Correlates of serum IGF-1 in young children with moderate acute malnutrition: a cross-sectional study in Burkina Faso

Thora W. Kjaer[5]; Benedikte Grenov[1]; Charles W. Yaméogo[1,2]; Christian Fabiansen[1,8]; Ann-Sophie Iuel-Brockdorff[1]; Bernardette Cichon[1]; Nick S. Nielsen[1]; Suzanne Filteau[3]; André Briend[1,7]; Jonathan C. K. Wells[4]; Kim F. Michaelsen[1]; Henrik Friis[1]; Daniel Faurholt-Jepsen[1]; Vibeke B. Christensen[5,6]

[1] Department of Nutrition, Exercise and Sports, University of Copenhagen, Denmark

[2] Département Biomédical et Santé Publique, Institut de Recherche en Sciences de la Santé, Ouagadougou, Burkina Faso

[3] Faculty of Epidemiology and Population Health, London School of Hygiene and Tropical Medicine, UK

[4] UCL Great Ormond Street Institute of Child Health, University College London, UK

[5] Department of Pediatrics and Adolescent Health, Rigshospitalet, Denmark

[6] Médecins Sans Frontières – Denmark, Copenhagen, Denmark

[7] Department for International Health, University of Tampere, School of Medicine, Tampere, Finland

[8] The Pediatric Department, Holbæk Sygehus, DK-4300 Holbæk, Denmark

Corresponding author:

Thora Wesenberg Kjaer, Blegdamsvej 9, DK-2100 Copenhagen, Denmark, phone +4535452360, Thora.wesenberg.kjaer@regionh.dk

Funding:

The study was funded by Danish International Development Agency (09-097 LIFE) Médecins Sans Frontières (Denmark, Norway); Arvid Nilsson's Foundation; The World Food Program, which was part of a donation to the World Food Program from the American people through the support of the US Agency for International Development's Office of Food for Peace; the Alliance for International Medical Action; and the European Union's humanitarian aid funds, in partnership with Action Contre la Faim. The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

Short running title:

Correlates of IGF-1 in children with MAM

Abbreviations:

AGP: α1-acid glycoprotein
CRP: C-reactive protein
DXA: Dual-energy X-ray absorptiometry
FFM: Fat free mass
FFMI: Fat free mass index
FM: Fat mass
FMI: Fat mass index
GH: Growth hormone
HRP2: Histidine rich protein 2
IGF-1: Insulin-like growth factor 1
IMCI: Integrated Management of Childhood Illnesses
LAZ: Length-for-age Z-score
MAM: Moderate acute malnutrition
MUAC: Mid-upper arm circumference
NEXS: Department of Nutrition, Exercise and Sports
RDT: Rapid diagnostic test
RPM: Revolutions per minute
SAM: Severe acute malnutrition

TBW: Total body water

VIF: Variance inflation factor

WAZ: Weight-for-age Z-score

WLZ: Weight-for-length Z-score

Clinical trial registration number:

ISRCTN42569496, https://www.isrctn.com/

Word count: 3390

Data described in the manuscript, code book, and analytic code will not be made available because the Danish Act on Data Protection does not allow for personal data to be made available to others without prior individual approval from the Danish Data Protection Agency.

Abstract

Background: Serum IGF-1 (sIGF-1) is an important growth factor in childhood. However, studies on sIGF-1 among children from low-income countries are few and the role of body composition are unknown.

Objective: To assess the associations of anthropometry, body composition, inflammation and breastfeeding with sIGF-1 among children with moderate acute malnutrition (MAM).

Design: A cross-sectional study based on admission data from 6-23 months old children with MAM participating in a nutrition intervention trial (Treatfood, ISRCTN42569496) in Burkina Faso. Linear regression analysis was used to identify correlates of sIGF-1.

Results: Among 1546 children, the median [interquartile range] sIGF-1 was 12 [8.2-18.3] ng/mL. Serum IGF-1 was highest at 6 months with a nadir around 10-11 months, and higher in girls than boys. Length-for-age Z-score (LAZ), weight-for-length Z-score (WLZ) and mid-upper arm circumference were positively associated with sIGF-1 ($p \leq 0.001$). Fat-free mass (FFM) was also positively associated, as sIGF-1 increased 1.5 (95%CI 0.5,2.5) ng/ml for each 1 kg increase in FFM. However, the association disappeared after adjustment for height. Elevated serum C-reactive protein (CRP) and α1-acid glycoprotein (AGP) were negatively associated with sIGF-1 ($p \leq 0.001$), as was fever ($p < 0.001$), but not a positive malaria test per se ($p = 0.15$). Children never breastfed had lower sIGF-1 (-5.1, 95%CI -9.8,-0.3).

Conclusion: LAZ and WLZ were positively and inflammation negatively associated with sIGF-1. As all children were moderately malnourished and many had inflammation, this probably explains the very low median sIGF-1. The association of FFM with sIGF-1 was fully explained by height. There was a marked age pattern, with a nadir in late infancy,

confirming findings from smaller studies from well-nourished populations. There is a need for prospective studies to disentangle the role of sIGF-1 in growth and health.

Key words: moderate acute malnutrition, insulin-like growth factor (IGF-1), low-income country, body composition, inflammation

Introduction

Moderate acute malnutrition (MAM) affects approximately 33 million children under five years of age worldwide and is associated with increased morbidity and mortality from infectious diseases [1]. Serum IGF-1 (sIGF-1) is an important growth factor both prenatally and during childhood [2, 3]. Positive associations between sIGF-1 concentrations and height and weight have been reported in children from high-income countries [4-6] and malnourished children seem to have lower sIGF-1 than age-matched controls [7]. Studies in well-nourished children have also found positive associations of fat-free mass (FFM), and to a lesser extent fat mass (FM), with sIGF-1 [5, 6]. Similarly, changes in sIGF-1 were shown to be associated with changes in FFM, but not FM in 6-9 year-old Ghanaian children with stunting and wasting prevalences of 12% and 10%, respectively [8]. In general, sIGF-1 concentrations vary considerably in children, depending on their age, sex, ethnicity, socio-economic status and nutrition [9-12].

