albumin, weight for height, and CHI, in a small group of critically ill veterans. They observed that both albumin and CHI had weak negative correlations with duration of ICU LOS, $r = -.36$ and $-.46$ respectively. Additionally, CHI was statistically higher in survivors compared to non-survivors, $p < .05$.[79]

Limitations of using UCE or CHI in the hospitalized patient:

A limitation in the use of urinary creatinine excretion or CHI to assess muscularity is the need for an accurate 24-hour urine collection.[80–82] Twenty-four-hour urine collections, in the hospital setting, can be fraught with problems. These include inconvenience on the part of

nursing or the patient, inaccurate timing and incomplete collections for those patients without urinary catheters. Inaccurate timing refers to when the collection extends beyond the 24-hour mark and thus will over-represent the amount excreted in 24-hours. Incomplete collections may occur due to patient incontinence, nursing accidently discarding part of the sample, or for patients without urinary catheters, forgetting and using the restroom rather than collecting the full sample.[80–82] Although spot urine samples, or time frames less than 24 hours have been utilized, difficulties with reliability persist, likely due to diurnal variations in nitrogen excretion, which have resulted in underestimating actual excretion.[80–82]

Novel solutions to limitations:

To allow wider application of CHI within the clinical setting, it has been proposed that in patients with normal renal function, the urinary creatinine excretion may be estimated from the plasma creatinine level.[83] The proposed estimation is accomplished with the following equation: CC = UV/P, in which CC is normal creatinine clearance of 80ml/minute, UV represents 24-hour urine creatinine, P represents plasma creatinine concentration and one solves for UV. CHI would then be calculated from the estimated urinary creatinine excretion.[83] This proposed manipulation has the additional benefit, in those with stable renal function, of allowing retrospective estimation of CHI and muscularity to examine changes over the course of the hospitalization.[83-85] The estimation of 24-hour urinary creatinine excretion from plasma creatinine level and subsequent estimation of CHI, although having the potential to be clinically useful, has not been modeled or validated to date.

Currently our understanding of protein requirements for critically ill, hospitalized adults and for specific subgroups is inadequate. It appears unrealistic to define protein requirements for

different diagnostic groups at different stages of illness, with varying degrees of nutrition risk and underlying muscularity with one set of recommendations.[13,17,83] It is important to further identify personalized factors, such as low muscle mass and degree of metabolic stress, to allow the clinician to individualize nutrient intake and identify those patients most in need of prompt adequate protein intake.[13,17,83] Although accurate methods exist to detect low muscularity, the identification of readily available, non-invasive biomarkers would allow rapid screening of the majority of hospital admissions and provide a potential method for subsequent longitudinal follow up.[57]

As no readily available biomarkers that screen for low muscularity exist, this project attempted to fill the existing gap by exploring if a robust model could be identified that predicted UCE from plasma creatinine level or other patient variables. This project also explored if model derived UCE and subsequent estimated CHI was related to patient outcomes and other commonly assessed measures of nutrition and metabolic stress.

Chapter 2: Theoretical Framework

Modeling

Mathematical modeling is the process of creating a mathematical model from a data set and is a method in which mathematical terms are used to represent the behavior of real-world functions, and describe their aspects, their interaction and dynamics. In broad terms, models are developed to describe what is seen, explain why results occurred and to predict future results and outcomes.[86–88] Models are a representation of reality in simplified forms. Mathematical models are used to develop scientific understanding, test the effect of changes in a system and aid in decision making. Mathematical modeling is routinely used throughout the disciplines of

medicine and health for widely ranging purposes such as drug metabolism and dosing[89], antibiotic resistance[90], disease diagnosis[91,92], simulation and prediction of organ damage associated with drug or alcohol consumption[93,94], infection control and infectious transmission[95,96] and estimates of mortality[97] to name a few. Nutrition professionals may be familiar with applications of modeling. The Prognostic Nutritional Index uses mathematical modeling to predict operative morbidity or mortality based off 4 nutrition parameters and the Nutrition Risk Index uses modeling to predict risk of post-operative complications based off of 2 nutrition parameters.[98–100]

To better discuss models, it can be considered helpful to identify classification categories between types of models. Models may be classified according to their structure and reflect their output and how they are used. A common classification method is to classify a model as deterministic or stochastic.

Deterministic Models

Deterministic models are designed to have precise outcomes through known relationships and exclude random variation. Thus, a deterministic model will always produce the exact same output when starting from a given initial state because the output of the model is fully determined by parameter values and the initial conditions of the system. It predicts outcomes with 100% certainty. Deterministic modeling is used when no randomness is involved in the development of future states of the system.[86–88] For example, when using linear regression, if deterministic relationships exist between variables, one can predict with 100% certainty where the y-value will be based on the x-value. Ordinary differential equations and regression equations are used in deterministic modeling using powerful computer programs.[86–88,101]

An example of deterministic modeling is one of the many models that was published during the early days of the Covid-19 pandemic.[102] As so little was known about the virus, researchers sought to use modeling to compute the infected population and number of casualties. Carcione and colleagues[102] used a version of the Susceptible-Exposed-Infectious-Removed (SEIR) model to describe the spread of the virus and compute the number of infected, recovered and dead individuals on the basis of number of contacts, probability of disease transmission, incubation period, recovery rate and fatality rate in a region of Northern Italy. The SEIR model predicts a peak of infected and dead as a function of time, assumes births and natural deaths are balanced, and that population decreases due to the disease are dictated by the fatality rate of the disease. The parameters of the SEIR model are per capita birth rate, per capita natural death rate, virus induced average fatality rate, probability of disease transmission, rate of progression from exposed to infectious, and recovery rate of infectious individuals. SEIR is a system of non-linear ordinary differential equations which are solved numerically using computer programs. Specific differential equations exist which govern 4 population classes of susceptible, exposed, infected-infectious and recovered. The researchers first solved the differential equations using published Covid-19 data from Hong Kong and Singapore to develop a Covid-19 model.

Next, they then applied the model to census and health data available from the Lombardy region in Italy. Use of the model allowed them to predict the number of infected individuals, the number who would require ICU admission, the day the peak of infected would occur on and predicted fatality rates. Using the model, they predicted that in less than three weeks the healthcare system in the region would be overwhelmed. The authors conclude that simulating the process of the infection is key to preparedness and to applying control measures.[102]

Deterministic models are not as commonly used in the field of medicine because deterministic modeling only allows the use of integers and does not allow for the random fluctuation that occurs within biological systems. Deterministic models are likely to have less clinical value because clinical cohorts have random variation that are not necessarily found in research settings.[86,101] Due to this, attention will be focused on stochastic models.

Stochastic models

Stochastic models are used to predict the distribution of possible outcomes and are based on random approaches. Stochastic models assume that the dynamics of the system being studied are partly driven by random fluctuations, such as blood glucose levels throughout the day, and because of this, the same set of parameter values and initial conditions can lead to different outcomes.[86–88] Stochastic modeling in medicine considers that each biologic process is a random event that can take place with a certain probability.[101,103] Stochastic modeling is conducted with computer programs utilizing probabilistic equations and analysis of variance.[87,101]

Steps of Model Building

The steps to modeling can be roughly thought of as follows.[88,101,104] First, the investigator needs to identify if the current problem or question is amendable to mathematical modeling. Second, a data source needs to be identified which can be used for modeling purposes. In medical research this data may come from the electronic medical record (EMR), FDA drug databases or from state or federal agencies such as the health department or CDC to name a few examples.

Next, the obtained data set is randomly divided into sets. A common approach is to divide the data into three subsets. A training set, to build the model, contains approximately 80% of the data. A test set of 10% of the data is set aside to select the best model from among the models developed. The validation set, which contains the remaining 10% of the data is then set aside for validation purposes. Ideally the validation set is not used in the development of the model and saved specifically for validation.[86]

The next step is to build the model using the training data set and several accepted mathematical equations, identifying variables which continue to improve the model. When no previous or limited knowledge exists that can adequately describe a relationship between variables of interest, the researcher generally starts with a body of data and attempts to fit equations to it. The most commonly used algorithms to accomplish this are support vector analysis, linear regression, logistic regression and linear discriminant analysis.[101] When modelling the data using the training set, the inputs consist of the input predictor variables and the outputs are the result after application of the model. Commonly used methods to create this transformation include sum of squared residuals, which is an aggregate measure of total variability, maximum likelihood estimation, a method of estimating the parameters of a probability distribution, or maximizing the constraining hyperplane, also called support vector machines (SVM). SVM, considered a non-probabilistic binary linear classifier, uses the training data set and identifies the variables as belonging to one of two categories, it then builds a model that maximizes the width of the gap between the two and then assigns new data to one or the other category.[101]

After models are identified, the investigator must select the one that best describes or fits the data. Thus, the next step in the process is to take the second set of data, the test set, and

decide which of the developed models works best by some measure of goodness. This is generally accomplished by identifying the model with the smallest mean squared error between predicted and measured data.[86]

The last step is model validation which is performed with the remaining 10% of data partitioned aside for this purpose. Validation is the objective method that establishes the generalizability of the model and tests it against observations from the system it is intended to represent. A model should be validated for both intrinsic bias and extrinsic variance. The most common way to validate the model is to measure the mean squared error from the residuals as the model is applied with the difference between the observed and predicted values providing a measure of accuracy.[86,104]

Development of a stochastic model

An example of stochastic modeling is the development of the NUTRIC risk assessment tool.[105] Heyland and colleagues postulated that not all critically ill patients have the same nutrition risk in that some benefit from nutrition intervention more than others. Their goal was to identify variables that would quantify the risk of a patient developing adverse events. Based on previously published research, they postulated that potential variables were age, the Acute Physiology and Chronic Health Evaluation (APACHE II) score, the Sequential Organ Failure Assessment (SOFA) score, interleukin-6 (IL-6), c-reactive protein, co-morbid illness, weight loss over the last 6 months, BMI <20, decreased oral intake over the previous week and pre-ICU hospital admission. The outcomes selected were ICU length of stay, 28-day mortality and ventilator free days within 28 days. The data set identified was the EMR from a convenience sample of 597 patients admitted to 3 tertiary care medical-surgical ICUs.

The investigators first validated the candidate variables by describing their association with 28-day mortality with chi-squared tests, Wilcoxon rank-sum tests, and Spearman correlation coefficients. Based on results and an additional sensitivity analysis, variables of oral intake, weight loss, BMI and c-reactive protein were excluded from further models. The remaining candidate variables were each fit as categorical predictors in separate single predictor logistic regression models predicting 28-day mortality. The parameters for each logistic regression model estimated the log of the odds ratio for each category of the variable compared to the lowest risk category. These parameters were rounded to whole numbers to provide the points used in the NUTRIC score. Variables were excluded if the inclusion in the NUTRIC score did not improve the scores ability to predict 28-day mortality. Additionally, using a multivariable fractional polynomial approach yielded a similar model with no improvement in performance. The final variables remaining were age, APACHE II score, SOFA score, number of co-morbidities, days from hospitalization to ICU admission and IL-6 level. Each variable remained independently statistically significant in the multivariable logistic model.

Validation of the model was performed by evaluating the quality of the NUTRIC model to predict 28-day mortality on a set of data. Model discrimination was assessed by the C-statistic derived from calculating the area under the receiving operating characteristic curve and the generalized max-rescaled R-squared statistic. Goodness of fit was assessed by comparing the predicted and actual mortality by the Hosmer-Lemeshow goodness of fit test.

Lastly, in a subset of 211 patients who had dietary intake data, the researchers examined if the NUTRIC score modified the association between intake and 28-day mortality. Logistic regression with nutritional intake, NUTRIC score and their product as continuous independent variables was used to generate a plot of association between intake and mortality by NUTRIC

score. They demonstrated that the association between risk score and mortality is attenuated in patients who meet calorie targets. The authors concluded that they believe they have identified a novel scoring tool, using a variety of mathematical techniques, to help identify which patient would benefit most from aggressive nutrition intervention.[105]

Model Application for Current Problem

As low muscularity has been strongly associated with outcomes, it is important to be able to promptly and easily identify those patients. Muscle measurements using techniques such as CT scans exist, however disadvantages of cost, exposure to radiation, lack of standardization of procedures, and limited access to techniques, trained radiologists and equipment will likely make these measurements infrequently available to the average clinician.[54] The nutrition focused physical exam remains a subjective measure and anthropometric measurements of mid-arm circumference and triceps skinfold are impractical for screening all new admissions. The identification of readily available, non-invasive biomarkers or measures would allow rapid screening of the majority of hospital admissions and provide a potential method for subsequent longitudinal follow up.[57] UCE and subsequent estimation of CHI have been strongly associated with muscularity and outcomes, however require a 24-hour urine collection. The postulation that UCE may be estimated from plasma creatinine or other patient variables would avoid the need for a 24-hour urine collection and is uniquely suited to mathematical modeling. The ability to predict CHI from model derived UCE may have clinical utility to become a quick screening tool to identify low muscularity, malnutrition and predict outcomes.

Chapter 3: Methodology

Study Purpose

The purpose of the study was to develop a model to predict UCE from patient variables. The study additionally sought to determine if application of this model, to predict UCE and subsequent estimation of CHI, in a cohort of hospitalized veterans, was correlated with outcomes.

