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Title: Health and nutrition of loggerhead sea turtles (*Caretta caretta*) in the southeastern United States

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Abstract

Loggerhead sea turtles (*Caretta caretta*) are opportunistic carnivores that feed primarily on benthic invertebrates and fish. Sea turtle rehabilitation requires provision of a species-specific, balanced diet that supplies nutrition similar to that of a wild diet; this can be challenging because free-ranging loggerheads' diets vary depending on their life stage and geographic location, with predominant prey species dictated by local availability. The goal of this study was to better understand the nutritional needs of subadult and adult loggerheads in rehabilitation. This was accomplished by conducting a retrospective survey of stomach contents identified during gross necropsy of 153 deceased loggerheads that stranded in coastal Georgia, USA. A total of 288 different forage items were identified; the most frequently observed prey items belong to the subphylum Crustacea (N=131), followed by bony fish (Osteichthyes; N=45), gastropod mollusks (N=40), bivalve mollusks (N=23), and Atlantic horseshoe crabs (*Limulus polyphemus;* N=15). The proportions of certain prey items differed significantly with turtle size; adult turtles ate proportionately more gastropods (P=0.001), and subadults ate proportionately more fish (P=0.01). Stomach contents information was used to determine common local prey items (blue crab, cannonball jellyfish, horseshoe crab, whelk), which were evaluated for nutritional content.

Additionally, we compared hematology and plasma biochemistry profiles (including proteins, trace minerals, and vitamins) between four cohorts of loggerhead turtles, including free-ranging subadults and adults, nesting females, and loggerheads undergoing rehabilitation. This information was applied to inform a regionally specific, formulated diet for tube feeding, and a supplement containing vitamins and minerals for captive loggerheads, to more closely approximate the nutritional content of their natural diet. Assessing the regional and temporal variability in loggerhead diets is an important component in their effective conservation because resultant data can be used to help understand the impacts of environmental perturbations on benthic food webs.

Keywords: Dietary supplement, hematology, nutrition, plasma biochemistry, prey items, rehabilitation

1. Introduction

Loggerhead sea turtles (*Caretta caretta*) are the most commonly occurring sea turtle species inhabiting the coastal waters of Georgia, USA (Norton, 2005). The turtles in this region comprise part of the Northwest Atlantic Distinct Population Segment of loggerheads inhabiting the waters of the eastern coast of the United States and are listed as Threatened by the United States Endangered Species Act (Conant et al., 2009; Wallace et al., 2010). Understanding sea turtle diets is an important component in their effective conservation. Previous studies demonstrate that loggerhead turtles are opportunistic carnivores that feed primarily on benthic invertebrates and fish (Donaton, Durham, Cerrato, Schwerzmann, & Thorne, 2019; Frick, Williams, Bolten, Bjorndal & Martins 2009; Tomas, Aznar, & Raga 2001). While individual loggerheads have been observed to specialize in a consistent mixture of prey species, considerable regional variability has been demonstrated, with predominant prey species dictated by local availability and ranging widely from crabs to jellyfish to bivalves (Frick et al, 2009; Lazar et al., 2011; Revelles, Cardona, Aguilar, San Félix, & Fernandez 2007). Changes in loggerhead diets over time have been associated with fishing pressure on benthic prey species (Seney & Musick 2007). Assessing the variability in loggerhead diets over time can help us understand the impacts of environmental perturbations on benthic food webs and maintain a current knowledge base about regional changes in their diet composition (Donaton et al., 2019).

The Georgia Sea Turtle Center (GSTC) is a rehabilitation facility on Jekyll Island, Georgia that serves to triage and rehabilitate sea turtles (Norton 2005). Part of the rehabilitation process is to provide a species-specific, balanced diet that provides nutrition similar to that of a wild diet to allow injured, ill, malnourished and debilitated turtles to gain weight and improve body condition. Because free-ranging loggerheads consume a variety of food items that vary depending on life stage and geographic location, providing appropriate food items and developing a nutritionally balanced gruel or gelatin-based formula for debilitated loggerheads undergoing rehabilitation is challenging (Jones & Seminoff 2013). Moreover, there is limited information regarding the nutritional needs of this species. Beyond considering the identity of loggerhead diet items, it is important to also evaluate the nutritional content and energetic contribution of their diet. For this study, we hypothesized that analyzing the stomach contents and clinical pathology data of wild loggerhead turtles would enable development of nutritional indices which could be applied to better address the dietary needs of captive loggerheads. Thus, the goal of this study was to better understand the nutritional needs of loggerheads in rehabilitation. This was accomplished by conducting a retrospective survey of stomach contents identified during gross necropsy of deceased loggerheads that stranded in coastal Georgia. This information was used to determine common local prey items, which were evaluated for nutritional content. Additionally, we compared clinical blood data from four cohorts of loggerhead sea turtles including free-ranging subadults and adults, nesting females, and loggerheads undergoing rehabilitation at GSTC. These comparisons allow us to relate different life history stages to differences in blood health analytes, including several nutritional parameters not previously reported for loggerheads in this region. Clinicians prefer species-specific baseline clinical pathology data in reptiles due to the diverse environmental conditions and life history stages that can affect these data in poikilothermic species (Lewbart et al., 2014). Ultimately, this baseline information, and information about commonly fed food items for loggerheads under human care, were applied to inform a regionally specific, formulated diet for tube feeding, and a supplement containing vitamins and minerals for captive loggerheads, to more closely approximate the nutritional content of their natural diet.

2. Materials and Methods

2.1. Retrospective analysis of stranded loggerhead stomach contents

Georgia Department of Natural Resources (Brunswick, Georgia USA) necropsy records were reviewed for observations of identifiable prey items found within the stomachs of deceased loggerheads that stranded along the Georgia coastline during 1998–2008. Turtle age class was determined based on curved carapace length (CCL); turtles were designated as subadults (<87 cm) or adults (≥87 cm) (Deem et al., 2009). Prey items were categorized based on lowest taxonomic groupings, ascertained from the available data in the necropsy reports. Stomach contents observations were grouped by relative abundance (i.e., number of each species) and richness (i.e., number of different species) based on year, month, turtle size class, and body condition (good/poor).

2.2. Prey item nutritional analysis

Several common loggerhead prey items obtained locally in coastal Georgia, including horseshoe crab (*Limulus polyphemus*), whelk (*Busycon* spp., *Busycotypus* spp.), blue crab (*Callinectes sapidus*), and cannonball jellyfish (*Stomolophus meleagris*), were submitted for whole body analysis of proximate composition (moisture, crude protein, crude fat, ash) at the Dairy One Laboratory (Ithaca, New York USA). Prey items were analyzed for macro minerals (calcium, chlorine, magnesium, phosphorus, potassium, sodium, sulfur), trace minerals (cobalt, copper, iron, manganese, molybdenum, selenium, zinc), and vitamins (A, E) at the Diagnostic Center for Population and Animal Health at Michigan State University (Lansing, Michigan USA) (Ernst & Jovich, 2009).

2.3. Live turtles and sample collection

Blood samples were collected from four cohorts of loggerhead turtles during 2008–2011: adults and subadults captured for in-water studies, nesting females, and live-stranded adults and subadults admitted to rehabilitation at GSTC. Free-ranging loggerheads were captured by fishery-independent trawlers during June–July each year as part of a long-term sea turtle abundance, demographic, and health assessment (Arendt et al., 2012). Turtles were captured in 4.5–12.2 m-deep, near-shore waters between Savannah, Georgia (32°5’N, 81°5’W) and St. Augustine, Florida USA (29°50’N, 81°15’W) using 20-m, four-seam trawl nets with 20-cm mesh without turtle excluder devices; maximum trawl duration was 30 minutes per trawl (Arendt et al., 2012). Nesting adult females were sampled on the beaches of Jekyll Island (31°4’N, 81°25’W) during May–August of 2008–2011 during oviposition. Live-stranded loggerheads were rescued along the coasts of Georgia and Florida and taken to GSTC for medical evaluation and treatment.