Inflammation is another important factor influencing sIGF-1 concentrations. Children with chronic inflammatory conditions often have growth failure and the underlying pathology seems to be multifactorial. Both an effect of the inflammatory response on the growth hormone (GH)-IGF-1 axis and an effect of suboptimal nutrition have been suggested to play important roles [13]. In low-income settings, environmental enteric dysfunction, characterized by malabsorption and intestinal inflammation and permeability, is associated with stunting [14]. A Brazilian case-control sub-study found relations between systemic inflammation, the GH-IGF-1 axis and growth in 6-24 month old children with varying degrees of malnutrition. Children with recent infections had lower sIGF-1, and serum C-reactive protein (CRP) was negatively associated with sIGF-1 [15]. A birth cohort study of Zimbabwean infants showed that acute infection decreased sIGF-1 both through acute phase

response and by direct downregulation of IGF-1 [16]. This suggests a complex relationship between malnutrition, inflammation and IGF-1.

The aim of this study was to investigate sex and age patterns of sIGF-1 concentrations from 1546 young children with MAM from Burkina Faso and to assess the association of anthropometry, body composition, inflammation and breastfeeding with sIGF-1 in this large population.

Subjects and Methods

Study design, area and population

This cross-sectional study was based on admission data from a cohort of children with MAM enrolled in a nutrition intervention trial (Treatfood, ISRCTN42569496). The study was carried out in the Passoré Province, Northern Region, Burkina Faso. The region is located in the Sudano-Sahaelian zone, with an average yearly rainfall of 600-700 mm. The catchment area covered 143 villages and a total population of approximately 258,000.

Children aged 6-23 months with MAM, resident in the catchment area, and whose parents/guardians consented for their child to participate, were eligible. Children were excluded if they had been treated for severe acute malnutrition (SAM) or hospitalized within the past two months, had participated in a nutritional program, required hospitalization, or had severe disability. Screening was carried out by community health workers using mid-upper arm circumference (MUAC) tapes or by designated screening teams assessing both MUAC and weight-for-length z-score (WLZ). In addition, children could be referred from a health center or could present at site on caretaker's initiative. Recruitment took place from September 2013 until August 2014. Children were classified with MAM if WLZ ≥ -3 and <-2 and/or MUAC was ≥115 and <125mm [17]. WLZ was determined using WHO field charts [18] and later recalculated. Final analyses of WLZ was recalculated using the package zscore06 in STATA 12 (StataCorp, US). Children could be enrolled in one of three groups fulfilling either MAM criteria for both WLZ (<-2 and ≥-3) and MUAC (<125 mm and ≥ 115 mm), WLZ only or MUAC only. WLZ was divided into 2 groups, WLZ ≥ -2 and <-2. MUAC was divided into 3 groups, MUAC ≥125 mm, ≥120 mm and <125 mm, and <120 mm.

Data collection

Weight was measured to the nearest 100 g using an electronic scale (Seca model 881 1021659), length was measured with a wooden length board to the nearest 1 mm and MUAC was measured at the midpoint between the olecranon and the acromion process to the nearest 1 mm using a standard measuring tape. All were measured in duplicate by trained study staff and the average was used. Study nurses were trained and supervised by a study physician and collected 2.5 mL of venous blood from the arm of each participant. One drop of blood was used for diagnosis of malaria on site using a rapid diagnostic test (RDT) (Bioline Malaria Ag P.f. Standard diagnostics inc.) that detects histidine rich protein 2 (HRP2) synthesized by the *Plasmodium falciparum* malaria parasite. Since HRP2, the protein detected by the RDT, can persist in blood for over a month after treatment of malaria, a positive RDT in the absence of clinical findings may be due to either a treated infection or asymptomatic malaria. We therefore present prevalence of children with a positive RDT accompanied by fever in addition to prevalence of positive RDT independent of fever. The remaining blood from each sample was collected in serum vacutainers (Becton Dickinson, reference #368492) and transported to the trial laboratory in a cold box at 2-8°C. Serum was isolated following centrifugation at 3000 RPM for 5 minutes (EBA 20S Hettich), stored at -20°C, and sent to VitMin Lab in Willstaedt, Germany for analysis of CRP and α1-acid glycoprotein (AGP). Serum CRP and AGP were determined using a simple sandwich enzyme-linked immunosorbent assay [19]. The intra- and interassay co-efficients of variation for serum CRP and AGP were <10%. Serum IGF-1 was analyzed on an Immulite 2000 Analyzer, (Siemens Healthcare, GmbH) at NEXS in Copenhagen, Denmark. Values below 25 ng/mL were not displayed automatically, but were calculated using algorithms according to correspondence with the manufacturer. The intra-assay co-efficient of variation was 20% when sIGF-1 was 10 ng/mL and 6% when sIGF-1 was 25 ng/mL. All samples were measured in duplicate and the average was used.

Study nurses collected data on illness using a patient history based on 14-day maternal recall and carried out a physical examination. A child was considered ill if the caretaker reported that their child in the previous two weeks had any of the following symptoms: fever, cough, diarrhea, vomiting, breathing problems, reduced appetite, rash, pain or swelling. A child was considered ill if the physical examination found skin problem (rash, ulcer, infection or other), respiratory tract infections, ear infection, diarrhea, oral thrush, mouth ulcer, fever or malaria. Binary morbidity variables were generated based on both maternal recall and physical examination. Diarrhea was defined as three or more loose or watery stools per day based on maternal information [20]. Respiratory tract infections were diagnosed using an adapted version of the Integrated Management of Childhood Illnesses (IMCI) guidelines. Fever was defined as an axillary temperature ≥37.5°C at physical examination [20]. Data on breastfeeding was collected based on maternal or caretaker information on present or past breastfeeding.