Study Aims

The primary aim of this research was to determine if 24-hour UCE can be reliably estimated from plasma creatinine or other patient variables by the development of a model and if the model can be validated. The secondary aims were to determine if low muscularity based on model derived CHI was correlated to low muscularity based on anthropometric measurements, to describe the degree of low muscle mass observed and determine if model derived UCE and CHI were associated with other assessment measures and outcomes in a cohort of hospitalized veterans. This study was reviewed and approved by the James A. Haley Veterans Hospital Research and Development Committee and the University of South Florida IRB and determined to meet criteria for an exempt study.

Hypothesis

Phase 1: Hθ There is no relationship between patient variables and UCE.

Phase 2: Hθ There is no relationship between UCE, CHI and outcomes.

Objectives

1. To build a model to predict 24-hour UCE from plasma creatinine or other patient variables using a large de-identified data set.
2. To validate the proposed model.

3. In a subset of patients, who had anthropometric measurements obtained, explore how well CHI, estimated from the model derived UCE, is associated with muscularity based on anthropometric measurements
4. Apply the model retrospectively to 120 intensive care unit patients to predict 24-hour UCE for each subject
5. Based on model derived 24-hour UCE, calculate CHI for the whole cohort and describe prevalence of low muscularity in the cohort.
6. To explore if a relationship exists between CHI and the identification of malnutrition using AND/ASPEN criteria
7. To explore if CHI is associated with outcomes of hospital LOS, ICU LOS, hospital mortality, 6-month mortality, 30-day readmission or discharge location.
8. To explore if relationships exist between CHI and commonly collected assessment parameters of albumin, prealbumin, c-reactive protein (CRP), NUTRIC score, APACHE 2 score and SOFA score.

Study design

Phase 1: Model Development Study Design

The first phase was the development of a model to predict 24-hour UCE from plasma creatinine level or other candidate variables of plasma blood urea nitrogen, sodium, potassium, glucose, chloride, carbon dioxide and magnesium and demographic variables of gender, age in years, height in centimeters, weight in kilograms and presence of spinal cord injury or neurologic disease. The model with the best R^2 was examined for validation. Lastly, whether muscularity, based on anthropometric measurements, was correlated to muscularity, based on model derived CHI, was examined.

Phase 1: Study Participants and Sample Size

Eligible participants were veterans who received care at the James A. Haley Veterans Hospital in Tampa Florida (JAHVA) who had 24-hour urine laboratory testing which included creatinine excretion between 10/01/2016 to 09/30/2021. It was estimated that approximately 300-1000 patients would be sufficient for model development.[86]

Phase 1: Data Collection

A data set was obtained from Data Analytics and Acquisition Services (DAAS) which contained the laboratory, demographic and diagnosis variables of patients who received care at JAHVA, and who had 24-hour UCE measured, between 10/01/2016 to 09/30/2021. Mid-arm muscle circumference, triceps skinfold and mid-arm muscle area measurements were extracted from the EMR in a subset of 44 patients who had anthropometric measurements obtained by searching with the note title of "Nutrition Assessment".

Phase 1: Statistical Methods

An initial test data set of 956 serum and urinary creatinine pairs was examined to identify potential mathematical relationships. A scatter plot was generated and visually examined for spline fitting. For plasma creatinine values greater than 5.0 mg/dL the relationship between serum and urinary creatinine appeared random. For pairs with plasma creatinine levels between 1.5 and 5.0mg/dL, a linear relationship appears to emerge but with substantial scatter. For plasma creatinine from 1.5mg/dL and below, a strong linear relationship was observed with a wide band of scatter around a central line. Using the 448 urine and plasma creatinine pairs with plasma creatinine of 1.5 or below, a regression analysis was run.

Next, additional patient input variables, readily available on admission, were added to develop models and improve the overall predictability. Input variables included were age, height,

weight, gender, serum urea nitrogen, glucose, sodium, potassium, chloride, carbon dioxide, magnesium and presence of a spinal cord injury (SCI) as surrogates to account for total body water, extracellular water and lean body mass. A data set of those patient variables for patients who had UCE measured in the previous 5 years was obtained from DAAS. The data set of 1592 patients were reduced to 967 after patients with missing or incomplete data were excluded. The subjects in the data set were 87.4% male and 12.6% female. For this step of model development, a plasma creatinine of $\leq$ 1.2 mg/dL was considered normal as it reflects the hospital's reference interval.

Descriptive statistics were completed, and all variables were examined for skewness and kurtosis. A correlation matrix was run and examined for multicollinearity. Age, weight, plasma creatinine and plasma BUN were observed to be the least correlated with one another. Next, a best fit regression was conducted which took every possible combination of variables and examined the R^2 for each regression equation. Using several mathematical and analytical modeling approaches, including support vector analysis, linear regression and polynomial regression, models were constructed. Using stepwise regression and backward elimination with Alpha-to-remove at the 0.1 level, deleting input variables which were not significant, linear regression models were produced. The amount of variance explained did not increase after input of the four variables of age, weight, plasma creatinine and plasma BUN. To avoid overfitting, further input variables were not included.

Where indicated, Box-Cox transformation, mathematical curve fitting, was performed to test if transforming the outcome variable improves the model and to optimize the mean squared error. Using Box-Cox transformation, the log of the outcome variable, which produced the smallest mean squared error, was identified. For the final identified statistically significant

multivariable models, each remaining input variable was found to be independently statistically significant.

Next, the model with the smallest mean squared error between predicted UCE and measured UCE, Model 3, was identified for validation. Validation was performed with two approaches. First the model was validated using cross validation methodology in which the data was randomly divided into 4 data sets. Then, using an additional set of 50 patients which were not included in the model development, the model was tested for goodness of fit, predictive performance and regression residual diagnostics.[101] Statistical analyses for the development of models were performed using Minitab, I., 2020. MINITAB, http://www.minitab.com/en-US/products/minitab/.

Lastly, in a subset of 44 subjects, with normal plasma creatinine and without the presence of SCI, who had upper arm anthropometry measured, CHI was calculated from the Model 3 predicted UCE. A correlation matrix was run comparing CHI to arm circumference (AC), arm muscle area (AMA) and arm muscle circumference (AMC).

Phase 2: Model Application Study Design

In the second phase, a retrospective review of the electronic medical records (EMR) of ICU patients who received a complete nutrition assessment by the registered dietitian at the JAHVA was conducted. Using patient data extracted from the EMR, Model 3 was applied to estimate UCE and CHI for the cohort. Based on CHI, calculated from UCE, the prevalence of low muscularity was described for the cohort.

Next, extracted data was examined to determine if UCE or CHI were clinically useful in predicting the presence of malnutrition, or were associated with outcomes of hospital LOS, ICU

LOS, hospital mortality, discharge location, 6-month mortality and 30-day readmission in the cohort. Lastly, extracted data was examined to determine if relationships exist between UCE and CHI and patient variables of albumin, prealbumin, c-reactive protein, NUTRIC score, APACHE 2 score, and SOFA score.

Phase 2: Study Participants and Sample Size

Patients who were hospitalized at JAHVA between 1/1/2016 to 9/30/2021, who during the course of their hospitalization required ICU admission, received full nutrition assessments and who had plasma creatinine levels of 1.2 mg/dL or less on admission were considered for inclusion. Patients were excluded if they were hospitalized < 72 hours, their admission plasma creatinine was greater than 1.2 mg/dL, or if they had a medical condition that is associated with muscle wasting such as spinal cord injury, multiple sclerosis, amyotrophic lateral sclerosis or other neurological diseases. It was estimated, for analysis, that a sample size of 120 patients was needed for a medium effect size, and an α of 0.05.[106,107]

Phase 2: Data collection

Eligible patients were identified from the EMR by searching note titles of "Nutrition Assessment", with the location of one of the intensive care units between the dates of 1/1/2016 to 9/30/2021. Patients identified for potential inclusion had their EMR reviewed for eligibility. The retrospective chart review began with those admitted 9/30/21 and proceeded backwards until 120 subjects that met criteria were reached. All data for Phase 2 was obtained and extracted from the medical record. Data after extraction from the EMR was entered into a password protected spreadsheet and de-identified upon entry.

All patients identified for inclusion in Phase 2 had the following extracted from the EMR for the admission:

- age on admission
- gender
- reported race
- height in centimeters
- admission weight in kilograms
- admission BMI
- Primary and secondary medical diagnosis
- date of hospital admission
- date of ICU admission
- date of ICU discharge
- date of hospital discharge
- admission plasma creatinine and blood urea nitrogen
- albumin from date of nutrition assessment
- pre-albumin from date of nutrition assessment
- C-reactive protein from date of nutrition assessment
- presence of malnutrition based on documentation in nutrition assessment
- mortality during admission and up to 6 months after discharge
- Discharge location
- Readmission within 30-days of discharge

The following were calculated for each patient from extracted data:

- ideal body weight (IBW)
- Model 3 derived UCE

- CHI calculated from Model 3 derived UCE
- APACHE 2 score
- SOFA score
- NUTRIC score
- Hospital and ICU LOS

Model 3 was applied to each subject to predict UCE based on admission weight and admission plasma creatinine and BUN. For patients with multiple ICU admissions during the course of their hospital stay, all ICU days were added together to represent total ICU LOS. Primary and secondary medical diagnosis were collected to calculate APACHE 2 and NUTRIC scores. APACHE 2 and SOFA scores were based on the first ICU admission for those who had multiple admissions. Discharge location was grouped as either discharge to home, skilled nursing facility, long term acute care, hospice, transfer to another hospital for care not available at JAHVA or not applicable for those that died during the admission. Degree of sarcopenia was determined based on CHI and defined by the following cut-offs: >75% of expected as normal/absent sarcopenia, <75-61% of expected as mild sarcopenia and <60% of expected as severe sarcopenia. Death dates, even if they occurred outside of a hospitalization, are recorded in the EMR as it serves as part of the veterans record of benefits.

Phase 2: Statistical Methods

Descriptive statistics were calculated to describe characteristics of the cohort. Continuous variables are reported as mean (± standard deviation) or median (25%-75% interquartile range [IQR]) and categorical data are reported as counts and percentage. Between-group comparisons for baseline characteristics and length of stay were analyzed using a χ^2 test for categorical variables and Student *t* test or MANOVA for continuous variables as appropriate. A correlation matrix was run for all predictor and outcome variables to identify possible relationships. Linear regression analysis models, ANOVA and MANOVA as appropriate, were performed to assess the association of UCE and CHI and outcomes and patient characteristics. Odds ratios were calculated using χ^2 and logistic regression to examine relationships between CHI and outcomes. For purposes of statistical analysis, CHI $\leq$60% was considered low and >60% was normal, malnutrition was classified as present or not present, and mid-arm muscle area $\leq 25^{th}$ percentile was classified as low and $> 25^{th}$ percentile as normal. All tests for statistical significance were two tailed and statistical significance was established at the threshold of $p < .05$. Missing data for laboratory measures was managed by insertion of means, as missing data represented only 2-6% of each measure. Statistical analyses were performed using IBM SPSS Statistics for Windows, version 25.0 (Armonk, NY: IBM Corp; 2017).

Chapter 4: Results

This chapter will present the results of the data analysis for Phase 1 and Phase 2 of the research study. First, the results of model development will be presented. This will include the identification of several viable models. Next, the results of model validation will be presented. This is then followed by results of the analysis of model derived output and correlation with

known measures of muscularity. Next, the results of the application of the model to a cohort of patients will be reported. Baseline demographics of the cohort will be presented and prevalence of low muscularity and malnutrition for the cohort will be described. Lastly, the relationship between model derived UCE and CHI and outcomes will be highlighted.

Phase 1

Model Building

An initial model was developed using only plasma and urinary creatinine pairs. Using the 448 urine and plasma creatinine pairs with plasma creatinine of 1.5 or below, it was identified that serum creatinine was a significant predictor of UCE but only accounted for 5% of the variability, $R^2 = .0549$; $p < .001$, rendering it unsuitable to use for the practical prediction of individual UCE values. Residual diagnostics were conducted and found to be normally distributed.

After the additional patient input variables were added, the following linear regression models were identified with potential utility for different patient populations (Table 2, Table 3, Table 4). Gender is defined as 1 = male and 2 = female. SCI is defined as 1 = presence of SCI and 2 = no SCI.

Model 1. For the general population with normal plasma creatinine:

Predicted Urinary Creatinine = 1426.93 – (262.82 x Gender) + (403.15 x SCI) – (20.47 x Age) + (7.39 x Weight) – (66.23 x Plasma Creatinine), $R^2 = .4085$, $p < .001$.

Model 2. For the general population with plasma creatinine >1.2mg/dL:

Predicted Urinary Creatinine^0.5 = 36.02 – (4.76 x Gender) + (7.12 x SCI) – (0.298 x Age) + (0.12 x Weight) – (1.04 x Plasma Creatinine) – (0.02 x Plasma BUN) – (0.17 x Plasma Potassium), $R^2 = .4051$, $p < .001$. In which the predicted urinary creatinine is transformed by the

square root, the result of which is then squared to revert back to the original scale, to obtain the final model predicted urinary creatinine.

Model 3. For patients without SCI and with a normal plasma creatinine:

Predicted Urinary Creatinine^0.833735 = 0 – (1.11 x Age) + (3.00 x Weight) + (231.92 x Plasma Creatinine) – (2.59 x Plasma BUN), $R^2 = .9098$, $p < .001$. In which the predicted urinary creatinine is transformed, and the result of which is then reverted back to original scale, to obtain the final model predicted urinary creatinine. Overall, Model 3 was found to be highly predictive of UCE.