Turtle age class was assigned based on the presence of a long tail in adult males, nesting in adult females, or by CCL as described above (Deem et al., 2009). For all turtles, using aseptic technique, 5–10 ml of blood was collected from the external jugular vein using a 20- or 22-gauge, 1.5 inch needle attached to a syringe. Blood was distributed into lithium heparin-coated Vacutainer® tubes (Becton Dickinson and Company, Franklin Lakes, New Jersey USA) and kept cool until processing, which occurred within 4 hours of collection for field-sampled turtles and immediately for rehabilitating turtles. Whole blood was reserved for packed cell volume (PCV) and two blood films, and the remaining blood was centrifuged at 3,500 rpm for 10 minutes and resulting plasma was aliquoted into cryovials. All plasma samples were stored frozen at –70°C for up to 6 months prior to analysis. Stranded loggerheads were released following a rehabilitation period (3–13 months), when deemed clinically healthy. A single blood sample was collected from free-ranging turtles, while paired samples (taken at admission and prior to release) were collected from rehabilitating turtles. The authors confirm that the ethical policies of the journal, as noted on the journal's author guidelines page, have been adhered to and the appropriate ethical review committee approval has been received. The authors confirm that they have followed EU standards for the protection of animals used for scientific purposes.

2.4. Blood sample analysis

Whole blood samples in capillary tubes were centrifuged for 5 minutes at 1,300 *g* (5,000 rpm) in a microhematocrit centrifuge and PCV (%) was interpreted using a hematocrit microcapillary tube reader. After centrifugation, plasma color was assessed visually for hemolysis. Plasma total protein concentration was determined by refractometer. Blood cell counts were conducted at GSTC by a certified veterinary technician, including erythrocyte (RBC) counts using the BD Unopette® brand test for manual RBC counts (Becton-Dickinson Diagnostics, Pre-analytical Systems), and total estimated white blood cell (tWBC) counts performed manually on blood films using the formula WBC estimate/µl = [average WBC/HPF] × objective power (Bjorndal 1997; Stacy & Innis 2017). Leukocyte morphology was also evaluated.

Plasma samples were evaluated for concentrations of glucose, blood urea nitrogen (BUN), creatinine, CO_2, sodium, potassium, amylase, lipase, calcium, phosphorus, cholesterol, triglycerides, high-density lipoproteins (HDL), low-density lipoproteins (LDL), very low-density lipoproteins (VLDL), uric acid, total protein, amylase aspartate aminotransferase (AST), alanine aminotransferase (ALT), lactate dehydrogenase (LDH), creatinine kinase (CK), and gamma glutamyl transferase (GGT) using standard dry-slide determinations with a Kodak 700XR™ chemical analyzer at the Comparative Pathology Laboratory, University of Miami (Miami, Florida USA [UMCPL]). Plasma protein fractions were measured using electrophoresis for total protein, pre-albumin, albumin, α_1-globulins, α_2-globulins, β-globulins, and γ-globulins (UMCPL, Miami, Florida USA) (Zais & Cray 2002). Plasma macro and trace mineral values (calcium, chlorine, cobalt, copper, iron, magnesium, manganese, molybdenum, phosphorus, potassium, selenium, sodium, sulfur, zinc) were measured using inductively coupled argon plasma-mass spectrometry (ICP-MS) at the Toxicology Laboratory of the New Bolton Center at the University of Pennsylvania (Kennett Square, Pennsylvania USA) (Chaffin et al., 2008). For all minerals, the detection limit was 0.05 μg/ml. Plasma vitamin D (25-hydroxycholecalciferol) was measured by radioimmunoassay at Boston University School of Medicine (Boston, Massachusetts USA) using previously described methods (Chen, Turner, & Holick 1990). Plasma vitamin A (retinol) and vitamin E (α−tocopherol) concentrations were determined using high-performance liquid chromatography at Mystic Aquarium (Mystic, Connecticut USA) (Catignani & Bieri 1983).

2.5. Statistical analysis

All blood analyte values were assessed for normality using Shapiro-Wilkes tests. Because data distributions violated the assumptions of normality, non-parametric tests were used. Kruskal-Wallis H-tests were used to compare clinical blood data between loggerheads entering rehabilitation versus ‘healthy’ free-ranging subadult and adult (including nesting female) loggerheads. Pearson’s correlation coefficients were calculated between the number of turtle strandings, prey item relative abundance, and prey item richness data per year, month, and size class. N–1 chi-squared tests were used to compare the proportions of larger taxonomic categories of prey items between turtles that stranded in warmer versus cooler months, between age classes, and between turtles in ‘good’ versus ‘poor’ body condition (Richardson 2011). All statistical analyses were conducted using IBM SPSS Statistics for Windows, v.26 (IBM Corp., Armonk, New York USA), with $\alpha = 0.05$.

3. Results

3.1. Stomach contents and prey item analysis

Of 1,628 necropsy records reviewed, stomach contents were documented for 153 deceased loggerheads that stranded along the coast of Georgia during 1998–2008, including 123 subadults, 19 adults, and 11 turtles of unknown/undocumented age class. Eighty-three turtles (54.2%) were considered to be in 'good' body condition at the time of death based on ample body fat; while 11 turtles (7.2%) were determined to be in 'poor' body condition since they were thin or emaciated, and in some cases with high epibiota loads. There were 59 turtles (38.6%) in 'unknown' body condition due to a paucity of data in the necropsy records.

Results of the stomach contents analysis are presented in **Table 1**. A total of 288 different forage items were identified. Of the 153 loggerheads, 76 (49.7%) had only one kind of identifiable forage item in their gastrointestinal tract, 42 (27.4%) had two, 22 (14.4%) had three, five (3.2%) had four, four (2.6%) had five, two (1.3%) had six, one (0.7%) had seven, and one (0.7%) had eight different kinds of forage items. Two turtles had hooks in their stomachs, and two had small pieces of plastic in their gastrointestinal tract. The most frequently observed prey items are classified in the subphylum Crustacea (N=131), including 106 crabs and 24 shrimp. The second most frequently observed prey item was bony fish (N=45); however, due to decomposition, no fish were identified below the taxonomic level of superclass Osteichthyes. The third most frequently observed group of prey items was gastropod mollusks (N=40), and whelks were the most frequently observed type of gastropod (N=19). The fourth most frequently observed group of prey items was bivalve mollusks (N=23), and the fifth was Atlantic horseshoe crabs (*Limulus polyphemus*, N=15). Unidentifiable stomach contents and other foreign materials were sparsely reported and are not included here.

Number of stranded loggerheads per year (**Figure 1**) and per month (**Figure 2**) during 1998–2008 are plotted alongside relative abundance and species richness of prey items identified in the stomach contents of the turtles. Pearson's correlation coefficients indicated that both relative abundance and richness of prey items were strongly correlated to the number of stranded turtles per year (abundance $r(8)=0.99$, $P<0.001$; richness $r(8)=0.96$, $P<0.001$) and per month

($r(9)=0.95$, $P<0.001$; $r(9)=0.78$, $P<0.01$). The results of N–1 chi-squared tests to compare proportions of broader taxonomic categories in stomach contents of 22 loggerheads that stranded in cooler months (September–January) and 131 loggerheads that stranded in warmer months (April–August) are shown in **Table 2**; none of the proportions examined significantly differed between seasons.