Fat-free mass (FFM) was assessed using the deuterium dilution technique as previously described [21, 22]. In short, pre-dose saliva samples were collected before an oral administration of 5 g D_2O diluted in 5 g of bottled water, weighed with 0.01 g precision. After a 3 hours equilibration period, post-dose saliva samples were collected. Duplicate measurements were performed on pre- and post-dose saliva samples and on a diluted sample of each child's dose. Fourier-transform infrared spectrometry was used to calculate D_2O abundance. From that, D_2O dilution space was calculated and further converted into total body water (TBW). Hydration coefficients based on age and sex were used to calculate FFM as TBW/hydration. Fat mass (FM) was defined as weight minus FFM. Typographical errors and implausible TBW values were cleaned from the data, based on the association of TBW

with length and cutoffs for FM of <-0.1 (to account for the normal technical variability in deuterium dilution studies) and >2.4 kg. To obtain length-adjusted indices, FFMI and FMI were calculated as FFM and FM divided by length in meters squared [23].

Outcomes and sample size

IGF-1 was a secondary outcome in the Treatfood study together with FFM, FM, FMI, weight, length, knee-heel length, MUAC, triceps skinfold, and nutritional recovery. The primary outcome FFMI has already been published [21]. The aims to identify sex and age patterns and correlates of serum IGF-1 were exploratory. The sample size of the current study was fixed as it involves baseline data from the Treatfood study.

Data handling and statistical analysis

Data was double entered into EPIDATA 3.1 software (Epidata Association, Odense, Denmark). All statistical analyses were carried out using STATA version 12 (StataCorp, Collage Station TX, USA). Children with missing values were excluded from the analysis. Baseline characteristics, reported illness, body composition, and serum concentrations of IGF-1, AGP and CRP were summarized as percentage (n), mean (standard deviation, SD) for normally distributed variables or median (interquartile range, IQR) for non-normally distributed variables based on visual inspection of histograms and probability plots. Linear regression models were used to assess the association of admission criteria, WLZ, length-for-age z-score (LAZ), weight-for-age z-score (WAZ), MUAC, body composition, CRP, AGP, breastfeeding, fever, positive malaria test, and a positive or negative malaria test both with and without fever with sIGF-1. CRP was divided into five groups (≤2mg/L, >2 and ≤5 mg/L, >5 and ≤10 mg/L, >10 and ≤50 mg/L and >50 mg/L). AGP was divided into three groups (<0.8 g/L, 0.8-1.2 g/L and >1.2 g/L). All linear regressions were reported unadjusted and

adjusted for age and sex and checked for collinearity using variance inflation factors (VIF). Collinearity was not present in any of the models as all VIFs ≤ 3. Additionally, the linear regression for sIGF-1 as a function of WLZ was adjusted for MUAC, sIGF-1 as a function of MUAC was adjusted for WLZ, and sIGF-1 as a function of admission criteria was adjusted for AGP. Model assumptions were checked using residual and normal probability plots respectively. An alpha of 0.05 was used in test of significance. Fractional polynomials with 95% confidence intervals (95%CI) were used to present sIGF-1 depending on age for boys and girls separately.

Results

Of 1609 children enrolled, sIGF-1 data were available on 96% (n=1546) (**Figure 1**). Of these, 21% (n=324) were enrolled based on WLZ, 29% (n=451) based on MUAC, and 50% (n=771) based on both WLZ and MUAC. Mean (±SD) age of the children was 12.4 (±4.9) months, 45% (n=699) were boys, WLZ was -2.2 (±0.5), MUAC was 123 (±4.0) mm,, 79% (1208) had been ill in the last two weeks and 94% (1453) were still breastfed (**Table 1**). The median [interquartile range] sIGF-1 at enrollment was 12 [8.2-18.3] ng/mL (Table 1). Serum IGF-1 was highest in children aged 6 months with nadir just before 10 months for girls and around 11 months for boys and it was positively associated with age from nadir to 24 months (**Figure 2**). As seen from the confidence intervals, girls had higher mean serum IGF-1 than boys throughout the age range.

Anthropometry, body composition and serum IGF-1

After adjustment for age and sex, those admitted based on MUAC only had 2.2 (95%CI 0.9,3.5) ng/mL higher sIGF-1 and those admitted based on WLZ only had 1.6 (95%CI 0.2,3.1) ng/mL higher sIGF-1 than those admitted based on both WLZ and MUAC (**Table 2**). Further adjustment for elevated serum AGP did not change these estimates (data not shown). LAZ was positively associated with sIGF-1. The regression coefficient of 1.0 (95%CI 0.5,1.5) reflects that sIGF-1 is 1.0 ng/mL higher for each 1 unit increase in LAZ. If used as categorical variable with LAZ ≥-2 as reference, then LAZ <-3 (severe stunting) was associated with a 2.8 (95%CI 0.9,4.7) ng/mL lower sIGF-1, whereas those with LAZ between -3 and -2 (moderate stunting) did not differ from the reference. WAZ and WLZ were also positively associated with sIGF (2.6 95%CI 1.7,3.4 and 2.2, 95%CI 1.0,3.3) Further adjustment of WLZ for MUAC or height did not considerably change the estimate (data not shown). Similarly, MUAC was positively associated with sIGF-1 (0.3, 95%CI 0.1, 0.4), and