Table 2. Model 1 For Patients with or without SCI and Plasma Creatinine < 1.2 mg/dL

	B	*SE (B)*	*β*	*p*
Constant	1426.93	282.14		< .001
Gender	-262.82	120.07	-0.16	0.03
SCI	403.16	88.33	0.24	< .001
Age	-20.47	1.81	-0.44	< .001
Weight	7.39	0.87	0.30	< .001
Plasma Creatinine	-66.23	5.09	-0.03	<.001

$R^2 = .408$, $p < .001$

Table 3. Model 2 For Patients with or without SCI and Plasma Creatinine >1.2 mg/dL

	B	*SE (B)*	*β*	*p*
Constant	36.02	4.93		< .001
Gender	-4.76	1.94	-0.001	.014
SCI	7.12	1.43	0.003	< .001
Age	-0.30	0.03	-0.006	< .001
Weight	0.12	0.01	0.005	< .001
Plasma Creatinine	-1.05	0.09	-0.008	< .001
Plasma BUN	-0.02	0.01	-0.0009	.16
Plasma Potassium	-0.17	0.52	-0.0002	.74

$R^2 = .405$, $p < .001$

Table 4. Model 3 For Patients without SCI and Plasma Creatinine < 1.2 mg/dL

	B	*SE (B)*	*β*	p
Age	-1.11	0.36	-0.095	.002
Weight	2.99	0.25	0.486	< .001
Plasma Creatinine	231.92	30.71	0.339	< .001
Plasma BUN	-2.59	0.61	-0.196	< .001

R^2= .91, p <.001

Model Validation

Model 3 validation was conducted by two methods. First, cross validation was conducted using the original data set. Four-fold cross validation resulted in R^2 of .4171, .4485, .4488 and .4488, all p < .001. Next, Model 3 was validated using a data set of 50 subjects, which were not included in model development. Mean predicted UCE was 1019.46 mg $\pm$ 318.63 SD, and mean measured UCE was 1033.04 mg $\pm$ 342.08 SD. Predicted and measured UCE were found to be highly correlated, r = .716, p < .001. Linear regression analysis yielded R^2 = .502, F = 50.478, p < .001. Durbin-Watson statistic = 2.071 indicated no autocorrelation in the residuals of the regression.

Relationship between model derived CHI and muscularity

In a subset of 44 subjects, with normal plasma creatinine and without the presence of SCI, who had upper arm anthropometry measured, CHI was calculated from the Model 3 predicted UCE. A correlation matrix was run comparing CHI to arm circumference (AC), arm muscle area (AMA) and arm muscle circumference (AMC). Model derived CHI was found to be strongly correlated to AC, r (42) = .665, p < .001, AMA, r (42) = .551, p < .001, and AMC, r (42) = .559, p < .001. Overall, CHI calculated from Model 3 predicted UCE was found to be highly correlated with accepted measures of muscularity.

Phase 2

Application of Model

A total of 191 medical records were retrospectively screened for inclusion. Forty-seven were excluded due to admission plasma creatinine >1.2mg/dL and 24 were excluded due to medical diagnosis which are associated with muscle wasting. A cohort of 120 patients had Model 3 applied and UCE and CHI estimated, demographic and outcome data collected and were included in the final analysis.

Baseline demographics of study population

Baseline admission characteristics are presented on Table 5. All subjects were male. The mean age for the cohort was 69.08 $\pm$ 8.7 years, mean height 176.4 $\pm$ 6.8 cm, mean weight 83.5 $\pm$ 19.5 kg, mean BMI 26.9 $\pm$ 6.4 and mean IBW 73.8 $\pm$ 7.3 kg. Self-reported racial composition of the cohort was 93 (77.5%) were white, 24 (20%) were black and 3 (2.5%) did not report race. Admission laboratory values of the cohort were mean plasma creatinine 0.94 $\pm$ 0.17 mg/dL and mean plasma BUN 15.6 $\pm$ 5.1 mg/dL.

Table 5. Baseline Demographics

Characteristic	Whole Cohort (n=120)	Subjects with CHI ≤ 60% (n=53)	Subjects with CHI >60% (n=67)
Age, median (IQR), years	69.00 (65.00,73.00)		
Mean (SD)			
Age, years	69.08 (8.75)	69.00 (9.24)	69.13 (8.41)
Height, cm	176.42 (6.77)	177.61 (6.98)	175.48 (6.51)
Weight, kg	83.50 (19.51)	69.93 (12.86)	94.24 (17.06) ***
BMI	26.90 (6.39)	22.11 (3.78)	30.69 (5.42) ***
Plasma BUN, gm/dL	15.64 (5.05)	15.24 (5.13)	15.96 (5.00)
Plasma Creatinine, mg/dL	0.94 (0.17)	0.83 (0.12)	1.03 (0.14) ***
Albumin, gm/dL	2.55 (0.58)	2.42 (0.48)	2.65 (0.63) *
Prealbumin, mg/dL	10.48 (5.67)	9.00 (4.18)	11.69 (6.42) **
CRP, mg/L	9.46 (7.69)	9.25 (7.69)	9.63 (7.76)
APACHE Score	15.93 (4.88)	16.09 (5.29)	15.81 (4.57)
SOFA Score	3.59 (2.64)	3.57 (2.49)	3.61 (2.76)
NUTRIC Score	3.63 (1.39)	3.62 (1.47)	3.64 (1.33)

CHI = creatinine height index; * $p < .05$; ** $p < .01$; *** $p < .001$

The mean severity of illness scores and nutrition screening scores for the cohort were APACHE II 15.93 ± 4.88, SOFA score 3.59 ± 2.64, and NUTRIC score 3.63± 1.39. The mean serum protein levels, on the day of the nutrition assessment, were Albumin 2.55 ± 0.58 g/dL, Prealbumin 10.48 ± 5.66 mg/dL and CRP 9.46 ± 7.70 mg/dL.

There was a statistically significant difference in baseline characteristics based on CHI, $F(13,106) = 18.48$, $p < .001$; Wilks' $\Lambda = 0.306$, partial $\eta^2 = .694$ (Table 5). Subjects with CHI ≤ 60% were observed to have significantly lower body weight, BMI, plasma creatinine, albumin and prealbumin levels compared to those subjects with CHI > 60%.

Prevalence of low muscularity in the cohort

Application of Model 3 to estimate UCE and subsequent calculation of CHI resulted in mean UCE of 1102.81 ± 284.56 mg/24-hours and mean CHI of 0.65 ± 0.17. For the entire

cohort, 33 (27.5%) were considered to have adequate muscle mass and had CHI >75%, 34 (28.3%) had CHI levels between 75-61% and considered to have mild sarcopenia and 53 (44.2%) were found to have CHI levels ≤ 60% and were considered to have severe sarcopenia, Table 6.

Table 6. Prevalence of Sarcopenia Based on Creatinine Height Index

Degree of Sarcopenia	Frequency	Percent of Total
CHI >75% Normal, No sarcopenia	33	27.5
CHI levels between 75-61% Mild Sarcopenia	34	28.3
CHI levels ≤ 60% Severe sarcopenia	53	44.2

Relationship with Malnutrition

Based on the nutrition assessment by the registered dietitian and using the AND/ASPEN criteria for malnutrition, for the entire cohort, 50 (41.7%) were identified with malnutrition and 70 (58.3%) were identified as not having malnutrition, Table 7. Differences in prevalence of malnutrition were noted when subjects with CHI ≤ 60% were compared to those with CHI >60%. For subjects with CHI ≤ 60%, 36 of 53 subjects, 68%, were identified with malnutrition. For subjects with CHI >60%, 14 of 67 subjects, 21%, were identified with malnutrition (Table 7, Figure 1). Subjects with CHI ≤ 60%, were 8.0 times more likely to be identified with malnutrition (OR = 8.0; 95% CI = 3.5, 18.3; $p < .001$), Table 8. UCE and CHI were found to be moderately correlated to malnutrition, $r = .57$, $p < .001$ and $r = .56$, $p < .001$ respectively. Conversely, NUTRIC score, was found to have no relationship with presence of malnutrition, $r = .004$, $p = .96$.

Table 7. Prevalence of Malnutrition Based on AND/ASPEN Clinical Characteristics

	Entire Cohort n = 120	CHI levels > 60% n = 67	CHI levels ≤ 60% n = 53
Number	50	14	36
Percent	41.7	21	68

Figure 1. Malnutrition Based on CHI Level

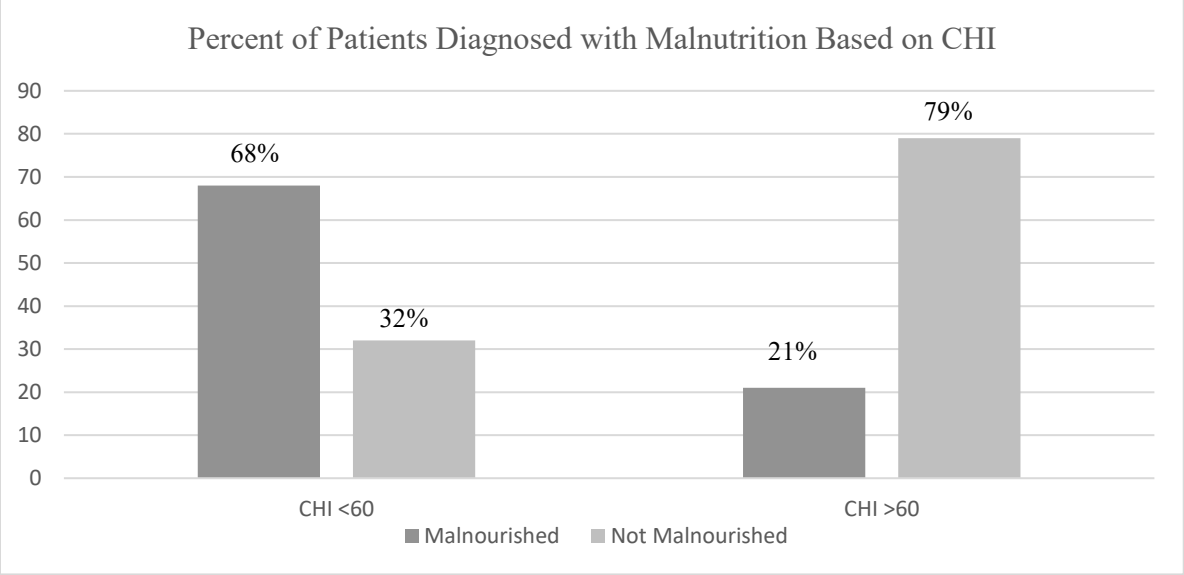


Table 8. Comparison of Malnutrition between Subjects with Low or Normal CHI

Outcome	CHI ≤ 60%[a] n = 53	CHI > 60%[a] n = 67	Odds Ratio (95% CI)	p value
Malnourished	36 (68)	14 (21)	8.02 (3.52-18.28)	p < .001

[a] = number (%)

Outcomes for the cohort

Table 9. Outcomes

Outcome	Frequency	Percent of Total
Hosp Mortality		
Yes	13	10.80
No	107	89.22
Six Month Mortality		
Yes	23	19.20
No	84	70.00
NA (Died during hospital stay)	13	10.80
Malnutrition		
Yes	50	41.70
No	70	58.30
Discharge Destination		
Home	68	56.70
Skilled nursing facility	27	22.50
Long term acute care facility	2	1.70
Hospice	8	6.70
Transfer to another hospital	2	1.70
NA (Died during hospital stay)	13	10.80
Readmit in 30 Days		
Yes	28	23.30
No	78	65.00
NA (Died during/after hospital stay)	14	11.70

Table 9 displays the overall summary of outcomes for the cohort. A total of 13 (10.8%) of subjects expired during the hospitalization, with 107 (89.2%) surviving to be discharged. For those who survived to discharge 68 were discharged home and 39 discharged elsewhere, with 27 discharge to a SNF, 2 discharged to a LTAC facility, 8 discharged to hospice care and two transferred to another hospital for care not available at JAHVA. For those that were discharged from the hospital, 28 were readmitted within 30-days and 23 expired within 6-months of initial hospital discharge.

The mean hospital length of stay for the entire cohort was 34.6 ± 24.6 and median 25.0 (IQR, 19.0-46.5) days. The mean ICU length of stay for the entire cohort was 14.7 ± 19.3 and median 10.0 (IQR, 4.0-17.8) days, Table 10.

Table 10. Length of Stay

	Days, mean	SD	Days, median	IQR
Hospital Length of Stay	34.6	24.6	25.0	19.0-46.5
ICU Length of Stay	14.7	19.3	10.0	4.0-17.8

Relationship between UCE, CHI and outcomes

Linear regression analysis determined that UCE was significantly associated with outcomes of presence of malnutrition, hospital mortality, 6-month mortality, hospital LOS, ICU LOS, discharge location and readmission within 30-days of discharge, $r = .594$, $R^2 = .353$, ($F(7,112) = 8.72$, $p < .001$). Similarly, using linear regression, CHI was significantly associated with outcomes of presence of malnutrition, hospital mortality, 6-month mortality, hospital LOS, ICU LOS, discharge location and readmission within 30-days of discharge, $r = .575$, $R^2 = .296$, ($F(7,112) = 7.92$, $p < .001$).