Both relative abundance and richness of prey items seemingly correlated to the number of stranded turtles per size class (**Figure 3**), although correlation coefficients were not calculated due to low sample sizes for adult turtles. The results of N–1 chi-squared tests to compare proportions of broader taxonomic categories in stomach contents of 19 adults and 123 subadults are shown in **Table 2**. The proportion of certain prey items differed significantly with turtle size; adult turtles ate proportionately more gastropods, and subadults ate proportionately more fish. None of the other proportions significantly differed between size classes. No gastropods, bivalve mollusks, or mixed invertebrates were identified in stomach contents of turtles in 'poor' body condition, and turtles in 'good' body condition ate proportionately more gastropods (**Table 2**).

Prey item nutritional analysis results are presented in **Table 3**. Crude protein levels (dry matter basis) in prey ranged from to 11.4% (blue crab) to 73.7% (horseshoe crab), while crude fat content was generally low (1.3–2.6%). Whelks are a rich source of calcium (24.2 %) and ash (63.6%) due to their calcium carbonate shells. Cannonball jellyfish had the highest moisture content (94.7%) of the four prey items based on the analytes presented here. Vitamins A and E were only measured in horseshoe crabs; vitamin E was detected at 3.53 mg/kg, but vitamin A concentration was below the assay detection limit which is typical of invertebrates that are low in fat (Pfaller et al., 2020).

An average consumption of target nutrients in a wild loggerhead diet, including protein, fat, calcium, copper, iodine, iron, manganese, phosphorus, zinc, vitamins A, E, C, and thiamine, were calculated based on these data and previously published nutrient data, assuming a consumption of 30% crab, 40% fish and 30% gastropods and mollusks (Hoopes, Koutsos, & Norton 2017). Additionally, estimated consumption of the same micronutrients was calculated based on a potential diet of loggerheads under human care, of 80% fish and 20% squid (Hoopes et al., 2017). Average nutrient values for fish and squid were derived from previously reported data (Hoopes et al., 2017). Nutrient compositions for both diets are presented in **Table 5**. A vitamin/mineral supplement and a critical care diet were designed based on these data (Mazuri 5B48 Sea Turtle Supplement and Mazuri 5S94 Sea Turtle Meal Diet for Carnivorous Turtles).

3.2. Live turtle blood sample analysis

Blood samples were collected from 54 free-ranging turtles, including 15 subadults and 24 adults captured in trawl nets, 15 nesting adult females, and from 15 live-stranded turtles admitted to the GSTC for rehabilitation. All free-ranging turtles were deemed healthy based on behavioral observation, physical exam, and body condition; all live-stranded loggerheads were determined to be clinically unhealthy based the same criteria. Reasons for stranding included abnormal neurologic status, coelomitis, cold-stunning, debilitation, gastric ulceration, and trauma. Selected morphometric data are presented in **Table 4**. Based on life history stages and assumed physiological states including foraging in trawl-captured turtles and capital breeding in nesting turtles, the nesting and live-stranded turtles are predicted to be in a fasted state, and the trawl-captured turtles are predicted to be in a 'fed' state (Perrault & Stacy 2018; Pfaller et al., 2020; Stacy et al., 2018). Blood results for free-ranging subadult and adult, nesting, and sick/rehabilitating turtles, including the results of the Kruskal-Wallis H-tests, are presented in **Table 6**.

4. Discussion

4.1. Loggerhead diet

Analysis of necropsy records reporting stomach contents of deceased loggerheads that stranded in Georgia during the study period revealed that crabs (Brachyura) were by far the most common prey item, followed by fish (Osteichthyes), shrimp (*Penaeidae*), gastropods (e.g., *Busycon* spp.), horseshoe crabs, bivalve mollusks (e.g., *Spisula solidissima*), and other invertebrates including tunicates, sponges, sea cucumbers, and soft coral. These data support previous observations that loggerhead sea turtles are opportunistic carnivores that feed in the benthic zone of coastal continental shelf areas, as well as within the water column (Youngkin, 2001). These data also represent a continuation of previous work on loggerhead diet data from turtles that stranded on Cumberland Island, Georgia during 1979–1999 (Youngkin, 2001; Youngkin & Wyneken, 2005). In both studies, crabs were the most commonly observed prey item in loggerhead stomach contents. Whereas Youngkin (2001) found that mollusks and fish were the next two consecutive most common prey items, here we found that fish occurred more frequently than mollusks, a phenomenon possibly related to turtles opportunistically feeding on bycaught fish from shrimp trawl nets (Shoop & Ruckdeschel, 1982; Youngkin, 2001). Such differences support the hypothesis that loggerhead diet composition shifts and adapts over time to changing prey availability. In turn, such fluctuations in the food web may be related to environmental shifts such as climate change, and also to human activities such as trawl fishing, which alters the food web composition by removing benthic crustaceans along with bycatch, and dredging, a practice that totally destroys benthic habitats (Bjorndal, 1997; Donaton et al., 2019; National Research Council, 2002). It has been previously theorized that loggerheads exploit trawl bycatch [fish] as food, and forage on fishes in times when their 'natural' diet of crustaceans decreases (Shoop & Ruckdeschel, 1982; Youngkin, 2001). This trend was not obvious in the data presented here, which may be due to the implementation of bycatch reduction devices in the study area in 1998 (Youngkin, 2001).

Seasonal effects were apparent in relative abundance of certain prey items. For example, crabs were not identified in stomach contents of turtles that stranded in cooler months (December–January), and bivalve mollusks were mostly identified in turtles that stranded in warmer months (May–August) (**Figure 2**). Low numbers of stranded turtles in the cooler months likely mask seasonal effects to some extent, as both total abundance of prey items and species richness correlated strongly to the number of turtles stranded per month (**Figure 2**). Such seasonal effects are likely related to local movement and abundance of specific prey types, which in turn are responsive to biological influences such as reproductive seasonality, as well as local external influences such as species-specific commercial and recreational fishery seasons including oyster (October–May), shrimp (June–January), and whelk (February–March) seasons (Georgia DNR, 2020). While it is possible that loggerheads take advantage of seasonal abundance of certain prey items, the opposite may also be true, wherein seasonal decreases in consumption of certain preferred prey items such as *Libinia* spp. crabs may lead to the turtles consuming other prey items instead, such as whelks (*Busycon* spp.) or surf clams (*Spisula solidissima*) (Youngkin, 2001).

Some dietary differences were observed to be related to turtle size, as adult turtles consumed proportionately more gastropods, and subadults ate proportionately more fish (**Figure 3**). This is consistent with previous observations for loggerheads in coastal Georgia; it has been suggested that smaller turtles engage in more opportunistic feeding of fish from trawl net bycatch than larger turtles (Shoop & Ruckdeschel, 1982; Youngkin, 2001). This type of feeding strategy can be potentially deadly for loggerheads due to the risk of trawl entanglement and drowning (Crouse, Crowder, & Caswell 1987; Youngkin, 2001). While these differences may be mostly behavioral, there may also be a physiological component, with larger turtles having more developed jaw musculature, and thereby more easily able to crush and consume the thick shells of gastropods and bivalves. In general, turtles in 'good' body condition had many more prey items in stomach contents than turtles in 'poor' body condition, including proportionately more gastropods. This is likely because poor body condition is often associated with cachexia as a symptom of debilitated loggerhead syndrome, or with co-morbid conditions such as boat strike wounds that are often associated with anorexia (Tomas et al., 2001). Thus, one limitation of analyzing prey items from dead stranded turtles is that some of these animals may not have been foraging under normal conditions (depending on cause of death), and therefore stomach contents may not accurately reflect the turtle's typical diet (Revelles et al., 2007). Additionally, hard parts such as shell fragments remain in the stomach for a longer period of time than soft or gelatinous parts, which could bias the data towards prey items with hard parts such as mollusks (Gales. 1988). A third limitation is that a one-time sampling of stomach contents does not encompass temporal variation in the diet (Horswill, et al., 2018). Future studies focused on analyzing stomach contents from deceased loggerheads incidentally captured during fisheries activities, as well as studies combining multiple analysis techniques (e.g., molecular DNA sequencing of feces, stable isotope ratios, fatty acid signatures) may provide a more complete representation of the diet of free-ranging, healthy loggerheads.