192 further adjustment for WLZ or height did not considerably change the estimate (data not
193 shown). FFM was positively associated with sIGF-1 (1.5, 95%CI 0.5, 2.5), but the height-
194 adjusted index FFMI was not. Likewise, FM tended to be associated with sIGF-1 (p = 0.067),
195 but FMI was not (**Table 2**). FFM and FM were not associated with sIGF-1 after adjustment
196 for length (data not shown).
197
198 *Breastfeeding, paraclinical and clinical markers of inflammation and serum IGF-1*
199 After adjustment for age and sex, elevated serum CRP or AGP were associated with lower
200 sIGF-1 (**Table 3**). In addition, being ill in the last 2 weeks (-2.3 95%CI -3.7,-1.0), having
201 diarrhea (-2.6 95%CI -4.0,-1.3), cough (-1.5 95%CI -2.8,-0.3) or fever (-2.8 95%CI -4.3,-1.4)
202 were associated with lower sIGF-1. Similarly, fever, with (-3.6, 95%CI -5.7,-1.5) or without
203 (-2.9, 95%CI -4.9,-0.9) a positive malaria test, but not a positive malaria test without fever
204 (p=0.15), was associated with lower sIGF-1. Never (-5.1 95%CI -9.8,-0.3) and previous (-2.3
205 95%CI -5.1,0.4) breastfeeding were associated with lower sIGF-1, although the latter was
206 only a tendency (p = 0.095). Further adjustment for MUAC and WLZ did not change the
207 estimates (data not shown).

Discussion

The very low levels of sIGF-1 in our study are probably explained by all children having MAM, a high proportion with inflammation, and the young age. Previous studies in younger age groups have mainly been among children from high-income countries with little stunting and wasting.

The median sIGF-1 was 12 ng/mL, which is lower than previously found in healthy children [5, 9, 24, 25]. Another study from Burkina Faso in apparently healthy 6-23 months children found lower sIGF-1 concentrations compared to children from high-income countries [11]. However, those children had a mean sIGF-1 between 24.8 ng/mL and 28.9 ng/mL, approximately double the concentration compared to our study for both sexes. A small case-control study in malnourished children from Chile aged 5-26 months with mean WLZ of -2.7 found a mean sIGF-1 around 4 ng/mL [7]. This study supports our finding of lower sIGF-1 in malnourished children. The difference between sIGF-1 concentrations in the study from Burkina Faso and the other studies in healthy children may be due to differences in dietary intake, and perhaps differences in ethnicity [26] or analytical methods.

Age and sex

Serum IGF-1 was highest in children aged 6 months, had a nadir at around 10-11 months, and was positively associated with age from nadir until 24 months. Studies on well-nourished children have shown a similar pattern although it was less clear due to fewer children and sampling points [9, 24]. However, when combining results from different studies covering the age range up to 24 months, a similar age pattern was shown, which supports the clear age pattern shown in the present study [24]. A review showed that sIGF-1 concentrations

increased until 3 months, decreased from 3-8 months with a nadir at 8 months and then slowly increased until puberty [24].

Our study also found higher sIGF-1 in girls than in boys. This is in line with other large studies [27, 28]. Studies in children at 3 months of age found no difference in sIGF-1 between girls and boys [29, 30], but sIGF-1 in children at 12 months was higher in girls than boys [30]. The lack of difference in sIGF-1 concentrations between girls and boys at 3 months suggests that sIGF-1 are higher in girls than boys at most but not at all ages.

Inflammation and infections

The negative associations of inflammatory markers CRP and AGP with sIGF-1 supports the findings of earlier studies [15, 16]. A longitudinal study in both stunted and non-stunted Zimbabwean children between 0-18 months found that acute illness was associated with a suppression of the GH axis with lower sIGF-1 mediated via both an indirect pathway through the acute phase response and a direct pathway on sIGF-1 [16]. Not only acute infection, but also chronic inflammatory diseases in children are associated with lower sIGF-1 [13, 31]. Children from low-income settings with poor sanitation may suffer from frequent infections and environmental enteric dysfunction, a syndrome associated with intestinal inflammation [14]. Our study found that children with fever had lower sIGF-1 compared to children without fever both with or without a positive malaria test. This indicates that the inflammatory response present during fever suppresses sIGF-1 [15]. In addition, children with cough or diarrhea had lower sIGF-1. Similarly, Zimbabwean children with diarrhea and cough [16] and Brazilian children with diarrhea, but not those with cough [15], had lower sIGF-1.

A lower sIGF-1 in children with illness was further supported by the paraclinical findings. Children with a positive malaria test and fever had an even lower sIGF-1 compared to

children with only fever, diarrhea or cough, suggesting that malaria has a worse or more prolonged inflammatory response than other infections. Previous infection with malaria, without persisting fever, was not associated with lower sIGF-1, suggesting a reversibility of the depressed sIGF-1 when the inflammatory response is removed. This is supported in the Zimbabwean study, where fever the previous day was associated with lower sIGF-1, but fever two weeks before was not [16].

Anthropometry and body composition

Children with both low MUAC and low WLZ had lower sIGF-1 compared to children with only low MUAC or low WLZ. This supports previous findings that malnutrition negatively influences sIGF-1 [7]. This study found that WLZ, MUAC and HAZ were positively associated with sIGF-1. Previous studies in both high- and low-income countries have also found a positive association between anthropometric measurements and sIGF-1 [5, 11, 32, 33]. Our study found an association between FFM and sIGF-1 concurrent with other studies [5, 6, 8, 33, 34]. The association of FM with sIGF-1 in our study only tended towards significance. A Danish cohort of healthy children aged 9-36 months found that FM was positively associated with sIGF-1, but not as strongly as FFM [5]. Most other studies found similar results using both multiple skinfold measures [6, 30] and fat mass by DXA [33, 34]. Consistent with the Danish and Ghanaian studies, our study found no association between FFMI, FMI and sIGF-1, suggesting that sIGF-1 is primarily affecting linear growth in children [5, 8].