Table 11. Length of Stay by Creatinine Height Index

Length of Stay Days	Whole Cohort (n = 120)	Subjects with CHI $\leq$ 60% (n=53)	Subjects with CHI >60% (n=67)	p value
Hospital LOS, median (IQR)	25.0 (19.0- 46.5)	28.0 (20.0-55.5)	23.0 (17.0-23.0)	
Hospital LOS, mean (SD)	34.6 (24.6)	38.3 (24.5)	31.7 (24.5)	p = .351
ICU LOS, median (IQR)	10.0 (4.0-17.8)	11.0 (5.0-23.0)	8.0 (4.0-17.0)	
ICU LOS, mean (SD)	14.7 (19.3)	16.7 (19.7)	13.6 (19.3)	p = .351

Table 11 and Figure 2 display the results of the differences in LOS by CHI. Subjects with CHI $\leq$ 60% had mean hospital LOS of 38.3 $\pm$ 24.5 days compared to subjects with CHI > 60% who had hospital LOS of 31.7 $\pm$ 24.5 days. Subjects with CHI $\leq$ 60% had mean ICU LOS of 16.7 $\pm$ 19.7 days compared to subjects with CHI > 60% who had ICU LOS of 13.6 $\pm$ 19.3 days. Although differences in LOS may be clinically different, there was no statistically significant difference in LOS based on CHI, F (2,117) = 1.06, p = .351; Wilks' $\Lambda = 0.982$, partial η^2 = .018, Table 11.

Figure 2. Hospital and ICU Length of Stay in Days

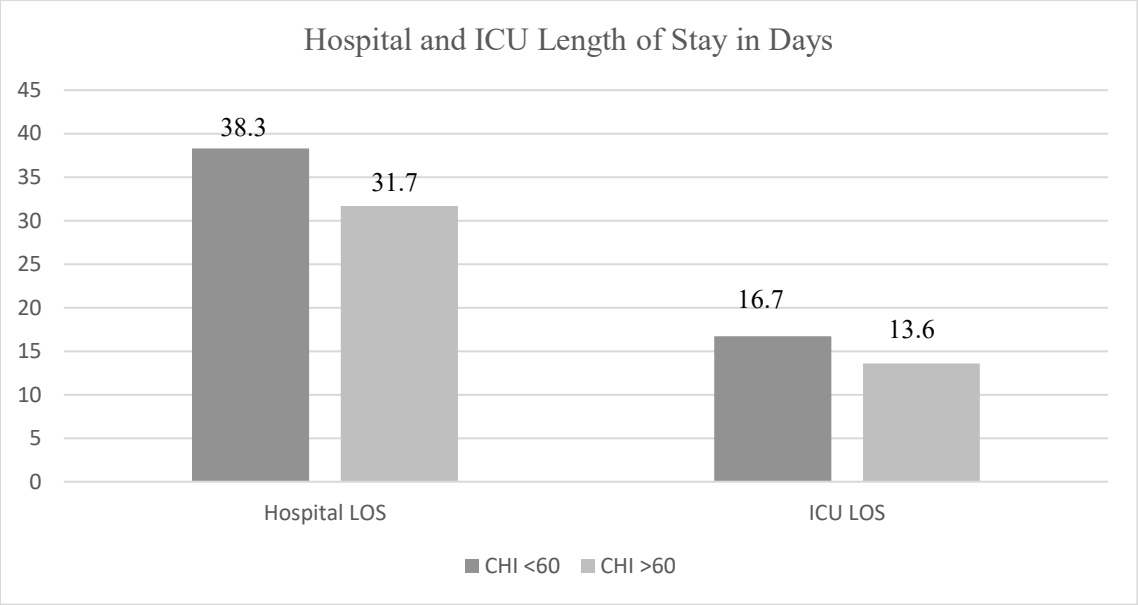


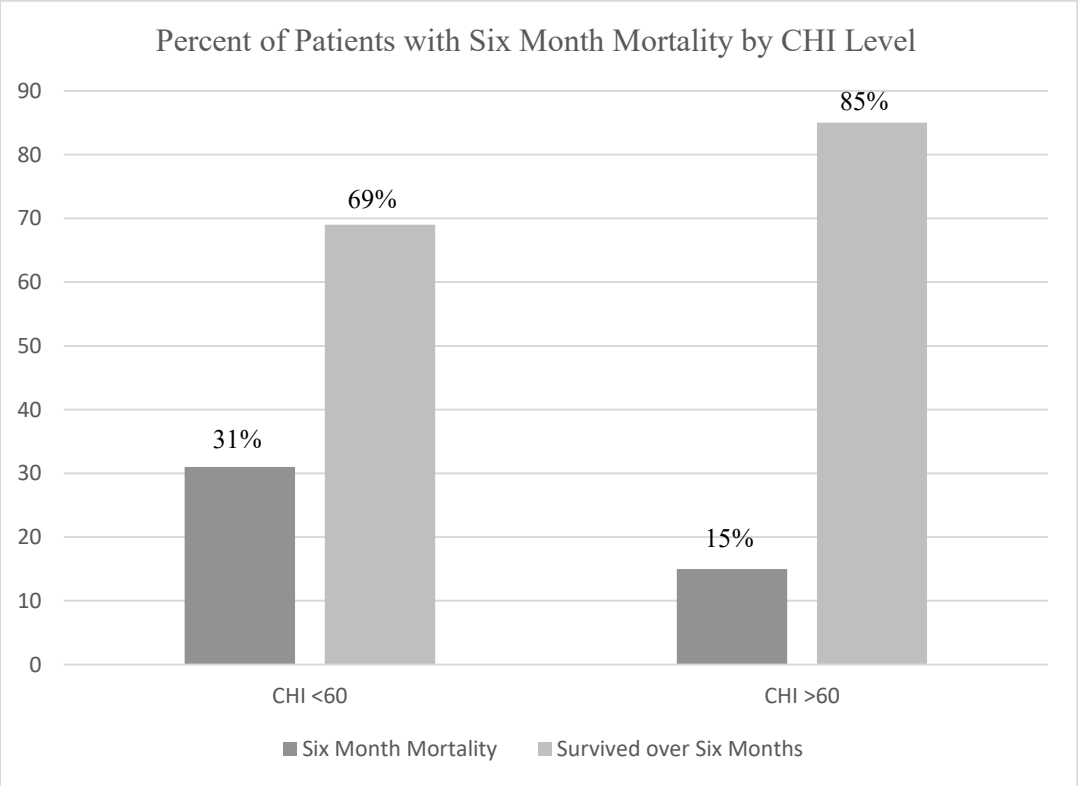
Table 12 displays differences in outcomes by CHI. Patients with CHI ≤60% were 2.2 times more likely to die during the hospitalization, (OR = 2.2; 95% CI 0.68, 7.18; p = .19) and 2.7 times more likely to die within 6-months of discharge, (OR = 2.66; 95% CI, 1.03, 6.9; p < .05). Patients with CHI > 60% were 1.8 times more likely to be discharged home, (OR = 1.8; 95% CI, 0.82, 4.2; p = .15). Of these outcomes, only death within 6-months of hospital discharge met statistical significance, Figure 3. No relationship was observed between CHI and 30-day readmission.

Table 12. Comparison of Outcomes between Subjects with Low or Normal CHI

Outcome	CHI ≤ 60%[a]	CHI > 60%[a]	Odds Ratio (95% CI)	p value
Died during hospitalization	8/53 (15)	5/67 (7)	2.20 (0.68-7.18)	p = .19
Discharged to location other than home	20/45 (44)	19/62 (31)	1.81 (0.82-4.02)	p = .14
Readmitted within 30-days	10/44 (23)	18/62 (29)	0.72 (0.29-1.76)	p = .47
Died within 6-months of discharge	14/45 (31)	9/62 (15)	2.66 (1.0-6.9)	p = .04

[a] = number (%)

Figure 3. Comparison of Six-Month Mortality by CHI



Relationship between UCE, CHI and Serum Proteins, NUTRIC and Severity of Illness Scores

Table 13 displays the correlation between UCE, CHI and serum protein levels and severity of illness scores. UCE and CHI were found to be only weakly associated with albumin, $r = .26$, $p < .005$ and $r = .22$, $p < .05$, respectively. UCE and CHI were also found to be only weakly associated with prealbumin, $r = .25$, $p < .005$ and $r = .24$, $p < .005$, respectively. UCE and CHI were observed to have no significant relationship with serum CRP levels, $r = .02$, $p = .84$ and $r = .01$, $p = .94$, respectively. UCE and CHI were observed to have no significant relationship with NUTRIC score, $r = .14$, $p = .12$ and $r = .03$, p = .75, respectively. UCE and CHI were also observed to have no relationship with severity of illness scores.

Table 13. Correlation between UCE, CHI, Protein Levels, Nutrition Risk and Severity of Illness Scores

	Ur Crt	CHI	Albumin	Prealb	CRP	NUTRIC	APACHE	SOFA
Ur Crt	-							
CHI	0.93***	-						
Albumin	0.26**	0.22*	-					
Prealb	0.25**	0.24**	0.59***	-				
CRP	0.02	0.01	-0.34***	-0.56***	-			
NUTRIC	-0.14	-0.03	-0.30**	-0.25**	0.15	-		
APACHE	-0.18	-0.09	-0.38***	-0.26**	0.03	0.81***	-	
SOFA	0.06	0.11	-0.32***	-0.31**	0.23*	0.56***	0.57***	-

Ur Crt = urinary creatinine excretion; CHI = creatinine height index; Prealb = prealbumin
*p < .05; **p < .01; ***p < .001

Chapter 5: Discussion

The purpose of this research was to investigate if a valid model could be developed to predict UCE, if the model predicted UCE was associated with known measures of muscle measurement and if model predicted UCE had clinical utility in predicting outcomes in a cohort of hospitalized veterans who required an ICU admission. The research included development of models to predict UCE using input variables from 956 patients. Using the model with the best R^2, predicted UCE was compared to anthropometric measurements of muscle in a cohort of 44 subjects who had measurements obtained. Lastly, the model with the best R^2 was applied retrospectively in a cohort of 120 ICU patients and demographic and outcome variables were examined.

Model Development

The first attempt at model development used plasma creatinine as the only input variable to predict UCE. This was trialed as it has been previously postulated in the literature as a potential method to predict UCE.[83] In patients with a normal plasma creatinine it is reasonable to assume that the creatinine level is a reflection of muscle mass and should be highly correlated with measurements of muscle mass. In individuals with normal, stable renal function, variations in muscle mass should be responsible for observed variations in plasma creatinine levels. This study identified, that in this veteran patient population, plasma creatinine was a significant predictor of UCE but only accounted for 5% of the variability, $R^2 = .0549$; $p < .001$, with 95% of the variance accounted for by other untested variables. This rendered the use of plasma creatinine alone unsuitable to use for the prediction of individual UCE values. It seems

reasonable that other variable, such as age, gender, height and weight, which can be associated with the overall amount of muscle mass would play a role in predicting UCE.

Next, additional patient input variables, readily available on admission, were added to develop models and improve the overall accuracy of the predictive equation. Input variables examined for inclusion in final models were age, height, weight, gender, serum urea nitrogen, glucose, sodium, potassium, chloride, carbon dioxide, magnesium and presence of a SCI as these can be considered surrogates to account for total body water, extracellular water and lean body mass.

The first model was developed for the general hospital population with plasma creatinine $\leq$ 1.2 mg/dL, and included patients both with and without SCI. The variables remaining in the model included input variables of gender, age, weight, plasma creatinine and presence or absence of SCI. A significant model was identified, $R^2 = .4085$, $p < .001$, for potential use in phase 2 of the study. The second model was developed for patients with plasma creatinine > 1.2 mg/dL. Input variables remaining in model 2 included gender, presence or absence of SCI, age, weight, plasma creatinine, BUN, and potassium. The identified model was also found to be significant, $R^2 = .4051$, $p < .001$. Lastly, to improve predictability further, a model was developed that excluded patients with the presence of a SCI and included only those with normal plasma creatinine, creatinine $\leq$ 1.2 mg/dL. The input variables remaining in model 3 included age, weight, and plasma creatinine and BUN. This final non-linear model, model 3, was found to be statistically significant, $R^2 = .9098$, $p < .001$, and was selected for further investigation.

The final step in model development was validation. Model 3 was able to be validated using two methods. First, 4-fold cross validation using the original data set was conducted resulting in R^2 of .4171, .4485, .4488 and .4488, all $p < .001$. Next, validation was conducted

using a separate data set of 50 subjects that were not included in the model development. Mean predicted UCE was 1019.46 mg ± 318.63 SD, and mean measured UCE was 1033.04 mg ± 342.08 SD. Predicted and measured UCE were found to be highly correlated, $r = .716$, $p < .001$. Linear regression analysis yielded $R^2 = .502$, $F = 50.478$, $p < .001$. This final model, Model 3, was found to be highly correlated, moderately predictive and statistically significant.

The final model with the best R^2, Model 3, selected for further investigation contained more input variables than had been proposed in the literature for the development of a model.[83] Creatinine is non-protein bound, diffusible across cell membranes and flows freely from one compartment to another with the same permeability as water. The input variables remaining in the model identified in this study are not surprising as they are surrogates for total body water, extracellular water and lean body mass.[112-114] Importantly, these input variables are all readily available on admission. Furthermore, this model was able to estimate UCE and CHI with the benefit of not requiring a 24-hour urine sample, which has been a limitation with using UCE or CHI to assess muscularity.[80–82] Use of a model, such as this, to estimate UCE and CHI rather than 24-hour urine collections, in the hospital setting, can be done within minutes rather than waiting for the urine collection and analysis. It also avoids the inconvenience and pitfalls associated with the urine collection. This model may represent a potential surrogate "biomarker" to screen for low muscularity shortly after admission and allow for early identification of those patients most in need of nutrition intervention.[57]

Relationship between model derived CHI and muscularity

Although this model was found to be correlated and predictive of UCE, it was important to determine if this predicted UCE was related to known measures of muscle mass. To this aim,

this study examined if CHI, estimated from Model 3, was correlated with AC, AMA and AMC in a subset of subjects, who had normal renal function, were without the presence of SCI, and who had upper arm anthropometrics obtained during their nutrition assessment. Model derived CHI was found to be strongly correlated to AC, r (42) = .665, p < .001, AMA, r (42) = .551, p < .001, and AMC, r (42) = .559, p < .001. Overall, CHI calculated from Model 3 predicted UCE was found to be highly correlated with accepted measures of muscularity.