Here, we report the nutritional content of four prey items commonly consumed by loggerheads in the southeastern United States. These data can be used to enable caretakers to more closely approximate a 'normal' diet for captive loggerhead turtles, including providing vitamin and mineral supplementation when appropriate. Of course, understanding the current nutrient profile of diet items being fed is of critical importance as fish and other food items for aquatic species can vary dramatically with regards to micronutrient and macronutrient content.

As there are few published trials in which sea turtle nutrient requirements have been empirically determined, data published for other species has generally been a primary resource (Hoopes et al., 2017). The addition of more data on wild-type diet components and their composition will allow for further refinement of recommendations for animals under managed care. Macronutrient needs may vary between animals in the wild and those under managed care, due to the differences in energy requirements for thermoregulation, foraging, migration, and reproduction (generally higher in wild situations) (Bjorndal 1997). Relative proportions of protein, lipid, and carbohydrate to which animals have evolved in their wild-type diets may be very useful to predict appropriate ratios of these components in their diets under managed care. Additionally, knowledge of micronutrient composition of dietary items routinely consumed in the wild may offer specific insight into needs for animals under managed care.

4.2. Loggerhead blood analysis and comparisons

The blood analyte data provided here can be referenced to help evaluate the health status of free-ranging and captive loggerheads of various life-history stages. Overall, the foraging and nesting turtles were deemed to be healthy based on the results of physical examination and blood analysis (Deem et al., 2009; Stacy et al., 2018). To the authors' knowledge, this is the first study to present plasma concentrations of HDL, LDL, and VLDL, and Vitamins A, E, and D in free-ranging healthy and unhealthy loggerheads.

Lipoprotein fractions and triglycerides were higher in nesting females than for other classes of loggerheads, which corresponds with their need to mobilize lipids for egg yolk synthesis (Price, 2016). Triglycerides were dramatically lower in rehabilitating animals due to their fasting state, and slightly higher in younger animals which may be reflective of higher fish intake (generally higher in lipid content compared to crustaceans and gastropods) (Youngkin, 2001). There was no significant difference in plasma vitamin D in healthy versus rehabilitating turtles, whereas at the initiation of rehabilitative care, vitamin E levels were numerically lower and increased over the rehab period to similar (numerically) levels to healthy animals. More data are needed, but this information suggests that enhanced vitamin E supplementation during rehabilitation, particularly for animals with low circulating vitamin E, may be warranted.

Significant differences were observed in several health parameters between sick turtles entering rehabilitation and healthy turtles, with varying results when compared to foraging subadults, adults, and nesting turtles. These include relatively lower PCV, total solids, total protein, BUN, potassium, and higher sodium– all likely related to chronic conditions such as anemia, decreased food intake, dehydration, and poor nutrition (Deem et al., 2009; Stacy et al.

2018). Plasma protein electrophoresis analysis is a useful diagnostic tool in sea turtle medicine, indicating circulating concentrations of proteins involved in facilitating ion and hormone transport around the body, controlling osmotic pressure across membranes, and initiating immunological responses (Flint, Matthews, Limpus, & Mills, 2015; Gicking, Foley, Harr, Raskin, & Jacobson, 2004; Zaias & Cray 2002). Abnormalities in these proteins, particularly albumin, α-, β-, and γ-globulins, can be indicative of disease processes associated with acute and chronic inflammation (Flint et al., 2015). In sick loggerheads entering rehabilitation, concentrations of albumin, as well as α_1-, α_2-, β-, and γ-globulins were low compared to those of free-ranging, healthy loggerheads, likely indicating decreased immunity related to cachexia and prolonged illness (Stacy et al., 2018). Sick loggerheads also displayed relatively higher tWBC, heterophil, and lymphocyte counts, possibly due to antigenic stimulation from infection, and higher CO_2 which may be related to poor ventilation associated with respiratory distress such as caused by stranding or pneumonia (Hunt et al., 2016). Loggerhead sea turtles are capital breeders, mostly ceasing to feed during reproduction, and some differences in blood analytes observed for nesting loggerheads are indicative of reduced foraging, such as plasma concentrations of BUN and creatinine that were even lower than those of sick loggerheads in this study (Perrault & Stacy 2018; Price, Sotherland, Wallace, Spotila, & Dzialowski, 2019). These differences highlight the need to develop baseline blood parameter reference intervals that are specific to life history stage, that can be applied in a rehabilitation setting to help interpret clinical data for stranded loggerheads in various physiological states.

Disclosure statement

No potential conflict of interest was reported by the authors.

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Animal Welfare Statement

The authors confirm that the ethical guidelines of the journal, as noted on the journal's author guidelines page, have been adhered to and the appropriate ethical review committee approval has been received. The authors confirm that they have followed EU standards for the protection of animals used for scientific purposes.

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Table 1. Stomach contents from 153 deceased loggerheads as recorded in the Georgia Department of Natural Resources necropsy records from 1998–2008. Only identifiable items were evaluated. All items were classified according to the lowest identifiable taxonomic level.

Food item	Taxonomic	Common Name	Number of Reports (absolute)	%

Taxon	Level		Total N=288	Reports
Crustacea	Subphylum	Crustacean	1	0.4
Brachyura	Infraorder	Crab	52	18.2
Libinia spp.	Genus	Libinia crab	9	3.2
Callinectes sapidus	Species	Blue crab	10	3.5
Hepatus epheliticus	Species	Calico crab	17	5.9
Ovalipes ocellatus	Species	Lady crab	1	0.4
Menippe mercenaria	Species	Florida stone crab	1	0.4
Squilla mantis	Species	Mantis shrimp	3	1.1
Arenaeus cribarius	Species	Speckled swimming crab	1	0.4
Persephona punctata	Species	Purse crab	1	0.4
Paguroidea	Superfamily	Hermit crab	3	1.1
Majidae	Family	Spider crab	8	2.8
Penaeidae	Family	Shrimp	24	8.4
Limulus polyphemus	Species	Atlantic horseshoe crab	15	5.2
Osteichthyes	Superclass	Fish	45	15.7
Mollusca	Phylum	Mollusks	8	2.8
Gastropoda	Class	Gastropods	6	2.1
Uvanilla olivacea	Species	Blood-spotted star shell	3	1.1
Littorina littorea	Species	Common periwinkle	1	0.4
Urosalpinx cinerea	Species	Atlantic oyster drill	1	0.4
Busycon spp.	Genus	Whelk	19	6.3
Naticidae	Family	Moon snail	8	2.8
Neverita spp.	Genus	Moon snail	1	0.4
Sinum perspectivum	Species	White baby ear	1	0.4
Bivalvia	Class	Bivalve	3	1.1
Heterodonta	Subclass	Heterodonta clam	1	0.4
Spisula solidissima	Species	Surf clam	11	3.9

Crassostrea virginica	Species	Eastern oyster	2	0.7
Dinocardium robustum	Species	Atlantic giant cockle	1	0.4
Mercenaria mercenaria	Species	Hard clam	2	0.7
Donax variabilis	Species	Coquina clam	3	1.1
Pennatulacea	Order	Pen snail	1	0.4
Gorgoniidae	Family	Gorgoniidae soft coral	1	0.4
Anemone spp.	Genus	Anemone	5	1.8
Exaiptasia pallida	Species	Brown anemone	2	0.7
Echinoidea	Class	Sea urchin	1	0.4
Holothuroidea	Class	Sea cucumber	1	0.4
Alcyonidium hauffi	Species	Rubbery bryozoan	8	2.8
Tunicata	Subphylum	Tunicate	3	1.1
Porifera	Phylum	Sponge	1	0.4
Siboglinidae	Family	Tube worm	1	0.4
Sargassum spp.	Genus	Sargassum seaweed	1	0.4