Breastfeeding

Our study found that children still breastfed had higher sIGF-1 compared to children that were never breastfed. This differs from previous studies in well-nourished children where

breastfed children had lower sIGF-1 [4, 29, 30]. This is most likely because children in high-income countries receive infant formula as a replacement for breastmilk. Infant formula has a higher concentration of protein compared to breast milk, which leads to increased levels of sIGF-1 [24, 29, 35], and even low-protein formula results in higher sIGF-1 in children than breastmilk [35]. The children in our study did not receive infant formula, but rather family-prepared foods with a low content of animal protein.

Limitations and strength

There were some limitations in our study. Firstly, the analysis kit to measure sIGF-1 was not validated to measure the low concentrations observed in this study. The intra-assay coefficient of variation was 20% when sIGF-1 was 10 ng/mL and 6% when sIGF-1 was 25 ng/mL or above, indicating that the analysis was less precise at low values. However, more than 1500 tests measured in duplicate should compensate for the larger variation. Secondly, the never-breastfed group was small and may have been insufficient to evaluate the effect of breastfeeding with accuracy, nevertheless the data for previously breastfed children tended towards a smaller but similar effect. In addition, there was an increased risk of Type 1 errors due to the multiple statistical analyses. However, for the associations with very low significance levels ($p<0.001$), the risk of chance findings is considered low. Finally, due to the cross sectional study design, there is a risk of residual confounding, which could not be adjusted for. A strength of the study is the well-defined nutritional status of the children and the large sample size, which provided greater power to identify correlates of sIGF-1.

Conclusion

LAZ and WLZ were strongly positively associated, and inflammation strongly negatively associated, with sIGF-1. As all children had MAM and many had inflammation, this probably

307 explains the very low sIGF-1 values found. Nevertheless, the age-sex pattern confirms what
308 has been found in smaller studies in high-income settings. The associations of FFM and FM
309 with sIGF-1 were fully explained by height. There is a need for prospective and nutrition
310 intervention studies to further disentangle the role of sIGF-1 in growth and health in a
311 population with undernutrition.

Acknowledgement

We acknowledge the contributions of the study participants and their families, the research staff, the Ministry of Health in Burkina Faso, the health and village authorities in Province du Passoré, and the staff at the health centres.

Statement of authors' contributions to manuscript:

SF, AB, JW, VBC, HF, and KFM designed the Treatfood trial. AI, CWY, CF, and BC conducted the research. TWK analyzed data and wrote the manuscript. BG, KM, HF, NSN, DF and VBC contributed to data analysis. TWK had primary responsibility for final content. All authors read and approved the final manuscript.

Conflicts of interest:

The authors declare no conflicts of interest.

References

1. UNICEF, W., and World Bank. *UNICEF/WHO/The World Bank Group joint child malnutrition estimates: levels and trends in child malnutrition: key findings of the 2020 edition*. 2020; Available from: https://www.who.int/publications/i/item/jme-2020-edition.
2. Benyi, E. and L. Savendahl, *The Physiology of Childhood Growth: Hormonal Regulation.* Horm Res Paediatr, 2017. **88**(1): p. 6-14.
3. Hill, D.J. and J. Hogg, *Growth factors and the regulation of pre- and postnatal growth.* Baillieres Clin Endocrinol Metab, 1989. **3**(3): p. 579-625.
4. Madsen, A.L., A. Larnkjær, C. Mølgaard, and K.F. Michaelsen, *IGF-I and IGFBP-3 in healthy 9 month old infants from the SKOT cohort: breastfeeding, diet, and later obesity.* Growth Horm IGF Res, 2011. **21**(4): p. 199-204.
5. Ejlerskov, K.T., A. Larnkjaer, D. Pedersen, C. Ritz, C. Mølgaard, and K.F. Michaelsen, *IGF-I at 9 and 36 months of age — relations with body composition and diet at 3 years — the SKOT cohort.* Growth Horm IGF Res, 2014. **24**(6): p. 239-44.
6. Ong, K., J. Kratzsch, W. Kiess, and D. Dunger, *Circulating IGF-I levels in childhood are related to both current body composition and early postnatal growth rate.* J Clin Endocrinol Metab, 2002. **87**(3): p. 1041-4.
7. Palacio, A.C., F. Perez-Bravo, J.L. Santos, L. Schlesinger, and F. Monckeberg, *Leptin levels and IgF-binding proteins in malnourished children: effect of weight gain.* Nutrition, 2002. **18**(1): p. 17-9.
8. Grenov, B., A. Larnkjær, R. Lee, A. Serena, C. Mølgaard, K.F. Michaelsen, and M.J. Manary, *Circulating Insulin-Like Growth Factor-1 Is Positively Associated with Growth and Cognition in 6- to 9-Year-Old Schoolchildren from Ghana.* J Nutr, 2020.
9. Yuksel, B., M.N. Ozbek, N.O. Mungan, F. Darendeliler, B. Budan, A. Bideci, E. Cetinkaya, M. Berberoglu, O. Evliyaoglu, E. Yesilkaya, et al., *Serum IGF-1 and IGFBP-3 levels in healthy children between 0 and 6 years of age.* J Clin Res Pediatr Endocrinol, 2011. **3**(2): p. 84-8.
10. Juul, A., P. Bang, N.T. Hertel, K. Main, P. Dalgaard, K. Jorgensen, J. Muller, K. Hall, and N.E. Skakkebaek, *Serum insulin-like growth factor-I in 1030 healthy children, adolescents, and adults: relation to age, sex, stage of puberty, testicular size, and body mass index.* J Clin Endocrinol Metab, 1994. **78**(3): p. 744-52.
11. Kouanda, S., R. Tonglet, V. De Coninck, B. Doulougou, B. Sondo, J.M. Ketelslegers, and A. Robert, *Reference values of IGF-I in children from birth to 5 years of age, in Burkina Faso, using blood samples on filter paper.* Growth Horm IGF Res, 2008. **18**(4): p. 345-52.
12. Murray, P.G. and P.E. Clayton, *Endocrine control of growth.* Am J Med Genet C Semin Med Genet, 2013. **163c**(2): p. 76-85.
13. Wong, S.C., R. Dobie, M.A. Altowati, G.A. Werther, C. Farquharson, and S.F. Ahmed, *Growth and the Growth Hormone-Insulin Like Growth Factor 1 Axis in Children With Chronic Inflammation: Current Evidence, Gaps in Knowledge, and Future Directions.* Endocr Rev, 2016. **37**(1): p. 62-110.
14. Crane, R.J., K.D. Jones, and J.A. Berkley, *Environmental enteric dysfunction: an overview.* Food Nutr Bull, 2015. **36**(1 Suppl): p. S76-87.