Although there are a variety of techniques to measure and estimate muscle mass, such as CT, DEXA or BIA, these advanced tools were unavailable to the dietitians at JAHVA. Some dietitians at JAHVA, who have the skill and experience, obtain upper arm anthropometry when doing a full nutrition assessment. Thus, for this study, anthropometric measurements were the only available objective measures of muscularity to compare CHI against. The findings of this study, which identified good correlation between model derived CHI and anthropometric measures, confirms what has been found in another study that examined the relationship between UCE and muscularity. Heymsfield et al[55] demonstrated that muscle mass, as estimated from upper arm anthropometry, was strongly correlated with UCE, r = .94, p < .001. Lambell et al[115] recently demonstrated that the bedside technique of upper arm anthropometry was strongly correlated with CT measured muscle area, r = .67, p < .001. UCE has also been found to be strongly correlated with LBM as measure by K^{40} counting and by densitometry and deuterium dilution.[59-61] Thus, the model identified in this study was found to be highly correlated and moderately predictive of UCE and was also strongly correlated with a known objective measure of muscularity.

Model Application

The second phase of this study sought to examine if UCE and CHI, derived from Model 3 had potential clinical usefulness. Phase 2 of the research was conducted by retrospectively applying the model to a cohort of 120 veterans who had been hospitalized and required an ICU admission. Phase 2 examined if UCE was associated with or predicted outcomes of malnutrition, mortality, discharge location or readmission.

Baseline Demographics

Baseline demographics of the cohort are reported on Table 5. As this study was conducted at a Veterans Affairs hospital, all subjects were male, and the median age was 69.08 $\pm$ 8.7 years. The cohort had a mean height of 176.4 $\pm$ 6.8 cm, mean weight of 83.5 $\pm$ 19.5 kg, and mean BMI of 26.9 $\pm$ 6.4, which could imply that the group was well nourished. However, as weight or BMI may be poorly correlated with muscle mass, reliance on these measures may result in the clinician failing to identify low muscle mass and ultimately prevent the clinician from directing their interventions accordingly and should be viewed with caution.[19,29-32]

The mean severity of illness scores for the cohort were APACHE II 15.93 $\pm$ 4.88, which is associated with a 12-24% risk of mortality, and SOFA score 3.59 $\pm$ 2.64, which is associated with a <10% risk of mortality.[116-117] As such, the cohort for this study is considered to have a lower risk for mortality.

The mean serum protein levels, on the day of the nutrition assessment, were albumin 2.55 $\pm$ 0.58 g/dL, prealbumin 10.48 $\pm$ 5.66 mg/dL and CRP 9.46 $\pm$ 7.70 mg/dL. This reduction from normal levels in albumin and prealbumin occurs in the presence of an inflammatory response,

which is reflected by the elevated CRP level and is commonly observed in the critically ill, regardless of nutrition status.[118-119]

When the cohort was examined by whether or not subjects had CHI $\leq 60\%$, considered to be sarcopenic, compared to >60%, some differences between the groups were noted. MANOVA analysis identified a statistically significant difference in baseline characteristics based on CHI, F (13,106) = 18.48, p < .001; Wilks' $\Lambda = 0.306$, partial η^2 = .694 (Table 5). Subjects with CHI $\leq$ 60% were observed to have significantly lower body weight, BMI, plasma creatinine, albumin and prealbumin levels compared to those subjects with CHI > 60%. A higher body weight and higher BMI may not always indicate greater lean body mass, as in those with sarcopenic obesity.[19,29-32] However in this study, those with sarcopenia as identified with model derived CHI, did have significantly lower weight and BMI.

As plasma creatinine varies directly and proportionally with muscle mass, in those with normal renal function, and plasma creatinine levels have been strongly correlated with lean body mass, it was expected that those with low CHI would have lower plasma creatinine.[54-57] This was confirmed by the findings observed in this cohort as those subjects with low CHI had significantly lower plasma creatinine levels (Table 5).

The differences in albumin and prealbumin levels, between those with CHI >60% compared to those with CHI $\leq$ 60%, observed in this study was unexpected as the CRP levels were not different between the groups. In acute illness, some protein levels change in response to cytokines. Those proteins that increase in concentration are referred to as positive acute phase proteins and include CRP and ceruloplasmin. Other proteins fall in the face of an inflammatory response and these are referred to as negative acute phase proteins and include albumin, prealbumin and transferrin.[118] As serum albumin and prealbumin are presently considered to be

associated with inflammation and not markers of nutrition status, one would have expected that lower levels would be observed in the presence of higher CRP levels and thus be similarly low in both groups.[118-120] However, this was not observed. It should be noted that both groups demonstrated below normal albumin and prealbumin levels and elevated CRP levels as expected in the critically ill. Additionally, the severity of illness scores, APACHE II and SOFA, were not different between the two groups. Previous studies have demonstrated that albumin and prealbumin were correlated with measures of inflammation and did not change in response to nutrition intervention.[118,121-122]

It has recently been suggested that low circulating levels of albumin suggests a long-term insufficient nitrogen intake and that treatment should be focused on resolving inflammation and provision of nitrogen.[123] Other potential explanations for the lower levels of albumin and prealbumin observed in those with sarcopenia in this study may be extrapolated from what is known about skeletal muscle. Skeletal muscle is an important metabolically active organ, and plays key roles in protein synthesis.[19,20] In the adequately fed state, amino acids from food supply the needed precursors for new protein synthesis. When nutrient intake is insufficient, muscle protein becomes the principal reservoir of the needed amino acids.[21] Normal protein turnover and synthesis can continue provided adequate muscle mass is available for cannibalism.[21] In the setting of acute illness, requirements for amino acids from skeletal muscle increases due to increased synthesis of acute phase proteins, synthesis of protein components of the immune system and synthesis of proteins necessary for wound healing.[12,20–22] It could be hypothesized that the lower albumin and prealbumin levels seen in the group with sarcopenia reflects the lack of available amino acid substrates from skeletal muscle necessary for protein

synthesis. As this study was not designed to address this interesting finding, it will require additional investigation.

Prevalence of low muscularity in the cohort

Application of Model 3 to estimate UCE and subsequent calculation of CHI resulted in mean UCE of 1102.81 $\pm$ 284.56 mg/24-hours and mean CHI of 0.65 $\pm$ 0.17. For the entire cohort, 33 (27.5%) were considered to have adequate muscle mass and had CHI >75%, 34 (28.3%) had CHI levels between 75-61% were considered to have mild sarcopenia and 53 (44.2%) were found to have CHI levels $\leq$ 60% and were considered to have severe sarcopenia.

Others have reported varying rates of prevalence of sarcopenia in the critically ill, mostly through the use of CT scans. Joyce et al[36] reported a prevalence of sarcopenia of 68% in their cohort of ICU patients. Looijard et al[17] observed a 60% prevalence of low skeletal muscle area using CT scans in their cohort of ICU patients and Weijs et al[28] reported a 63% prevalence. Using DEXA, Abramowitz et al[41] reported that 14% of their study cohort had sarcopenia. Differences in the reported prevalence of sarcopenia in these ICU patients and our findings may reflect the different method of measurement, CT versus DEXA, and for this study, an indirect method of estimating muscularity. However, the findings of this study, a prevalence of 44%, appear reasonable and within the range of what others have reported.

Relationship with Malnutrition

Based on the nutrition assessment by the registered dietitian and using the AND/ASPEN criteria for malnutrition, for the entire cohort, 50 (41.7%) were identified with malnutrition and 70 (58.3%) were identified as not having malnutrition, Table 7. Varying prevalence of

malnutrition in the critically ill have been reported. A recent review of 20 studies found the prevalence of malnutrition to be from 38% to 78%.[124] These widely varying rates are generally considered to be due to different tools and different criteria utilized to diagnosis malnutrition and include the use of Subjective Global Assessment, the Mini Nutritional Assessment or the Malnutrition Universal Screening Tool.[124] The AND/ASPEN publication of standardized malnutrition diagnostic criteria has allowed more uniform analysis and comparisons between groups.[31] A recent study by Hiura et al[125] utilized the standardized malnutrition diagnostic criteria and identified a 23.9% and 21.1% prevalence of severe malnutrition in the medical ICU and surgical ICU respectively. Unfortunately, Hiura et al[125] did not report the prevalence of moderate malnutrition in their groups so the overall prevalence of malnutrition is unclear. The findings of this current study of a 41.7% prevalence of malnutrition in the cohort is in line with the findings of others.[124-125]

Differences in prevalence of malnutrition were noted when subjects with CHI $\leq$ 60% were compared to those with CHI >60%. For subjects with CHI $\leq$ 60%, 36 of 53 subjects, 68%, were identified with malnutrition, Figure 1. For subjects with CHI >60%, 14 of 67 subjects, 21%, were identified with malnutrition. Subjects with CHI $\leq$ 60%, were 8.0 times more likely to be identified with malnutrition (OR = 8.0; 95% CI = 3.5, 18.3; $p < .001$), Table 8. These findings are consistent with the AND/ASPEN standardized malnutrition diagnostic criteria as two of the diagnostic criteria, loss of muscle mass and diminished functional status as measured by hand grip strength, involve measurements of muscle mass and identification of sarcopenia.[31] Importantly, applying Model 3 retrospectively to a cohort of patients identified, on admission, those that would be diagnosed with malnutrition.

Conversely, the NUTRIC score, was found to have no relationship with presence of malnutrition, $r = .004$, $p = .96$. The NUTRIC score, although commonly used as a nutrition screening tool for ICU patients, was designed to identify ICU patients who would benefit from prompt nutrition intervention and not screen for malnutrition or nutrition risk.[105] The NUTRIC score does not actually include any classic nutrition indicators, but is scored based on age, APACHE 2 score, SOFA score, number of co-morbidities, numbers of days from hospital admission to ICU admission and Interleukin 6 level, a pro-inflammatory cytokine.[105] In this study, reliance on the NUTRIC score, alone, to screen patients would have failed to identify those with malnutrition and possibly delayed nutrition intervention.

Outcomes for the cohort

A total of 13 (10.8%) subjects expired during the hospitalization. This confirms predicted estimates from the mean APACHE 2 and SOFA scores with anticipated mortality rates of 12-24% and <10% respectively. Overall, the mean hospital LOS was 34.6 $\pm$ 24.6 days and the mean ICU LOS for was 14.7 $\pm$ 19.3 days (Table 10). Of the 107 patients who were discharged, 28 were readmitted within 30-days and 23 expired within 6-months of discharge.

Relationship between UCE, CHI and Outcomes

This study identified, with linear regression analysis, that UCE and CHI, were both significantly associated with the outcomes of presence of malnutrition, hospital mortality, 6-month mortality, hospital LOS, ICU LOS, discharge location and readmission within 30-days of discharge, $r = .594$, $R^2 = .353$, $p < .001$ and $r = .575$, $R^2 = .296$, $p < .001$, respectively.

Several important differences in outcomes were also observed when subjects with CHI ≤ 60% were compared with those CHI > 60% (Table 11, Table 12, Figure 2, Figure 3). Patients with low CHI were 2.2 times more likely to die during the hospitalization, 1.8 times more likely to be discharged to a location other than their home, and 2.66 times more likely to expire within 6 months of discharge. However, difference in six-month mortality was the only outcome that met statistical significance. Hospital and ICU LOS was longer for those patients with CHI ≤ 60% and could represent a clinical difference however did not meet statistical significance.

The findings of this study of worse outcomes, or a trend towards worse outcomes, in those with low muscularity, coincides with the findings of others which have used various methods to estimate muscle mass. Previous research using CT scans to identify low muscularity have identified an increase in hospital, 30-day, 60-day, 6-month or 1-year mortality in those with sarcopenia.[17,28,37,38,40] Sarcopenia identified with BIA has also been found to be associated with 28-day mortality.[33] Other studies have identified significant relationships between sarcopenia and increased hospital LOS using CT or US measurements.[35,36] Lastly, Hessels et al[66] identified a significant relationship between UCE and hospital mortality. This study, by use of a model, was able to obtain similar findings without the use of invasive testing or a 24-hour urine collection.

Strengths of the Study

Strengths of this study include the availability of a large de-identified data set from which models could be constructed and validated. Additionally, as this data set came from a specific patient population, identified models were highly predictive. Also, the model identified was

simple enough that it can be easily applied by the average clinician and utilized variables that are available for patients shortly after admission.

Several strengths for the design of Phase 2 have been identified. The data collected was readily available within the EMR. Limiting application of the model in Phase 2 to only ICU patients minimized heterogeneity of the study population. Limiting the study population to those that had received a full nutrition assessment by the RDN allowed comparison of model derived UCE and CHI to the presence or absence of malnutrition. Extensive documentation and laboratory testing are done for all patients who receive full nutrition assessments minimizing the likelihood of missing data. Lastly, collecting data during the selected time period ensured that the AND/ASPEN malnutrition criteria were used to identify malnutrition and avoided confounders of differing definitions or criteria of malnutrition.