Table 2. Results of N–1 chi-squared tests used to compare proportions of broader taxonomic categories of prey items identified in the stomach contents of 22 loggerheads that stranded in cooler months and 131 that stranded in warmer months, between 19 stranded adult loggerheads and 123 stranded subadults, and between 83 loggerheads that stranded in 'good' body condition, and 11 that stranded in 'poor' body condition.
df = degrees of freedom; * denotes statistically significant differences

	Stranded in Cooler Months	Stranded in Warmer Months	% Difference	95% CI	χ^2	df	*P*-value
Gastropods	5%	17%	12%	–6.3–20.4	2.1	1	0.2
Fish	18%	31%	13%	–8.6–26.7	1.5	1	0.2
Crabs	91%	97%	6%	–1.9–24.8	1.8	1	0.2
Bivalve mollusks	27%	27%	0%	–22.0–16.2	0.0	1	1.0
Invertebrates, mixed	9%	15%	6%	–13.4–15.6	0.6	1	0.5
	Adults	**Subadults**	**% Difference**	**95% CI**	χ^2	**df**	***P*-value**
Gastropods	58%	22%	36%	12.9–56.0	10.8	1	0.001*
Fish	5%	33%	28%	7.3–37.7	6.2	1	0.01*
Crabs	100%	87%	13%	–4.5–20.1	2.8	1	0.1
Bivalve mollusks	11%	14%	3%	–18.6–13.7	0.1	1	0.7
Invertebrates, mixed	11%	14%	3%	–18.6–13.7	0.1	1	0.7
	Good Body Condition	**Poor Body Condition**	**% Difference**	**95% CI**	χ^2	**df**	***P*-value**
Gastropods	30%	0%	30%	2.7–40.6	4.4	1	0.04*
Fish	29%	27%	2%	–28.6–22.4	0.02	1	0.9
Crabs	96%	82%	14%	–0.5–43.6	3.5	1	0.06
Bivalve mollusks	19%	0%	19%	–7.8–28.7	2.5	1	0.1
Invertebrates, mixed	14%	0%	5%	–24.2–16.7	0.2	1	0.7

Table 3. Nutritional analysis of common prey items of loggerhead turtles in the southeastern United States. When available, nutritional contents are presented on an as-fed (AF) and dry matter (DM) basis.

	Horseshoe Crab		**Whelk**		**Cannonball Jellyfish**		**Blue Crab**	
Parameter	**AF**	**DM**	**AF**	**DM**	**AF**	**DM**	**AF**	**DM**
Gross energy (kcal/g)	--	--	436	578	--	--	725	2,328
Moisture (%)	70.9	--	24.7	--	94.7	--	68.9	--
Dry Matter (%)	--	29.1	--	75.3	--	5.3	--	31.1
Crude Protein (%)	21.5	73.7	8.6	11.4	1.6	30.8	11.9	38.1
Crude Fat (%)	0.5	1.9	1.0	1.3	0.1	2.5	0.8	2.6
Ash (%)	5.08	17.46	63.62	84.48	3.17	59.82	16.05	51.55
Calcium (%)	0.47	1.35	24.22	32.16	0.04	0.67	5.0	16.05
Phosphorus (%)	0.13	0.38	0.06	0.08	0.01	0.26	0.56	1.81
Magnesium (%)	0.18	0.52	0.08	0.11	0.08	1.50	0.27	0.86
Potassium (%)	0.2	0.59	0.11	0.14	0.06	1.10	0.2	0.64
Sodium (%)	1.07	3.11	0.53	0.71	0.93	17.61	0.68	2.19
Sulfur (%)	0.28	0.81	0.19	0.25	0.08	1.46	0.17	0.54
Cobalt (mg/kg)	0.23	0.67	0.1	0.13	0.02	0.39	0.06	0.18
Copper (mg/kg)	67.2	194.7	3.0	4.0	<1.0	4.0	12.0	39.0
Iron (mg/kg)	337.4	977.2	578.0	767.0	10.0	185.0	28.0	91.0
Manganese (mg/kg)	13.43	38.9	13.0	17.0	<1.0	7.0	55.0	176.0
Molybdenum (mg/kg)	0.41	1.19	0.3	0.4	<0.1	0.8	0.3	1.0
Selenium (mg/kg)	1.36	3.94	0.33	0.44	0.15	2.9	0.56	1.8
Zinc (mg/kg)	61.9	179.2	47.0	63.0	2.0	42.0	20.0	64.0
Vitamin A (IU/g)	<0.0015	<0.0045	--	--	--	--	--	--
Vitamin E (mg/kg)	1.22	3.53	--	--	--	--	--	--

Table 4. Selected morphometric parameters for free-ranging subadult and adult loggerheads, nesting female loggerheads, and loggerheads at entry and end of rehabilitation. Values represent average ± standard deviation (SD).

CCL_{min} = minimum curved carapace length measured from notch to notch; CCL_{max} = maximum curved carapace length from notch to tip; SCL_{max} = maximum straight carapace length measured from notch to tip; CCW = curved carapace width.

Cohort	N	Body Mass (kg)	CCL_{min} (cm)	SCL_{max} (cm)	CCL_{max} (cm)	CCW (cm)
Free-ranging subadults	15	65.2 ± 15.4	81.0 ± 6.5	76.8 ± 7.0	82.6 ± 7.0	75.6 ± 6.0
Free-ranging adults	24	91.2 ± 16.6	91.0 ± 4.5	86.5 ± 4.2	92.5 ± 4.6	83.3 ± 3.9
Rehabilitation entry	15	43.9 ± 18.4	74.2 ± 10.3	69.0 ± 10.3	75.4 ± 10.6	71.3 ± 9.9
Rehabilitation end	15	49.6 ± 8.0	73.8 ± 8.0	68.4 ± 8.3	75.2 ± 8.0	70.8 ± 7.2

Table 5. Estimated consumption of select nutrients from two diet scenarios, the addition of a vitamin/mineral tablet supplement, or the use of a critical care diet formulation. All nutrients ate presented on a dry matter basis.

	Diet A	Diet B	Recommended Intake Level	Carnivorous Turtle Tablet Contribution	Carnivorous Turtle Critical Care Diet Contribution
Crude Protein (%)	49	73	40–47	NA	48
Crude Fat (%)	7	52	9	NA	22
Ca (%)	13	2	1–4	NA	3.6
P (%)	1	2	0.67–0.89	NA	2.2
Fe (mg/kg)	420	62	50–100 min	112	640
Mn (mg/kg)	34	3	20–50	96	120
Zn (mg/kg)	87	79	14–83	39	250
Cu (mg/kg)	37	29	1–6	2	20
Vitamin A (IU/kg)	44,000	92,000	2,700–20,000	7,440	5,380
Vitamin E (IU/kg)	179	420	30–198	384	103
Vitamin C (mg/kg)	9	0	100–1100	560	538
Thiamine (mg/kg)	0	0	1–12	384	90

Diet A consisted of 30% crab, 40% fish (50:50 capelin:lean herring), 30% gastropods and mollusks

Diet B consisted of 80% fish (50:50 capelin:lean herring), 20% squid

Diet recommendations from Hoopes et al. (2017)[18]

Tablet consumption based on 1.5 g tablet per 0.5 kg fish as fed.

NA–not applicable.

625

Figure Legend

Figure 1. (**A**) Number of turtles, and relative abundance and richness of prey items (by year), and (**B**) relative abundance of prey item categories (by year) identified in stomach contents of 153 loggerhead sea turtles (*Caretta caretta*) that stranded in coastal Georgia, USA during 1998–2008.