15. DeBoer, M.D., R.J. Scharf, A.M. Leite, A. Ferrer, A. Havt, R. Pinkerton, A.A. Lima, and R.L. Guerrant, *Systemic inflammation, growth factors, and linear growth in the setting of infection and malnutrition.* Nutrition, 2017. **33**: p. 248-253.
16. Jones, A.D., S. Rukobo, B. Chasekwa, K. Mutasa, R. Ntozini, M.N. Mbuya, R.J. Stoltzfus, J.H. Humphrey, and A.J. Prendergast, *Acute illness is associated with suppression of the growth hormone axis in Zimbabwean infants.* Am J Trop Med Hyg, 2015. **92**(2): p. 463-70.
17. Organization), W.W.H., *Supplementary Foods for the Management of Moderate Acute Malnutrition in Infants and Children 6–59 Months of Age.* 2012, WHO: Geneva.
18. Organization), W.W.H. *Weight-for-length/height: Charts* Available from: https://www.who.int/tools/child-growth-standards/standards/weight-for-length-height.
19. Erhardt, J.G., J.E. Estes, C.M. Pfeiffer, H.K. Biesalski, and N.E. Craft, *Combined measurement of ferritin, soluble transferrin receptor, retinol binding protein, and C-reactive protein by an inexpensive, sensitive, and simple sandwich enzyme-linked immunosorbent assay technique.* J Nutr, 2004. **134**(11): p. 3127-32.
20. WHO (World Health Organization), D.o.C.a.A.H.D.C., *Handbook: IMCI Integrated Management of Childhood Illness*, UNICEF, Editor. 2004.
21. Fabiansen, C., C.W. Yameogo, A.S. Iuel-Brockdorf, B. Cichon, M.J.H. Rytter, A. Kurpad, J.C. Wells, C. Ritz, P. Ashorn, S. Filteau, et al., *Effectiveness of food supplements in increasing fat-free tissue accretion in children with moderate acute malnutrition: A randomised 2 x 2 x 3 factorial trial in Burkina Faso.* PLoS Med, 2017. **14**(9): p. e1002387.
22. Fabiansen, C., C.W. Yameogo, S. Devi, H. Friis, A. Kurpad, and J.C. Wells, *Deuterium dilution technique for body composition assessment: resolving methodological issues in children with moderate acute malnutrition.* Isotopes Environ Health Stud, 2017. **53**(4): p. 344-355.
23. VanItallie, T.B., M.U. Yang, S.B. Heymsfield, R.C. Funk, and R.A. Boileau, *Height-normalized indices of the body's fat-free mass and fat mass: potentially useful indicators of nutritional status.* Am J Clin Nutr, 1990. **52**(6): p. 953-9.
24. Larnkjær, A., C. Mølgaard, and K.F. Michaelsen, *Early nutrition impact on the insulin-like growth factor axis and later health consequences.* Curr Opin Clin Nutr Metab Care, 2012. **15**(3): p. 285-92.
25. Wiley, A.S., S.M. Joshi, H.G. Lubree, D.S. Bhat, N.S. Memane, D.A. Raut, and C.S. Yajnik, *IGF-I and IGFBP-3 concentrations at 2 years: associations with anthropometry and milk consumption in an Indian cohort.* Eur J Clin Nutr, 2018. **72**(4): p. 564-571.
26. Higgins, P.B., J.R. Fernández, M.I. Goran, and B.A. Gower, *Early ethnic difference in insulin-like growth factor-1 is associated with African genetic admixture.* Pediatr Res, 2005. **58**(5): p. 850-4.
27. Closa-Monasterolo, R., N. Ferré, V. Luque, M. Zaragoza-Jordana, V. Grote, M. Weber, B. Koletzko, P. Socha, D. Gruszfeld, R. Janas, et al., *Sex differences in the endocrine system in response to protein intake early in life.* Am J Clin Nutr, 2011. **94**(6 Suppl): p. 1920s-1927s.
28. Juul, A., K. Holm, K.W. Kastrup, S.A. Pedersen, K.F. Michaelsen, T. Scheike, S. Rasmussen, J. Müller, and N.E. Skakkebaek, *Free insulin-like growth factor I serum levels in 1430 healthy children and adults, and its diagnostic value in patients*

suspected of growth hormone deficiency. J Clin Endocrinol Metab, 1997. **82**(8): p. 2497-502.