Limitations of the Study

For Phase 1 several weaknesses are acknowledged. Models are strongly linked to the population which they were derived from, thus the model identified in this study, developed from a group of veterans, may be less applicable in other populations. As this study was conducted at a Veterans' hospital, although data from females was not excluded, female data would be underrepresented in the deidentified data set from which the models were built. For the development of the models, the study assumes that there have been accurate 24-hour urine collections. Cases in which the collection lasts for more or less than 24-hours or in which some urine is lost may result in inaccurate estimates of 24-hour creatinine excretion. Lastly, there is a risk that some patients had elevated UCE not due to increased muscle mass but rather due to consumption of a very high protein diet or certain amino acid supplements.

For Phase 2 of the study additional limitations are acknowledged. Limitations associated with retrospective studies and small sample size may have precluded definitive results. Retrospective studies have the limitation of only identifying associations and not causation. These results may not be generalizable to females, non-veterans and patients of different age groups. The results will also not be applicable to patients with admission plasma creatinine levels of >1.2 mg/dL. It is unclear if these results would hold up if applied to those patients who had not received a full nutrition assessment or who were not ICU patients. Unfortunately, the sample size was too small to detect differences in LOS. These results may also not be applicable to patients with different medical acuity levels, represented by APACHE 2 and SOFA scores, as JAHVA has a patient acuity similar to a community hospital.

Chapter 6: Conclusions, Implications for Practice and Recommendations for Future Research

Conclusions

The primary aim of this research was to determine if 24-hour UCE can be reliably estimated from plasma creatinine or other patient variables by the development of a model and if the model can be validated. Additional aims of the study were to determine if low muscularity based on model derived CHI is correlated to low muscularity based on anthropometric measurements. The last aim was to apply the model to a cohort of hospitalized critically ill veterans, to describe the degree of low muscle mass observed and determine if model derived UCE and CHI are associated with other assessment measures and outcomes.

The study successfully identified a final model to estimate UCE utilizing the input variables of plasma creatinine, plasma BUN, age and weight. Each remaining input variable was found to be independently statistically significant. The final model was found to be highly correlated, moderately predictive of UCE and statistically significant. This model was then appropriately validated using two methods, four-fold cross validation and using a separate data set of subjects not used to construct the model. Next, the study sought to determine if model derived CHI was correlated to known measures of muscularity. Using a subset of patients in whom anthropometric measurements were obtained, the study identified that model derived CHI was highly correlated with arm muscle area, arm muscle circumference and arm circumference.

In Phase 2 of the study, the model was retrospectively applied to a group of critically ill veterans. The model identified that 44.2% of the subjects were found to have CHI levels $\leq$ 60% and were considered to have severe sarcopenia. Subjects with model estimated CHI $\leq$ 60% were found to have significantly lower body weight, BMI, plasma creatinine, albumin and prealbumin levels. Subjects with CHI $\leq$ 60% were found to be 8.0 times more likely to be diagnosed with malnutrition and 2.6 times more likely to be readmitted in 6 months. Subjects with low CHI trended towards longer hospital and ICU LOS, however it did not meet statistical significance. Lastly, a commonly used ICU nutrition screening tool, NUTRIC, was found to have no relationship with the presence of malnutrition.

Implications for practice

The development of a model which predicts UCE and correlates with muscle mass offers a novel method for the RDN to readily identify patients with sarcopenia on hospital admission. This method could allow the RDN to quickly screen new admissions for potential sarcopenia without the use of CT or DEXA scans and without the inconvenience of a 24-hour urine

collection by using readily available patient variables. A model such as this is also ideally suited to be automatically computer generated.

Recent recommendations have been made that muscle mass should be at the core of nutrition screening and management strategies, and that tools and techniques be developed to assess muscle mass.[19] As nutrition screening is moving away from evaluation of weight and towards identifying sarcopenia, this study potentially fills a gap by providing a clinical tool to accomplish this. This would thus allow the RDN to begin prompt nutrition intervention to those patients most in need.

Recommendations for future research

This study was limited by applying the identified model in a retrospective fashion to a group of critically ill veterans. Several future research studies are warranted to determine the applicability of the model to other populations. First, research should be designed to apply the model in a prospective fashion to the same patient population, using a larger sample size, to determine if the findings are consistent and if additional relationships with outcomes can be identified. Next, the model could be applied prospectively to sample populations of hospitalized veterans admitted to the general medical and surgical wards. This would allow analysis of the model's ability to function as an admission nutrition screening tool. If these studies demonstrate positive findings, the model could then be applied to non-veteran patient populations.

Lastly, the findings of differences in albumin and pre-albumin levels between those subjects with or without sarcopenia was unexpected. These findings should be explored with an appropriately powered study examining relationships between these serum protein levels and direct and indirect measures of muscularity.

References

1. McClave SA, Taylor BE, Martindale RG, et al. Guidelines for the Provision and Assessment of Nutrition Support Therapy in the Adult Critically Ill Patient: Society of Critical Care Medicine (SCCM) and American Society for Parenteral and Enteral Nutrition (A.S.P.E.N.). *J Parenter Enter Nutr*. 2016;40(2):159-211. doi:10.1177/0148607115621863
2. Singer P, Blaser AR, Berger MM, et al. ESPEN guideline on clinical nutrition in the intensive care unit. *Clin Nutr*. 2019;38(1):48-79. doi:10.1016/j.clnu.2018.08.037
3. Nicolo M, Heyland DK, Chittams JESSE, Sammarco T, Compher C. Clinical Outcomes Related to Protein Delivery in a Critically Ill Population: A Multicenter, Multinational Observation Study. *J Parenter Enter Nutr*. 2016;40(1):45-51. doi:10.1177/0148607115583675
4. Weijs PJM, Stapel SN, De Groot SDW, et al. Optimal protein and energy nutrition decreases mortality in mechanically ventilated, critically Ill patients: A prospective observational cohort study. *J Parenter Enter Nutr*. 2012;36(1):60-68. doi:10.1177/0148607111415109
5. Weijs PJM, Looijaard WGPM, Beishuizen A, Girbes ARJ, Oudemans-van Straaten HM. Early high protein intake is associated with low mortality and energy overfeeding with high mortality in non-septic mechanically ventilated critically ill patients. *Crit Care*. 2014;18(1):1-10. doi:10.1186/s13054-014-0701-z
6. Allingstrup MJ, Esmailzadeh N, Wilkens Knudsen A, et al. Provision of protein and energy in relation to measured requirements in intensive care patients. *Clin Nutr*. 2012;31(4):462-468. doi:10.1016/j.clnu.2011.12.006
7. Ferrie S, Allman-Farinelli M, Daley M, Smith K. Protein Requirements in the Critically

Ill: A Randomized Controlled Trial Using Parenteral Nutrition. *J Parenter Enter Nutr*. 2016;40(6):795-805. doi:10.1177/0148607115618449

8. de Koning MSLY, Koekkoek WAC, Kars JCN, van Zanten ARH. Association of PROtein and CAloric Intake and Clinical Outcomes in Adult SEPTic and Non-Septic ICU Patients on Prolonged Mechanical Ventilation: The PROCASEPT Retrospective Study. *J Parenter Enter Nutr*. 2020;44(3):434-443. doi:10.1002/jpen.1663
9. Casaer MP, Mesotten D, Hermans G, et al. Early versus late parenteral nutrition in critically ill adults. *N Engl J Med*. 2011;365(6):506-517. doi:10.1177/175114371101200422
10. Casaer MP, Wilmer A, Hermans G, Wouters PJ, Mesotten D, Van Den Berghe G. Role of disease and macronutrient dose in the randomized controlled epanic trial a post hoc analysis. *Am J Respir Crit Care Med*. 2013;187(3):247-255. doi:10.1164/rccm.201206-0999OC
11. Singer P, Anbar R, Cohen J, et al. The tight calorie control study (TICACOS): A prospective, randomized, controlled pilot study of nutritional support in critically ill patients. *Intensive Care Med*. 2011;37(4):601-609. doi:10.1007/s00134-011-2146-z
12. Hoffer LJ, Bistrian BR. Appropriate protein provision in critical illness: A systematic and narrative review. *Am J Clin Nutr*. 2012;96(3):591-600. doi:10.3945/ajcn.111.032078
13. Weijs PJM, Dickerson RN, Heyland DK, Moore FA, Rugeles SJ, McClave SA. Experimental and Outcome-Based Approaches to Protein Requirements in the Intensive Care Unit. *Nutr Clin Pract*. 2017;32(1_suppl):77S-85S. doi:10.1177/0884533617692774
14. Compher C, Chittams J, Sammarco T, Nicolo M, Heyland DK. Greater protein and energy intake may be associated with improved mortality in higher risk critically ill patients: A

multicenter, multinational observational study. *Crit Care Med*. 2017;45(2):156-163. doi:10.1097/CCM.0000000000002083

15. Mogensen KM, Robinson MK, Casey JD, et al. Nutritional status and mortality in the critically ill. *Crit Care Med*. 2015;43(12):2605-2615. doi:10.1097/CCM.0000000000001306

16. Weijs PJM, Mogensen KM, Rawn JD, Christopher KB. Protein Intake, Nutritional Status and Outcomes in ICU Survivors: A Single Center Cohort Study. *J Clin Med*. 2019;8(1):43. doi:10.3390/jcm8010043

17. Looijaard WGPM, Dekker IM, Beishuizen A, Girbes ARJ, Oudemans-van Straaten HM, Weijs PJM. Early high protein intake and mortality in critically ill ICU patients with low skeletal muscle area and -density. *Clin Nutr*. 2020;39(7):2192-2201. doi:10.1016/j.clnu.2019.09.007

18. Alberda C, Gramlich L, Jones N, et al. The relationship between nutritional intake and clinical outcomes in critically ill patients: Results of an international multicenter observational study. *Intensive Care Med*. 2009;35(10):1728-1737. doi:10.1007/s00134-009-1567-4

19. Deutz NEP, Ashurst I, Ballesteros MD, et al. The Underappreciated Role of Low Muscle Mass in the Management of Malnutrition. *J Am Med Dir Assoc*. 2019;20(1):22-27. doi:10.1016/j.jamda.2018.11.021

20. Thongprayoon C, Cheungpasitporn W, Kashani K. Serum creatinine level, a surrogate of muscle mass, predicts mortality in critically ill patients. *J Thorac Dis*. 2016;8(5):E305-E311. doi:10.21037/jtd.2016.03.62

21. Wolfe RR. The underappreciated role of muscle in health and disease. *Am J Clin Nutr*.

2006;84(3):475-482. doi:10.1093/ajcn/84.3.475

22. Hoffer LJ. Human Protein and Amino Acid Requirements. *J Parenter Enter Nutr*. 2014;40(4):460-474. doi:10.1177/0148607115624084

23. Biolo G, Toigo G, Ciocchi B, et al. Metabolic response to injury and sepsis: Changes in protein metabolism. *Nutrition*. 1997;13(9 SUPPL.):52. doi:10.1016/s0899-9007(97)83044-4

24. Weijs PJM, Cynober L, DeLegge M, Kreymann G, Wernerman J, Wolfe RR. Proteins and amino acids are fundamental to optimal nutrition support in critically ill patients. *Crit Care*. 2014;18(6):1-13. doi:10.1186/s13054-014-0591-0

25. Puthucheary ZA, Rawal J, McPhail M, et al. Acute skeletal muscle wasting in critical illness. *JAMA - J Am Med Assoc*. 2013;310(15):1591-1600. doi:10.1001/jama.2013.278481

26. Mogensen KM, Horkan CM, Purtle SW, et al. Malnutrition, Critical Illness Survivors, and Postdischarge Outcomes: A Cohort Study. *J Parenter Enter Nutr*. 2018;42(3):557-565. doi:10.1177/0148607117709766

27. Weijs PJM. Fundamental determinants of protein requirements in the ICU. *Curr Opin Clin Nutr Metab Care*. 2014;17(2):183-189. doi:10.1097/MCO.0000000000000029

28. Weijs PJM, Looijaard WGPM, Dekker IM, et al. Low skeletal muscle area is a risk factor for mortality in mechanically ventilated critically ill patients. *Crit Care*. 2014;18(1):1-7. doi:10.1186/cc13189

29. Prado CM, Purcell SA, Alish C, et al. Implications of low muscle mass across the continuum of care: a narrative review. *Ann Med*. 2018;50(8):675-693. doi:10.1080/07853890.2018.1511918.Implications

30. Skipper A, Coltman A, Tomesko J, et al. Adult Malnutrition (Undernutrition) Screening: An Evidence Analysis Center Systematic Review. *J Acad Nutr Diet*. 2020;120(4):669-708. doi:10.1016/j.jand.2019.09.010

31. White J V., Guenter P, Jensen G, Malone A, Schofield M. Consensus statement: Academy of nutrition and dietetics and American society for parenteral and enteral nutrition: Characteristics recommended for the identification and documentation of adult malnutrition (undernutrition). *J Parenter Enter Nutr*. 2012;36(3):275-283. doi:10.1177/0148607112440285

32. Jensen GL, Cederholm T, Correia MITD, et al. GLIM Criteria for the Diagnosis of Malnutrition: A Consensus Report From the Global Clinical Nutrition Community. *J Parenter Enter Nutr*. 2019;43(1):32-40. doi:10.1002/jpen.1440