Figure 2. (**A**) Number of turtles, and relative abundance and richness of prey items (by month), and (**B**) relative abundance of prey item categories (by month) identified in stomach contents of 153 loggerhead sea turtles (*Caretta caretta*) that stranded in coastal Georgia, USA during 1998–2008.

Figure 3. (**A**) Number of turtles, and relative abundance and richness of prey items identified in stomach contents based on age class in 123 subadult and 19 adult loggerhead sea turtles that stranded in coastal Georgia, USA during 1998–2008. (**B**) Percent occurrence of different categories prey items identified in stranded loggerhead stomach contents, based on age class. *denotes statistically significant differences in proportions.

Table 6. Hematology, plasma biochemistry values including lipoproteins and immunoglobulins, and plasma mineral and vitamin concentrations are presented for free-ranging subadult loggerheads, free-ranging adults loggerheads, nesting female loggerheads, and loggerheads in rehabilitation.

Superscript numbers denote statistically significant differences ($P<0.05$) between variables.

PCV=packed cell volume; ALT=alanine transaminase; AST=aspartate aminotransferase; BUN=blood urea nitrogen; B/C=BUN:creatinine; CPK=creatine phosphokinase; GGT=gamma-glutamyl transferase; HDL=high-density lipoprotein; LDH=lactate dehydrogenase; LDL=low-density lipoprotein; VLDL=very low-density lipoprotein; A/G=albumin:globulin.

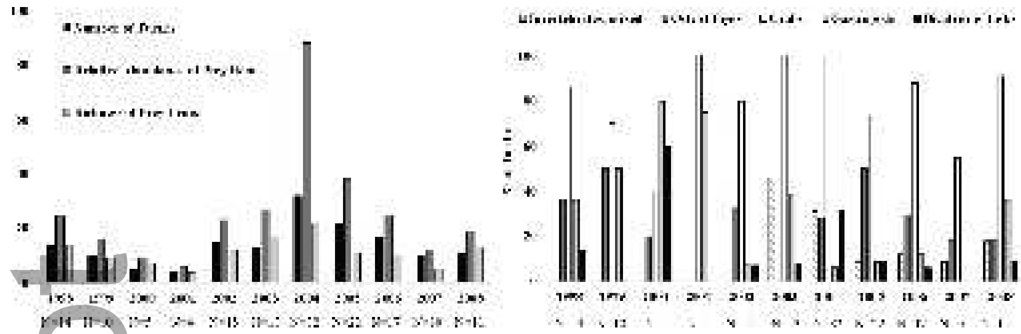
	Trawl-captured subadults[1] (N=15)			**Trawl-captured adults[2] (N=24)**			**Nesting females[3] (N=15)**			**Rehabilitating loggerheads at admission[4] (N=15)**			**Rehabilitating loggerheads prior to release[5] (N=15)**		
	Median ± SD	90% CI	Range	Median ± SD	90% CI	Range	Median ± SD	90% CI	Range	Median ± SD	90% CI	Range	Median ± SD	90% CI	Range
Hematology															
PCV (1.0 proportion)	0.35 ± 0.04^{4}	0.304–0.386	0.240–0.400	0.350 ± 0.043^{4}	0.280–0.394	0.280–0.430	0.340 ± 0.050^{4}	0.282–0.398	0.270–0.410	0.180 ± 0.095^{1,2,3}	0.124–0.326	0.050–0.400	0.280 ± 0.035	0.244–0.326	0.230–0.360
Total solids (g/L)	50 ± 9.0^{4}	44–57	26–70	48 ± 9.0^{4}	33–56	28–70	48 ± 70^{4}	46–59	44–63	33 ± 9.0^{1,2,3}	17–99	16–42	43 ± 7.0	34–50	30–53
tWBC ($\times10^9$/L)	0.0014 ± 0.0005^{4}	0.0008–0.0021	0.0007–0.0025	0.0014 ± 0.0012^{4}	0.0008–0.0031	0.0006–0.005	0.0028 ± 0.0014	0.0013–0.0043	0.0012–0.0051	0.0034 ± 0.0049^{1,2}	0.0014–0.0104	0.0003–0.0192	0.002 ± 0.0022	0.0007–0.0061	0.0003–0.007
Absolute Heterophils ($\times10^3$)	0.5 ± 0.3^{4}	0.3–0.9	0.3–1.3	0.6 ± 0.6^{4}	0.2–1.5	0.2–2.7	0.9 ± 0.7	0.4–1.7	0.4–2.2	1.7 ± 3.7^{1,2}	0.3–7.2	0.1–13.4	0.8 ± 1.0	0.3–0.7	0.1–3.2
Absolute Lymphocytes ($\times10^3$)	0.5 ± 0.2^{4}	0.4–0.9	0.1–0.9	0.6 ± 0.4^{4}	0.4–1.2	0.2–1.7	1.1 ± 0.6^{4}	0.8–2.1	0.8–2.5	1.3 ± 1.3^{1,2,3}	0.5–3.3	0.1–4.8	0.9 ± 0.9	0.3–2.0	0.1–3.5
Absolute Eosinophils	0.1 ± 0.2	0.0–0.4	0.0–0.5	0.2 ± 0.2^{3}	0.0–0.6	0.0–0.8	0.1 ± 0.2^{2}	0.0–0.4	0.0–0.5	0.1 ± 0.2	0.0–0.2	0.0–8	0.1 ± 0.3	0.0–0.6	0.0–1.2

$(\times 10^3)$															
Absolute Monocytes $(\times 10^3)$	0.0 ± 0.1	0.0–0.1	0.0–0.2	0.0 ± 0.1	0.0–0.2	0.0–0.6	0.1 ± 0.1	0.0–0.3	0.0–0.5	0.1 ± 0.1	0.0–0.3	0.0–0.5	00 ± 0.4	0.0–0.2	0.0–0.0
Absolute Basophils $(\times 10^3)$	0.0 ± 0.1	0.0–0.01	0.0–0.6	0.0 ± 0.0	0.0–0.0	0.0–0.0	0.0 ± 0.0	0.0–0.0	0.0–0.0	0.0 ± 0.0	0.0–0.0	0.0–0.0	0.0 ± 0.1	0.0–1.4	0.0–0.4
Plasma Biochemistry															
ALT (µKat/L)	0.23 ± 0.24[3]	0.14–0.40	0.13–1.1	0.22 ± 0.09[3]	0.13–0.32	0.07–0.53	0.10 ± 0.05[1,2,4]	0.05–0.15	0.05–0.18	0.30 ± 0.17[3]	0.13–0.54	0.10–0.65	0.33 ± 0.73	0.13–1.66	0.10–2.61
Amylase (µKat/L)	6.03 ± 1.90[3]	4.04-8.37	2.35–9.72	6.26 ± 1.39[3]	4.50–7.72	3.12–8.47	8.40 ± 1.57[1,2,4]	6.10–10.1	5.68–10.9	4.71 ± 2.55[3]	2.05–8.06	0.50–8.55	8.20 ± 2.94	3.66–102	1.47–11.9
AST (µKat/L)	4.04 ± 1.25	2.90–4.90	2.64–7.85	3.29 ± 0.990	7.02–15.3	6.35–16.5	2.84 ± 1.84	2.26–4.55	2.17–9.52	3.77 ± 2.61	2.24–7.46	1.67–11.6	4.07 ± 3.72	2.49–11.5	2.15–13.3
BUN (mmol/L)	34.6 ± 12.9	19.3–41.8	14.6–70.0	31.4 ± 11.3[3]	11.3–41.9	5.35–46.8	3.90 ± 0.0	2.14–6.50	1.78–8.92	21.1 ± 11.5[1,3]	9.35–35.2	6.78–47.5	46.1 ± 14.9	29.9–66.8	22.1–79.3
B/C ratio	267.5 ± 115.0[3]	89.5–365.3	65.0–386.7	262.5 ± 125.5[3]	164.8–448.0	103.3–655.0	60 ± 49.3[1,2,4]	32.0–142.0	20.0–190.0	213.3 ± 142.3[3]	58.0–408.0	22.0–460.0	322.5 ± 269.7	192.8–841.0	124.0–955.0
Calcium (mmol/L)	1.95 ± 2.62	1.57–2.30	0.5–2.33	2.0 ± 0.25[4]	1.65–2.27	1.18–2.3	2.63 ± 0.95[4]	1.38–3.63	0.78–41	1.55 ± 0.28[1,2,3]	1.13–1.82	1.0–1.82	1.82 ± 0.23	1.45–2.08	1.45–2.13
Cholesterol (mmol/L)	2.56 ± 0.70[3]	1.55–3.11	1.17–3.94	2.15 ± 0.77[3]	1.59–3.21	1.29–4.84	5.46 ± 1.20[1,2,4]	4.23–7.36	3.88–7.64	1.29 ± 1.22[3]	1.17–3.56	1.17–5.23	3.42 ± 1.13	2.06–4.53	1.17–5.54
CO_2 (mmol/L)	25.0 ± 5.1[4]	20.0–28.6	11.0–33.0	28.0 ± 4.7[3,4]	21.0–33.4	18.0–35.0	23.0 ± 6.0[2,4]	14.4–27.6	6.0–28.0	34.0 ± 6.0[1,2,3]	32.0–44.4	27.0–50.0	31.0 ± 3.9	28.4–36.2	26.0–41.0
CPK (µKat/L)	14.3 ± 3.88	11.5–19.3	9.97–25.1	12.5 ± 16.8	7.47–23.3	3.84–96.0	8.33 ± 41.0	5.01–76.4	4.54–145	18.2 ± 51.6	6.47–101	2.91–189	8.70 ± 25.3	4.46–26.3	2.86–104