29. Chellakooty, M., A. Juul, K.A. Boisen, I.N. Damgaard, C.M. Kai, I.M. Schmidt, J.H. Petersen, N.E. Skakkebaek, and K.M. Main, *A prospective study of serum insulin-like growth factor I (IGF-I) and IGF-binding protein-3 in 942 healthy infants: associations with birth weight, gender, growth velocity, and breastfeeding.* J Clin Endocrinol Metab, 2006. **91**(3): p. 820-6.
30. Ong, K.K., M. Langkamp, M.B. Ranke, K. Whitehead, I.A. Hughes, C.L. Acerini, and D.B. Dunger, *Insulin-like growth factor I concentrations in infancy predict differential gains in body length and adiposity: the Cambridge Baby Growth Study.* Am J Clin Nutr, 2009. **90**(1): p. 156-61.
31. Savage, M.O., *Insulin-like growth factors, nutrition and growth.* World Rev Nutr Diet, 2013. **106**: p. 52-9.
32. Brennan, B.M., M. Gill, L. Pennells, O.B. Eden, A.G. Thomas, and P.E. Clayton, *Insulin-like growth factor I, IGF binding protein 3, and IGFBP protease activity: relation to anthropometric indices in solid tumours or leukaemia.* Arch Dis Child, 1999. **80**(3): p. 226-30.
33. Garnett, S., C.T. Cowell, D. Bradford, J. Lee, C. Tao, V. Petrauskas, R. Fay, and L.A. Baur, *Effects of gender, body composition and birth size on IGF-I in 7- and 8-year-old children.* Horm Res, 1999. **52**(5): p. 221-9.
34. Mäntyselkä, A., J. Jääskeläinen, A.M. Eloranta, J. Väistö, R. Voutilainen, K. Ong, S. Brage, T.A. Lakka, and V. Lindi, *Associations of lifestyle factors with serum dehydroepiandrosterone sulphate and insulin-like growth factor-1 concentration in prepubertal children.* Clin Endocrinol (Oxf), 2018. **88**(2): p. 234-242.
35. Socha, P., V. Grote, D. Gruszfeld, R. Janas, H. Demmelmair, R. Closa-Monasterolo, J.E. Subías, S. Scaglioni, E. Verduci, E. Dain, et al., *Milk protein intake, the metabolic-endocrine response, and growth in infancy: data from a randomized clinical trial.* Am J Clin Nutr, 2011. **94**(6 Suppl): p. 1776s-1784s.

Table 1: Characteristics of 1546 children aged 6-23 months with moderate acute malnutrition

Age, months	12.4 (±4.9)
Male sex	45% (699)
Length-for-age Z-score	-1.7 (±1.1)
Weight-for-age Z-score	-2.5 (±0.6)
Weight-for-length Z-score	-2.2 (±0.5)
Mid-upper arm circumference, mm	123 (±4.0)
Illness	
Ill in the last 2 weeks[1]	78% (1208)
Diarrhea[1]	20% (304)
Cough[1]	29% (452)
Fever (≥37.5°C)[2]	17% (264)
Malaria Rapid-test positive	40% (617)
Serum concentrations	
IGF-1 ng/mL	12 [8.2-18.3]
CRP mg/L	2.3 [0.8-9.3]
AGP g/L	1.3 (±0.66)
Body composition (n=1489)	
Fat-free mass index (kg/m^2)	11.6 (±0.9)
Fat-free mass (kg)	5.8 (±0.9)
Fat mass index (kg/m^2)	2.3 (±0.8)
Fat mass (kg)	1.1 (±0.4)
Breastfeeding	
Still breastfed	94% (1453)
Previously breastfed	5% (70)
Never breastfed	1% (21)

Values are presented as % (n), mean (±SD) or median [interquartile range]
Abbreviations , AGP = α_1-acid glycoprotein, CRP = C-reactive protein, IGF-1 = Insulin-like growth factor-1
[1]Based on maternal recall and physical examination at inclusion by trained study nurse
[2]Based on physical examination at inclusion by trained study nurse

Table 2: Anthropometry and body composition as correlates of serum IGF-1 (ng/mL) among 1546 children aged 6-23 months with moderate acute malnutrition

			Unadjusted		Age-sex adjusted	
	N	Median [interquartile range]	B (95%CI)	P-value	B (95%CI)	P-value
Admission criteria						
MUAC and WLZ	771	11.5 [7.9-16.8]	Ref.		Ref.	
WLZ only	324	11.9 [7.8-18.1]	0.8 (-0.6,2.3)	0.25	1.6 (0.2,3.1)	0.028
MUAC only	451	13.3 [9.4-20.6]	2.9 (1.7,4.2)	<0.001	2.2 (0.9,3.5)	0.001
Length-for-age Z-score			1.2 (0.7,1.7)	<0.001	1.0 (0.5,1.5)	<0.001
≥-2	962	12.6 [8.5-19.1]	Ref.		Ref.	
<-2 and ≥-3	426	12.0 [9.0-17.7]	-1.2 (-2.5,-0.0)	0.045	-1.0 (-2.2,0.3)	0.14
<-3	158	10.4 [7.2-13.1]	-3.5 (-5.3,-1.6)	<0.001	-2.8 (-4.7,-0.9)	0.004
Weight-for-age Z-score			3.0 (2.2,3.8)	<0.001	2.6 (1.7,3.4)	<0.001
≥-2	316	14.7 [10.1-21.9]	Ref.		Ref.	
<-2 and ≥-3	887	12.0 [8.1-17.9]	-3.4 (-4.8,-2.0)	<0.001	-2.9 (-4.3,-1.5)	<0.001
<-3	343	10.8 [7.7-15.1]	-5.4 (-7.1,-3.7)	<0.001	-4.5 (-6.2,-2.7)	<0.001
Weight-for-length Z-score			3.0 (1.9,4.1)	<0.001	2.2 (1.0,3.3)	<0.001
≥-2	455	13.3 [9.4-20.6]	Ref.		Ref.	
<-2	1091	11.8 [7.9-17.2]	-3.0 (-4.2,-1.8)	<0.001	-2.1 (-3.4,-0.9)	0.001
Mid-upper arm circumference, mm			0.1 (-0.0,0.3)	0.11	0.3 (0.1,0.4)	0.001
≥125	324	11.9 [7.9-18.1]	Ref.		Ref.	
≥120 and <125	873	12.5 [8.5-19.0]	0.6 (-0.8,2.0)	0.37	-0.6 (-2.0,0.9)	0.45
>115 and <120	349	11.4 [8.1-16.4]	-0.7 (-2.4,1.0)	0.41	-2.3 (-4.0,-0.5)	0.011
Fat-free mass index (kg/m^2)	1432		-0.2 (-0.8,0.5)	0.61	0.2 (-0.5,0.9)	0.54
Fat-free mass (kg)	1432		-0.1 (-0.8,0.5)	0.71	1.5 (0.5,2.5)	0.005
Fat mass index (kg/m^2)	1432		0.6 (-0.1,1.4)	0.097	0.4 (-0.4,1.1)	0.34
Fat mass (kg)	1432		1.4 (-0.1,2.9)	0.059	1.4 (-0.1,2.9)	0.067