33. Thibault R, Makhlouf AM, Mulliez A, et al. Fat-free mass at admission predicts 28-day mortality in intensive care unit patients: the international prospective observational study Phase Angle Project. *Intensive Care Med*. 2016;42(9):1445-1453. doi:10.1007/s00134-016-4468-3

34. Moisey LL, Mourtzakis M, Cotton BA, et al. Skeletal muscle predicts ventilator-free days, ICU-free days, and mortality in elderly ICU patients. *Crit Care*. 2013;17(5). doi:10.1186/cc12901

35. Gruther W, Benesch T, Zorn C, et al. Muscle wasting in intensive care patients: Ultrasound observation of the M. quadriceps femoris muscle layer. *J Rehabil Med*. 2008;40(3):185-189. doi:10.2340/16501977-0139

36. Joyce PR, O'Dempsey R, Kirby G, Anstey C. A retrospective observational study of sarcopenia and outcomes in critically ill patients. *Anaesth Intensive Care*. 2020;48(3):229-

235. doi:10.1177/0310057X20922234

37. Loosen SH, Schulze-Hagen M, Püngel T, et al. Skeletal Muscle Composition Predicts Outcome in Critically Ill Patients. *Crit Care Explor*. 2020;2(8):e0171. doi:10.1097/cce.0000000000000171

38. Jaitovich A, Dumas CL, Itty R, et al. ICU admission body composition: Skeletal muscle, bone, and fat effects on mortality and disability at hospital discharge - A prospective, cohort study. *Crit Care*. 2020;24(1):1-11. doi:10.1186/s13054-020-03276-9

39. Imamura H, Haraguchi M, Minami S, et al. The Impact of Low Muscle Mass in Patients Undergoing Emergency Surgery for Colonic Perforation – A Single-center Experience. *In Vivo (Brooklyn)*. 2019;33(2):523-528. doi:10.21873/invivo.11505

40. Fuchs G, Thevathasan T, Chretien YR, et al. Lumbar skeletal muscle index derived from routine computed tomography exams predict adverse post-extubation outcomes in critically ill patients. *J Crit Care*. 2018;44:117-123. doi:10.1016/j.jcrc.2017.10.033

41. Abramowitz MK, Hall CB, Amodu A, Sharma D, Androga L, Hawkins M. Muscle mass, BMI, and mortality among adults in the United States: A population-based cohort study (PLoS ONE (2018) 13:4 (e0194697) DOI: 10.1371/journal.pone.0194697). *PLoS One*. 2018;13(5):1-16. doi:10.1371/journal.pone.0198318

42. Shibahashi K, Sugiyama K, Kashiura M, Hamabe Y. Decreasing skeletal muscle as a risk factor for mortality in elderly patients with sepsis: A retrospective cohort study. *J Intensive Care*. 2017;5(1):1-7. doi:10.1186/s40560-016-0205-9

43. Mundi MS, Patel JJ, Martindale R. Body Composition Technology: Implications for the ICU. *Nutr Clin Pract*. 2019;34(1):48-58. doi:10.1002/ncp.10230

44. Earthman CP. Body composition tools for assessment of adult malnutrition at the bedside.

J Parenter Enter Nutr. 2015;39(7):787-822. doi:10.1177/0148607115595227

45. Malone A, Hamilton C. The academy of nutrition and dietetics/the american society for parenteral and enteral nutrition consensus malnutrition characteristics: Application in practice. *Nutr Clin Pract*. 2013;28(6):639-650. doi:10.1177/0884533613508435

46. Sheean P, Gonzalez MC, Prado CM, McKeever L, Hall AM, Braunschweig CA. American Society for Parenteral and Enteral Nutrition Clinical Guidelines: The Validity of Body Composition Assessment in Clinical Populations. *J Parenter Enter Nutr*. 2020;44(1):12-43. doi:10.1002/jpen.1669

47. Teigen LM, Kuchnia AJ, Mourtzakis M, Earthman CP. The Use of Technology for Estimating Body Composition: Strengths and Weaknesses of Common Modalities in a Clinical Setting. *Nutr Clin Pract*. 2017;32(1):20-29. doi:10.1177/0884533616676264

48. Mareschal J, Achamrah N, Norman K, Genton L. Clinical Value of Muscle Mass Assessment in Clinical Conditions Associated with Malnutrition. *J Clin Med*. 2019;8(7):1040. doi:10.3390/jcm8071040

49. Yap J, Rafii M, Azcue M, Pencharz P. Effect of Intravenous Infusion Solutions on Bioelectrical Impedance Spectroscopy. *J Parenter Enter Nutr*. 2017;41(4):641-646. doi:10.1177/0148607115619598

50. Lopez-Ruiz A, Kashani K. Assessment of muscle mass in critically ill patients: role of the sarcopenia index and images studies. *Curr Opin Clin Nutr Metab Care*. 2020;23(5):302-311. doi:10.1097/MCO.0000000000000673

51. Paris MT, Mourtzakis M, Day A, et al. Validation of Bedside Ultrasound of Muscle Layer Thickness of the Quadriceps in the Critically Ill Patient (VALIDUM Study). *J Parenter Enter Nutr*. 2017;41(2):171-180. doi:10.1177/0148607116637852

52. Hernández-Socorro CR, Saavedra P, López-Fernández JC, Ruiz-Santana S. Assessment of muscle wasting in long-stay ICU patients using a new ultrasound protocol. *Nutrients*. 2018;10(12):1-11. doi:10.3390/nu10121849

53. Formenti P, Umbrello M, Coppola S, Froio S, Chiumello D. Clinical review: peripheral muscular ultrasound in the ICU. *Ann Intensive Care*. 2019;9(1). doi:10.1186/s13613-019-0531-x

54. Wang ZM, Gallagher D, Nelson ME, Matthews DE, Heymsfield SB. Total-body skeletal muscle mass: Evaluation of 24-h urinary creatinine excretion by computerized axial tomography. *Am J Clin Nutr*. 1996;63(6):863-869. doi:10.1093/ajcn/63.6.863

55. Heymsfield SB, Arteaga C, McManus CM, Smith J, Moffitt S. Measurement of muscle mass in humans: Validity of the 24-hour urinary creatinine method. *Am J Clin Nutr*. 1983;37(3):478-494. doi:10.1093/ajcn/37.3.478

56. Schutte JE, Longhurst JC, Gaffney FA, Bastian BC, Blomqvist CG. Total plasma creatinine: An accurate measure of total striated muscle mass. *J Appl Physiol Respir Environ Exerc Physiol*. 1981;51(3):762-766. doi:10.1152/jappl.1981.51.3.762

57. Walser M. Creatinine excretion as a measure of protein nutrition in adults of varying age. *J Parenter Enter Nutr*. 1987;11(5 SUPPL.):73-78. doi:10.1177/014860718701100510

58. Wyss M, Kaddurah-Daouk R. Creatine and creatinine metabolism. *Physiol Rev*. 2000;80(3):1107-1213. doi:10.1152/physrev.2000.80.3.1107

59. Forbes GB, Bruining GJ. Urinary creatinine excretion and lean body mass. *Am J Clin Nutr*. 1976;29(12):1359-1366. doi:10.1093/ajcn/29.12.1359

60. Welle S, Thornton C, Totterman S, Forbes G. Utility of creatinine excretion in body-composition studies of healthy men and women older than 60 y. *Am J Clin Nutr*.

1996;63:151-156.

61. Virgili F, Maiani G, Zahoor ZH, Ciarapica D, Raguzzini A, Ferro-Luzzi A. Relationship between fat-free mass and urinary excretion of creatinine and 3-methylhistidine in adult humans. *J Appl Physiol*. 1994;76(5):1946-1950. doi:10.1152/jappl.1994.76.5.1946

62. Proctor DN, O'Brien PC, Atkinson EJ, Nair KS. Comparison of techniques to estimate total body skeletal muscle mass in people of different age groups. *Am J Physiol - Endocrinol Metab*. 1999;277:489-495. doi:10.1152/ajpendo.1999.277.3.e489

63. Udy AA, Scheinkestel C, Pilcher D, Bailey M. The association between low admission peak plasma creatinine concentration and in-hospital mortality in patients admitted to intensive care in Australia and New Zealand. *Crit Care Med*. 2016;44(1):73-82. doi:10.1097/CCM.0000000000001348

64. Cartin-Ceba R, Afessa B, Gajic O. Low baseline serum creatinine concentration predicts mortality in critically ill patients independent of body mass index. *Crit Care Med*. 2007;35(10):2420-2423. doi:10.1097/01.CCM.0000281856.78526.F4

65. Thongprayoon C, Cheungpasitporn W, Kittanamongkolchai W, Harrison AM, Kashani K. Prognostic Importance of Low Admission Serum Creatinine Concentration for Mortality in Hospitalized Patients. *Am J Med*. 2017;130(5):545-554.e1. doi:10.1016/j.amjmed.2016.11.020

66. Hessels L, Koopmans N, Gomes Neto AW, et al. Urinary creatinine excretion is related to short-term and long-term mortality in critically ill patients. *Intensive Care Med*. 2018;44(10):1699-1708. doi:10.1007/s00134-018-5359-6

67. Khan J, Bath K, Hafeez F, Kim G, Pesola GR. Creatinine excretion as a determinant of accelerated skeletal muscle loss with critical illness. *Turk Anesteziyoloji ve Reanimasyon*

Dern Derg. 2018;46(4):311-315. doi:10.5152/TJAR.2018.60437

68. Ter Maaten JM, Damman K, Hillege HL, et al. Creatinine excretion rate, a marker of muscle mass, is related to clinical outcome in patients with chronic systolic heart failure. *Clin Res Cardiol*. 2014;103(12):976-983. doi:10.1007/s00392-014-0738-7

69. Bistrian BR, Blackburn GL, Sherman M, Scrimshaw NS. Therapeutic index of nutritional depletion in hospitalized patients. *Surgery, Gynecol Obs*. 1975;141:512-516.

70. Blackburn G l., Bistrian BR, Maini BS, Schlamm HT, Smith MF. Nutritional and metabolic assessment of the hospitalized patient. *J Parenter Enter Nutr*. 1977;1(1):11-21. doi:10.1177/014860717700100101

71. Daley BJ, Maliakkal RJ, Dreesen EB, Driscoll DF, Bistrian BR. Rapid clinical assessment of kidney function based on arm muscle circumference and serum creatinine. *Nutrition*. 1994;10(2):128-131.

72. Weisberg H. Evaluation of nutritional status. *Ann Clin Lab Sci*. 1983;13(2):95-105.

73. Datta D, Foley R, Wu R, Grady J, Scalise P. Can creatinine height index predict weaning and survival outcomes in patients on prolonged mechanical ventilation after critical illness? *J Intensive Care Med*. 2018;33(2):104-110. doi:10.1177/0885066616648133

74. Selberg O, Sel S. The adjunctive value of routine biochemistry in nutritional assessment of hospitalized patients. *Clin Nutr*. 2001;20(6):477-485. doi:10.1054/clnu.2001.0427

75. Tosato M, Marzetti E, Cesari M, et al. Measurement of muscle mass in sarcopenia: from imaging to biochemical markers. *Aging Clin Exp Res*. 2017;29(1):19-27. doi:10.1007/s40520-016-0717-0

76. Carney DE, Meguid MM. Current concepts in nutritional assessment. *Arch Surg*. 2002;137(1):42-45. doi:10.1001/archsurg.137.1.42

77. Schwebel C, Pin I, Barnoud D, et al. Prevalence and consequences of nutritional depletion in lung transplant candidates. *Eur Respir J*. 2000;16(6):1050-1055. doi:10.1034/j.1399-3003.2000.16f05.x

78. Medhat AS, Ahmed AO, Ahmed FT, Amal MAA. Creatinine height index as a predictor of nutritional status among patients with liver cirrhosis. *J Public Heal Epidemiol*. 2016;8(10):220-228. doi:10.5897/jphe2016.0850

79. Apelgren K, Rombeau J, Twomey P, Miller R. Comparison of nutritional indices and outcones in critically ill patients. *Crit Care Med*. 1982;10(5):305-307.

80. Gerber LM, Mann SJ. Development of a model to estimate 24-hour urinary creatinine excretion. *J Clin Hypertens*. 2014;16(5):367-371. doi:10.1111/jch.12294

81. John KA, Cogswell ME, Campbell NR, et al. Accuracy and Usefulness of Select Methods for Assessing Complete Collection of 24-Hour Urine: A Systematic Review. *J Clin Hypertens*. 2016;18(5):456-467. doi:10.1111/jch.12763

82. Jędrusik P, Symonides B, Gaciong Z. Performance of 24-hour urinary creatinine excretion-estimating equations in relation to measured 24-hour urinary creatinine excretion in hospitalized hypertensive patients. *Sci Rep*. 2019;9(1):1-11. doi:10.1038/s41598-019-40416-w

83. Bistrian BR, Mogensen KM, Christopher KB. Plea for Reapplication of Some of the Older Nutrition Assessment Techniques. *J Parenter Enter Nutr*. 2020;44(3):391-394. doi:10.1002/jpen.1808

84. Bistrian BR. A Catabolic Index Adjusted for the Creatinine Height Index: Can It Help in Nutrition Assessment? *J Parenter Enter Nutr*. 2020;44(8):1376-1377. doi:10.1002/jpen.1942

85. Bistrian BR. A simple techinque to estimate severity of stress. *Surgery, Gynecol Obs*. 1979;148:675-678.

86. Fabri PJ. Analysis by modeling data. In: *Measurement and Analysis in Transforming Healthcare Delivery*. Switzerland: Springer International Publishing; 2016:93-100. doi:10.1007/978-3-319-40812-5

87. Verma B, Ray S, Srivastava R. Mathematical models and their applications in medicine and health. *Heal Popul Perspect Issues*. 1981;4(1):42-58.