Creatinine (µmol/L)	35.4 ± 26.5[2,3]	26.5–61.9	26.5–106	26.5 ± 8.84[1,3]	17.7–35.4	8.84–44.2	8.84 ± 8.84[4]	8.84–35.4	8.84–44.2	26.5 ± 44.2[3]	17.7–53.0	17.7–177	26.5 ± 17.7	17.7–44.2	17.7–61.9
GGT (µKat/L)	0.08 ± 0.01	0.08–0.10	0.08–	0.08 ± 0.0	0.08–0.08	0.08–0.08	0.11 ± 0.09	0.08–0.12	0.08–0.42	0.08 ± 0.0	0.08–0.08	0.08–0.08	0.08 ± 0.0	0.08–0.08	0.08–0.08
Glucose (mmol/L)	6.49 ± 1.19	5.23–7.76	4.00–8.55	5.72 ± 1.28	4.70–7.80	4.05–9.32	5.38 ± 0.81	4.10–6.06	3.77–6.77	6.99 ± 3.49	3.20–11.3	0.56–13.1	6.05 ± 1.38	4.21–7.39	3.66–9.05
HDL (mmol/L)	23.0 ± 9.4[3]	14.8–37.6	12.0–42.0	19.0 ± 10.3[3]	13.6–31.0	11.0–56.0	55.0 ± 17.6[1,2,4]	31.0–72.6	28.0–112.0	13.0 ± 14.7[3]	4.8–40.4	4.0–45.0	37.0 ± 19.0	17.4–56.0	5.0–74.0
LDH (µKat/L)	11.7 ± 7.37	6.91–23.7	6.20–30.2	9.32 ± 3.10	7.02–15.3	6.35–16.5	14.4 ± 8.79	6.12–29.5	3.77–31.3	6.00 ± 11.1	3.31–27.3	1.67–37.6	7.10 ± 3.59	1.67–10.8	1.67–10.8
LDL (mmol/L)	18.51 ± 12.0	8.85–37.0	0.82–45.7	16.5 ± 8.85[3]	9.71–30.0	0.82–37.9	43.2 ± 22.4[2]	8.15–61.6	0.41–80.2	22.6 ± 10.3	16.3–35.0	15.2–38.7	33.3 ± 18.0	14.2–56.6	11.9–66.7
Lipase (µKat/L)	0.12 ± 0.03[3]	0.11–0.15	0.02–0.78	0.10 ± 0.09[3]	0.03–0.21	0.02–0.43	0.33 ± 0.16[1,2,4]	0.24–0.62	0.15–0.67	0.05 ± 0.26[3]	0.02–0.23	0.02–1.05	0.08 ± 0.07	0.02–0.17	0.02–0.22
Phosphorus (mmol/L)	2.81 ± 0.29[2,3,4]	2.29–2.94	2.16–3.17	2.36 ± 0.45[1,3,4]	1.97–2.91	1.58–3.68	2.78 ± 0.81[1,2,4]	2.23–4.23	2.00–4.72	2.42 ± 0.68[1,2,3]	1.61–3.29	1.32–3.52	2.00 ± 0.45	1.61–2.29	0.52–2.52
Potassium (mmol/l)	4.8 ± 0.4[3,4]	4.4–5.4	4.3–5.6	4.5 ± 0.4[3,4]	4.2–5.0	3.8–5.3	3.8 ± 0.5[1,2]	3.4–4.7	3.3–4.7	3.6 ± 0.5[1,2]	2.6–3.9	2.6–3.9	3.8 ± 0.6	3.3–5.0	3.3–5.2
Sodium (mmol/l)	159.0 ± 7.9[3]	155.2–173.6	150.0–177.0	161.0 ± 5.0[3]	154.6–166.4	152.0–170.0	148.0 ± 4.7[1,2,4]	1440–153.6	143.0–161.0	158.0 ± 9.1[3]	149.0–169.8	147.0–178.0	151.0 ± 13.1	145.0–172.2	132.0–186.0
Triglycerides (mmol/L)	1.53 ± 1.36[3,4]	0.58–3.95	0.52–4.37	1.15 ± 0.83[3,4]	0.46–2.58	0.31–3.29	3.49 ± 1.42[1,2,4]	3.08–6.19	1.57–6.35	0.11 ± 0.18[1,2,3]	0.11–0.31	0.11–0.79	0.42 ± 0.35	0.17–0.71	0.11–1.59
g/Uric acid (mmol/L)	0.05 ± 0.02[3]	0.03 – 0.08	0.03–0.09	0.04 ± 0.02	0.04–0.07	0.02–0.09	0.04 ± 0.01[1,4]	0.02–0.05	0.02–0.08	0.06 ± 0.02[3]	0.04–0.09	0.03–0.12	0.03 ± 0.02	0.02–0.05	0.02–0.11
VLDL (kU/l)	27.0 ± 24.2[3,4]	10.4–71.4	9.0–77.0	20.0 ± 17.7[3,4]	8.2–45.4	5.0–58.0	62.0 ± 25.1[1,2,4]	54.4–109.4	28.0–80.0	5.0 ± 4.0[1,2,3]	3.5–10.0	3.0–14.0	8.0 ± 6.0	6.0–13.2	5.0–28.0