Abbreviations B = Beta coefficient, IGF-1 = Insulin-like growth factor-1, MUAC = Mid-upper arm circumference, WLZ = Weight-for-length Z-score

Associations were analyzed by univariate and age- and sex adjusted linear regressions

Table 3: Breastfeeding, paraclinical and clinical markers of inflammation as correlates of serum IGF-1 (ng/mL) among 1546 children aged 6-23 months with moderate acute malnutrition

			Unadjusted		Age-sex adjusted	
	N	Median [interquartile range]	B (95%CI)	P-value	B (95%CI)	P-value
CRP mg/l						
≤2	721	12.0 [8.5-17.6]	Ref.		Ref.	
>2 and ≤5	274	13.3 [9.1-19.7]	-1.3 (-2.8,0.2)	0.10	-1.4 (-2.9,0.1)	0.065
>5 and ≤10	182	12.0 [7.6-19.6]	-1.5 (-3.3,0.3)	0.11	-1.6 (-3.4,0.1)	0.068
>10 and ≤50	277	10.5 [7.2-15.7]	-2.7 (-4.2,-1.2)	0.001	-2.6 (-4.1,-1.1)	0.001
>50	92	9.6 [7.1-12.1]	-5.4 (-7.8,-3.0)	<0.001	-5.6 (-7.9,-3.2)	<0.001
AGP g/l						
<0.8	300	15.2 [10.4-22.6]	Ref.		Ref.	
≥0.8 and ≤1.2	461	12.7 [9.0-18.8]	-2.5 (-4.1,-1.0)	0.002	-2.6 (-4.2,-1.0)	0.001
>1.2	785	10.9 [7.5-15.3]	-5.0 (-6.4,-3.5)	<0.001	-5.0 (-6.5,-3.6)	<0.001
Ill in the last 2 weeks						
No	322	14.4 [10.1-21.0]	Ref.		Ref.	
Yes	1208	11.7 [7.9-17.4]	-2.5 (-3.8,-1.1)	<0.001	-2.3 (-3.7,-1.0)	0.001
Diarrhea						
No	1242	12.7 [8.7-19.1]	Ref.		Ref.	
Yes	304	9.9 [6.9-14.4]	-2.9 (-4.3,-1.6)	<0.001	-2.6 (-4.0,-1.3)	<0.001
Cough						
No	1091	12.5 [8.7-18.9]	Ref.		Ref.	
Yes	452	11.2 [7.3-16.6]	-1.5 (-2.7,-0.3)	0.016	-1.5 (-2.8,-0.3)	0.012
Fever (≥37.5˚C)						
No	1280	12.5 [8.5-19.1]	Ref.		Ref.	
Yes	264	10.5 [7.1-14.7]	-3.0 (-4.5,-1.6)	<0.001	-2.8 (-4.3,-1.4)	<0.001
Malaria (Rapid test)						
Negative	923	12.4 [8.3-19.1]	Ref.		Ref.	
Positive	617	11.8 [8.2-17.2]	-1.1 (-2.3,-0.0)	0.049	-1.1 (-2.2,0.1)	0.064
Malaria/fever						
No malaria + no fever	782	12.8 [8.5-19.5]	Ref.		Ref.	
No malaria + fever	139	10.5 [7.4-15.1]	-2.9 (-4.9,-0.9)	0.004	-2.9 (-4.9,-0.9)	0.004
Malaria + no fever	492	12.1 [8.4-18.4]	-0.9 (-2.2,0.3)	0.14	-0.9 (-2.2,0.3)	0.15
Malaria + fever	125	10.5 [6.8-14.5]	-4.0 (-6.1,-1.9)	<0.001	-3.6 (-5.7,-1.5)	0.001
Breastfeeding						
Still breastfed	1453	12.1 [8.3-18.5]	Ref.		Ref.	
Previously breastfed	70	11.2 [7.0-16.5]	-2.2 (-4.9,0.4)	0.10	-2.3 (-5.1,0.4)	0.095
Never breastfed	21	8.3 [5.1-13.3]	-4.7 (-9.5,0.03)	0.052	-5.1 (-9.8,-0.3)	0.035

Abbreviations AGP = α_1-acid glycoprotein, B = Beta coefficient, CRP = C-reactive protein, IGF-1 = Insulin-like growth factor-1,

Associations were analyzed by univariate and age- and sex adjusted linear regressions

Figure 1: Participant flow chart

Figure 2: The relationship between age and serum IGF-1 for boys and girls in 1546 children aged 6-23 months with moderate acute malnutrition. Data are presented as fractional polynomials with 95% confidence intervals.