88. Marion G. An introduction to mathematical modelling. 2015 https://people.maths.bris.ac.uk/~madjl/course_text.pdf.

89. Branham ML, Ross EA, Govender T. Predictive models for maximum recommended therapeutic dose of antiretroviral drugs. *Comput Math Methods Med*. 2012;2012. doi:10.1155/2012/469769

90. Grima DT, Webb GF, D'Agata EMC. Mathematical model of the impact of a nonantibiotic treatment for Clostridium difficile on the endemic prevalence of vancomycin-resistant enterococci in a hospital setting. *Comput Math Methods Med*. 2012;2012. doi:10.1155/2012/605861

91. Su CT, Chen KH, Chen LF, Wang PC, Hsiao YH. Prediagnosis of obstructive sleep apnea via multiclass MTS. *Comput Math Methods Med*. 2012;2012. doi:10.1155/2012/212498

92. Bazan I, Ramos A, Calas H, et al. Possible patient early diagnosis by ultrasonic noninvasive estimation of thermal gradients into tissues based on spectral changes modeling. *Comput Math Methods Med*. 2012;2012. doi:10.1155/2012/275405

93. Ghosh A, Berger I, Remien CH, Mubayi A. The role of alcohol consumption on acetaminophen induced liver injury: Implications from a mathematical model. *J Theor*

Biol. 2021;519:110559. doi:10.1016/j.jtbi.2020.110559

94. Remien CH, Adler FR, Waddoups L, Box TD, Sussman NL. Mathematical modeling of liver injury and dysfunction after acetaminophen overdose: Early discrimination between survival and death. *Hepatology*. 2012;56(2):727-734. doi:10.1002/hep.25656

95. Kucharski AJ, Russell TW, Diamond C, et al. Early dynamics of transmission and control of COVID-19: a mathematical modelling study. *Lancet Infect Dis*. 2020;20(5):553-558. doi:10.1016/S1473-3099(20)30144-4

96. Doan TN, Kong DCM, Kirkpatrick CMJ, McBryde ES. Optimizing Hospital Infection Control: The Role of Mathematical Modeling. *Infect Control Hosp Epidemiol*. 2014;35(12):1521-1530. doi:10.1086/678596

97. Shine D. Risk-adjusted mortality: Problems and possibilities. *Comput Math Methods Med*. 2012;2012. doi:10.1155/2012/829465

98. Buzby GP, Mullen JL, Matthews DC, Hobbs CL, Rosato EF. Prognostic nutritional index in gastrointestinal surgery. *Am J Surg*. 1980;139(1):160-167. doi:10.1016/0002-9610(80)90246-9

99. Buzby GP, Blouin G, Colling CL, Crosby LO. Perioperative total parenteral nutrition in surgical patients. *N Engl J Med*. 1991;325(8):525-531.

100. Baker JP, Detsky AS, Wesson DE, et al. Nutritional assessment a comparison of clinical judgment and objective measures. *N Engl J Med*. 1982;306(16):969-972.

101. Fabri PJ. Principles of supervised learning. In: *Measurement and Analysis in Transforming Healthcare Delivery*. Switzerland: Springer International Publishing; 2016:101-120. doi:10.1007/978-3-319-40812-5

102. Carcione JM, Santos JE, Bagaini C, Ba J. A Simulation of a COVID-19 Epidemic Based

on a Deterministic SEIR Model. *Front Public Heal*. 2020;8(May). doi:10.3389/fpubh.2020.00230

103. Irurzun-Arana I, Rackauckas C, McDonald TO, Trocóniz IF. Beyond Deterministic Models in Drug Discovery and Development. *Trends Pharmacol Sci*. 2020;41(11):882-895. doi:10.1016/j.tips.2020.09.005

104. Torres N V., Santos G. The (mathematical) modeling process in biosciences. *Front Genet*. 2015;6(DEC):1-9. doi:10.3389/fgene.2015.00354

105. Heyland DK, Dhaliwal R, Jiang X, Day AG. Identifying critically ill patients who benefit the most from nutrition therapy: the development and initial validation of a novel risk assessment tool. *Crit Care*. 2011;15(6). doi:10.1186/cc10546

106. Cohen J. A Power Primer. *Psychol Bull*. 1992;112(1):155-159. doi:10.1037/0033-2909.112.1.155

107. Green SB. How many subjects does it take to do a regression analysis. *Multivar Behav Reserach*. 1991;26(3):499-510.

108. Bonett DG, Wright TA. Sample Size Requirements for Estimating Pearson, Kendall and Spearman Correlations. *Psychometrika*. 2000;65(1):23-28.

109. Cassidy R, Singh NS, Schiratti PR, et al. Mathematical modelling for health systems research: A systematic review of system dynamics and agent-based models. *BMC Health Serv Res*. 2019;19(1):15-17. doi:10.1186/s12913-019-4627-7

110. López-Palau NE, Olais-Govea JM. Mathematical model of blood glucose dynamics by emulating the pathophysiology of glucose metabolism in type 2 diabetes mellitus. *Sci Rep*. 2020;10(1):1-11. doi:10.1038/s41598-020-69629-0

111. van Riel NAW. Dynamic modelling and analysis of biochemical networks: Mechanism-

based models and model-based experiments. *Brief Bioinform*. 2006;7(4):364-374. doi:10.1093/bib/bbl040

112. Doolan PD, Alpen EL, Theil GB. A clinical appraisal of the plasma concentration and endogenous clearance of creatinine. *Am J Med.* 1962;32:65-79. doi:10.1016/0002-9343(62)90183-3

113. Narayanan S, Appleton HD. Creatinine: a review. *Clin Chem*. 1980;26:1119-1126. https://doi.org/10.1093/clinchem/26.8.1119

114. Bhave G, Neilson EG. Body fluid dynamics: back to the future. *J Am Soc Nephrol*. 22(12):2166-2181. doi:10.1681/ASN.2011080865

115. Lambell KJ, Earthman CP, Tierney AC, Goh GS, Fortsyth A, King SJ. How does muscularity assessed by bedside methods compare to computed tomography muscle area at intensive care unit admission? A pilot prospective cross-sectional study. *J Hum Nutr Diet*. 2021;34:345-355. doi:10.1111/jhn.12804

116. Knaus WA, Draper EA, Wagner DP, Zimmerman JE. APACHE II: a severity of disease classification system. *Crit Care Med*. 1985;13(10):818-829.

117. Jones AE, Trzeciak S, Kline JA. The Sequential Organ Failure Assessment score for predicting outcomes in patients with severe sepsis and evidence of hypoperfusion at the time of emergency department presentation. *Crit Care Med*. 2009;37(5)1649-1654. doi:10.1097/CCM.0b013e31819def97

118. Evans DC, Corkins MR, Malone A, et al. The use of visceral proteins as nutrition markers: an ASPEN position paper. *Nutr Clin Pract*. 2021;36(1):22-28. doi:10.1002/ncp:10588

119. Soeters PB, Rolfe RR, Shenkin A. Hypoalbuminemia: pathogenesis and clinical significance. *JPEN J Parenter Enteral Nutr*. 2019;43(2):181-193. doi:10.1002/jpen.1451

120. Doweiko JP, Nompleggi DJ. The role of albumin in human physiology and pathophysiology. Part III: albumin and disease states. *JPEN J Parenter Enteral Nutr*. 1991;15(4):476-483.

121. Davis CJ, Sowa D, Keim KS, Kinnare K, Peterson S. The use of pre-albumin and C-reactive protein for monitoring nutrition support in adult patients receiving enteral nutrition in an urban medical center. *JPEN J Parenter Enteral Nutr*. 2012;36(2)197-204.

122. Yeh DD, Johnson E, Harrison T, et al. Serum levels of albumin and prealbumin do not correlate with nutrient delivery in surgical intensive care unit patients. *Nutr Clin Pract.* 2018;33(3)419-425.

123. Dioguardi FS. Low plasma albumin levels should be interpreted, but not ignored. *Nutr Clin Pract.* 2021;36(2):502-503. doi:10.1002/ncp.10619

124. Lew CCH, Yandell R, Fraser RJL, Chua AP, Chong MFF, Miller M. Association between malnutrition and clinical outcomes in the intensive care unit: a systemic review. *JPEN J Parenter Enteral Nutr*. 2017;41(5):744-758. doi:10.1177/0148607115625638

125. Hiura G, Lebwohl B, Seres DS. Malnutrition diagnosis in critically ill patients using 2012 Academy of Nutrition and Dietetics / American Society of Parenteral and Enteral Nutrition standardized diagnostic characteristics is associated with longer hospital and intensive care unit length of stay and increased in-hospital mortality. *JPEN J Parenter Enteral Nutr*. 2020;44(2)256-264. doi: 10.1002.jpen.1599

Appendix A. IRB Approval

October 12, 2021

Regarding: Protocol amendment

Study Title: Protein needs of the critically ill

PI: Lynn D. Hiller

IRB#: Pro00010950

VA IRBNet # 1573343-2

Dear RDC Chair,

I am requesting several modifications to my open exempt retrospective study referenced above. These include modifications to personnel, date range, sample size, collection of additional laboratory values and the collection of outcome data. Below are the specifics.

The addition of the following personnel:

- Susan Goldsmith MD, as an additional PI, is a staff gastroenterologist with training in nutrition.
- James Epps PhD, from USF, as a consultant, has experience in nutrition research to aid in the interpretation of findings.
- Lauri Wright PhD, RD, from UNF, as a consultant, has extensive experience in nutrition research to aid in interpretation of findings and implications on future practice.

Date range and sample size:

- To expand the date range from the previous date range of 01/01/2010 to 04/30/2013 to the new range of 01/01/2010 to 09/30/2021.
- To increase sample size from 100 to 150 subjects

Labs and Acuity Data: to additionally collect serum glucose, BUN, creatinine, sodium, potassium, chloride, carbon dioxide, albumin, prealbumin and NUTIRC score that corresponds to the day of the other data collection.

Outcome data: to collect length of stay, intensive care unit length of stay, hospital mortality and 6-month mortality for the subjects.

Thank you for your time and consideration.

Lynn Hiller

DEPARTMENT OF VETERANS AFFAIRS
James A. Haley Veterans Hospital Research and Development Committee
James A. Haley Veterans Hospital

Date:	**December 8, 2021**
To:	**Lynn Hiller, MS, RD** Principal Investigator
From:	Research & Development Committee (R&DC) Chair
Protocol Title:	**[1573343-5] Protein Needs of the Critically Ill**
Submission Type:	**Amendment/Modification**
Review Type:	**Designated Review**
Action:	**Amendment Approved**
Effective Date:	**December 8, 2021**

+++This letter is only for studies where the R&DC is the sole oversight committee or when the modification requires RDC review per your local SOP +++

The amendment to the above-named project was reviewed and **approved** by the R&D Committee Chair or designee on via designated review procedures. This approval will be reported to the committee during the next convened R&D Committee meeting.

The R&DC reminds you of the following requirements:

1. Any changes to the protocol must be proposed to the R&DC in writing as an amendment to the approved project via IRBNet and must be approved before changes are implemented.
2. You are required to submit reportable events, UPIRTSOs, and DSMB reports, as applicable per timeframes defined in the local SOP.
3. Acknowledgment of the VA's contribution is required in any publications and presentations that may result from this research.

The following documents were reviewed:

- Conflict of Interest - Declaration - S Goldsmith MD Conflict of Interest 2021.pdf (UPLOADED: 10/12/2021)
- Data Collection - Hiller Track protein data collection form 9-14-21 version 3.docx (UPLOADED: 10/12/2021)
- Data Collection - Hiller Clean protein data collection form 9-14-21 version 3.docx (UPLOADED: 10/12/2021)
- Letter - Hiller Study amendment memo.pdf (UPLOADED: 10/12/2021)
- Other - Goldsmith PI Request Form.pdf (UPLOADED: 10/15/2021)
- Other - Epps PI Request Form.pdf (UPLOADED: 10/13/2021)

- Other - Wright L PI Request Form.pdf (UPLOADED: 10/13/2021)
- Other - Hiller Protein Amended (PSSF)_9.2021.pdf (UPLOADED: 10/12/2021)
- Protocol - Hiller Protocol 09-05-2021 version09 track.docx (UPLOADED: 10/12/2021)
- Protocol - Hiller Protocol 09-05-2021 version09 clean.docx (UPLOADED: 10/12/2021)

If you have any questions, please contact **Stanley Pettermon** at or **stanley.pettermon@va.gov**. Please include your project title and reference number in all correspondence with this committee.

SHANNON R. MILES 166807

Digitally signed by SHANNON R. MILES 166807
Date: 2021.12.17 16:05:53 -05'00'

Dr. Bradley Stein
Research & Development Committee Chair

This electronically generated document serves as official notice to sponsors and others of approval, disapproval or other RDC decisions. Only those individuals who have been granted authority by the institution to create letters on behalf of the RDC are able to do so. A copy of this document has been retained within the James A. Haley Veterans Hospital IRBNet records. The IRBNet System is fully compliant with the technology requirements for Electronic Records per CFR 21, Part 11, Section 11.10 - Controls for Closed Systems, and the technology requirements for Electronic Signatures per CFR 21, Part 11 Subpart C - Electronic Signatures.