Plasma protein electrophoresis															
Total protein (g/dl)	50 ± 8.0[4]	44–56	31–69	48 ± 10[4]	35–59	26–66	47 ± 10[4]	41–66	40–70	30 ± .8[1,2,3]	19–37	14–41	38 ± 0.0	26–48	20–60
A/G ratio	0.2 ± 0.1[3]	0.2–0.3	0.2–0.4	0.2 ± 0.1	0.2–0.4	0.2–0.5	0.3 ± 0.1[1]	0.2–0.4	0.2–0.4	0.2 ± 0.1	0.2–0.4	0.1–0.4	0.3 ± 0.1	0.2–0.4	0.1–0.5
Pre-albumin (mg/L)	1000 ± 1000	0.0–2000	0.0–2000	1000 ± 0.0[3]	1000–2000	1000–2000	2000 ± 1000[1,4]	1000–3000	1000–4000	1000 ± 0.0[3]	0.0–1000	0.0–1000	1000 ± 1000	0.0–2000	0.0–3000
Albumin (g/L)	9.0 ± 2.0[4]	8.0–11	5.0–13	8.0 ± 2.0[4]	6.0–10	4.0–14	8.0 ± 3.0[4]	7.0–11	2.0–13	5.0 ± 2.0[1,2,3]	2.0–8.0	2.0–8.0	8.0 ± 3.0	3.0–11	2.0–13
Alpha-1 globulins (g/L)	2.0 ± 1.0[4]	1.0–3.0	1.0–3.0	2.0 ± 1.0[4]	1.0–3.0	1.0–3.0	2.0 ± 0.0	1.0–2.0	1.0–2.0	1.0 ± 1.0[1,2]	1.0–2.0	0.0–2.0	2.0 ± 2.0	1.0–3.0	1.0–7.0
Alpha-2 globulins (g/L)	3.0 ± 1.0[3,4]	2.0–4.0	2.0–4.0	3.0 ± 1.0[3,4]	2.0–4.0	2.0–5.0	4.0 ± 1.0[1,2,4]	3.0–6.0	3.0–6.0	2.0 ± 1.0[1,2,3]	1.0–2.0	1.0–3.0	2.0 ± 1.0	2.0–3.0	1.0–4.0
Beta globulins (g/L)	11 ± 3.0[3,4]	9.0–15	8.0–19	13 ± 3.0[3,4]	8.0–16	6.0–17	16 ± 4.0[1,2,4]	13–23	12–27	8.0 ± 3.0[1,2,3]	5.0–11	4.0–12	10 ± 4.0	6.0–14	3.0–17
Gamma globulins (g/L)	23 ± 5.0[3,4]	20–28	15–36	20 ± 6.0[3,4]	13–30	11–33	14 ± 4.0[1,2]	12–20	10–22	12 ± 4.0[1,2]	8.0–18	7.0–22	13 ± 5.0	9.0–18	8.0–27
Vitamins and minerals															
Copper (μmol/L)	0.09 ± 0.03	0.06–0.13	0.06–0.9	0.08 ± 0.02	0.05–0.09	0.05–0.11	0.08 ± 0.02	0.06–0.11	0.06–0.11	0.09 ± 0.03	0.05–0.11	0.05–0.13	0.08 ± 0.02	0.05–0.11	0.05–0.11
Iron (μmol/L)	0.29 ± 1.36[2,4]	0.25–2.17	0.20–4.74	0.23 ± 0.09[1,3]	0.14–0.30	0.11–0.57	0.43 ± 0.91[2,4]	0.29–1.25	0.27–3.71	0.23 ± 0.07[1,3]	0.13–0.27	0.11–0.41	0.29 ± 4.73	0.21–0.61	0.18–18.6

Magnesium (mmol/L)	31.1 ± 3.41[3,4]	26.5–34.1	22.0–34.5	29.8 ± 4.24[3,4]	25.2–35.7	20.2–38.6	26.3 ± 4.24[1,2,4]	20.8–28.2	20.0–37.2	20.2 ± 4.53[1,2,3]	14.3 – 26.0	13.2–26.5	15.1 ± 2.92	11.4–18.1	9.26–20.8
Selenium (µmol/L)	0.01 ± 0.0	0.0–0.1	0.0–0.02	0.0 ± 0.0	0.0–0.1	0.0–0.1	0.0 ± 0.0	0.0–0.0	0.0–0.0	0.0 ± 0.0	0.0–0.1	0.0–0.1	0.1 ± 0.0	0.0–0.1	0.0–0.1
Zinc (µmol/L)	0.14 ± 0.06[3]	0.11–0.26	0.08–0.29	0.12 ± 0.06[3]	0.08–0.17	0.05–0.20	0.26 ± 0.09[1,2,4]	0.18–0.38	0.14–0.44	0.14 ± 0.05[3]	0.11–0.20	0.11–0.23	0.15 ± 0.40	0.09–0.23	0.08–1.70
Vitamin A (nmol/L)	1.25 ± 0.75	0.75–2.50	0.50–3.00	1.75 ± 0.75	1.50–2.75	0.75–3.24	1.50 ± 0.75	1.00–3.00	0.75–3.24	1.50 ± 0.75	0.75–2.50	0.50–2.75	1.50 ± 0.75	1.25–2.75	1.00–3.00
Vitamin E (nmol/L)	9.98 ± 7.99[3]	5.49–22.5	3.24–32.5	10.2 ± 7.74[3]	5.74–21.2	4.24–42.4	25.0 ± 7.6[1,2,4]	12.2–56.7	8.24–74.6	7.74 ± 9.98[3]	3.74–13.7	2.5–43.9	11.0 ± 5.24	4.99–19.0	4.24–19.2
Vitamin D (nmol/L)	15.0 ± 12.2	7.49–37.9	4.99–42.4	12.5 ± 11.7	8.99–30.0	4.99–62.4	27.5 ± 28.2	12.5–75.9	0–87.4	15.0 ± 11.0	12.5–30.0	4.99–49.9	17.5 ± 16.0	12.5–48.9	9.98–59.9



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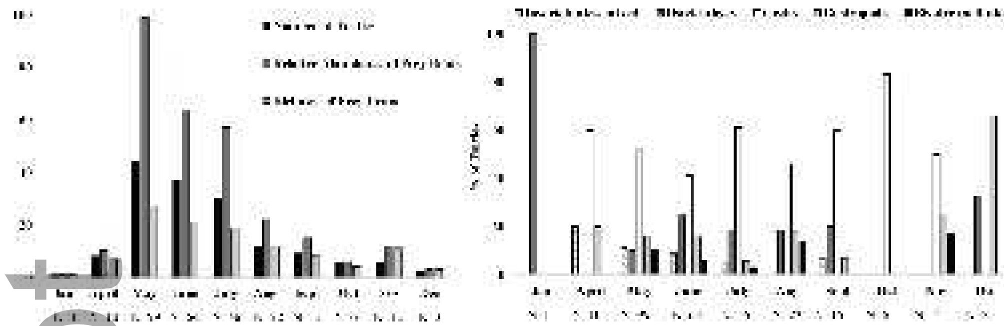

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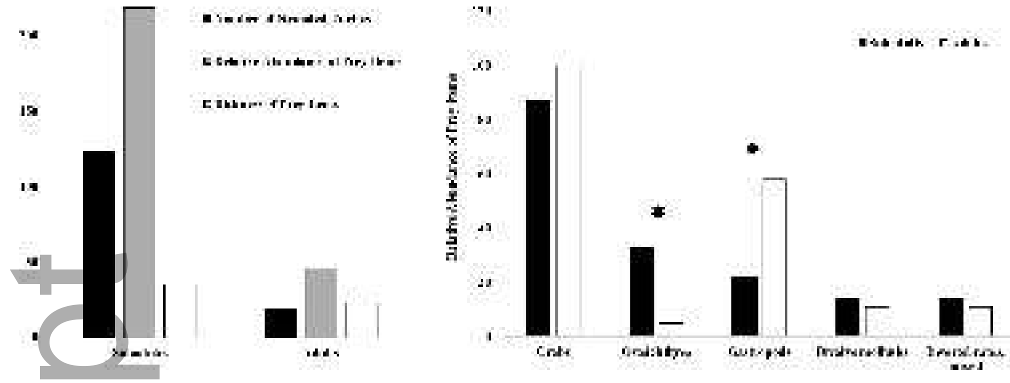

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