

Associations between Spanish children's physical activity and physical fitness with lean body mass

Abstract

The aim of the present study is to investigate the associations between physical activity (PA) and physical fitness (PF) with lean body mass (LBM) and to assess whether PA mediates the association between PF and LBM. 279 children (150 boys) aged 7.5 ± 0.3 years participated in the study. PA was assessed by accelerometry and PF with handgrip and the standing long jump test. Total lean soft tissue mass index (TLSTMI), muscle cross-sectional area index (MCSAI), and fat-free mass index (FFMI) were evaluated using dual-energy X-ray absorptiometry, peripheral quantitative computed tomography, and bioimpedance analysis, respectively.

Total ($\beta=0.247$) and vigorous PA ($\beta=0.143$) were associated with TLSTMI in girls. In boys, total ($\beta=0.337$), light ($\beta=0.290$), vigorous ($\beta=0.200$), and moderate-vigorous PA ($\beta=0.189$) were associated with TLSTMI. Only total PA was associated with FFMI ($\beta=0.299$). Handgrip strength does not mediate the relationship between total PA and TLSTMI. Positive associations were found between handgrip strength and TLSTMI, MCSAI, and FFMI in both girls and boys.

In children, there is a positive association between total and vigorous PA with TLSTMI. Handgrip strength does not mediate the relationship between total PA and TLSTMI. It was associated with TLSTMI, MCSAI, and FFMI.

Key words: lean body mass, muscle cross-sectional area, physical activity, physical fitness, schoolchildren

1. Introduction

Lean body mass (LBM) is mainly constituted by muscle mass, internal organ non-adipose components, and extracellular fluid (Kuriyan, 2018). In recent

33 years, LBM has been considered to play an essential role in growth
34 maintenance, normal development, and systemic glucose metabolism in
35 children (Liu et al., 2019). It has also been associated with the risk of
36 cardiovascular disease (S. Kim & Valdez, 2015), affecting bone health (bone
37 mineral density and structure in both sexes during childhood) (Dorsey et al.,
38 2010; Sioen et al., 2016), and cognitive development (Scheurer et al., 2018),
39 among others. Studies on children and adolescents with low lean mass showed
40 a higher cardiometabolic risk (S. Kim & Valdez, 2015), related to significantly
41 higher waist circumference, blood pressure, triglycerides, and total
42 cholesterol/high-density lipoprotein cholesterol values (Burrows et al., 2017;
43 Gracia-Marco et al., 2016). Other studies have shown an increased risk of
44 metabolic syndrome (Burrows et al., 2017; J. H. Kim & Park, 2016; K. Kim et al.,
45 2016). In this regard, the literature revealed that those presenting a phenotype
46 combining low lean mass and obesity had the most unfavorable
47 cardiometabolic risk profile (Burrows et al., 2017). Therefore, low levels of lean
48 mass in children and adolescents may represent a public health problem and a
49 burden on the health system for future stages in life.

50

51 Currently, it is accepted that several factors can influence the development of
52 lean mass/muscle mass throughout the life cycle, including fetal programming
53 (Isganaitis, 2019; Labayen et al., 2006; Larqué et al., 2019), early nutritional
54 status (Singhal et al., 2003), age, gender (Wells, 2000), hormones (Veldhuis et
55 al., 2005), diet, and physical activity and exercise (Kulkarni et al., 2014;
56 Westerterp et al., 2021)

57

58 Physical activity and exercise play an essential role in the development of both
59 the size of muscle fibers and the recruitment of motor units, thus, developing
60 strength (Dotan et al., 2012) and metabolic adaptations (Boisseau &
61 Delamarche, 2000).

62

63 A cross-sectional study, in preschool-age children between 5 and 6, showed
64 that a level of MVPA below the World Health Organization recommendation was
65 significantly associated with a lower content of muscle mass and FFM (0.8%

66 and 1.19%, respectively); these differences were more evident in boys than in
67 girls (Wyszyńska et al., 2020).

68

69 A longitudinal study (Leppänen et al., 2016, 2017) in 4.5-year-old children
70 demonstrated that children with higher and moderate to vigorous physical
71 activity (MVPA) levels had a higher fat-free mass index (FFMI) after a 12-month
72 follow-up. Similar results have been reported in preschool children (Henriksson
73 et al., 2016) and adolescents (Baxter-Jones et al., 2008; Ramires et al., 2016).

74

75

76 The relationship between physical activity levels, physical fitness, and body
77 composition with fat mass has been widely studied in children and adolescents
78 (Henriksson et al., 2016; Santos et al., 2019). Regarding its relationship with
79 LBM, Baxter-Jones et al. (Baxter-Jones et al., 2008) reported that daily physical
80 activity had a significant independent influence on LBM development during
81 adolescence (after controlling for biological maturity and stature). In addition,
82 studies in children have shown that higher fat-free mass (FFM) values are
83 associated with better cardiorespiratory fitness, improved upper and lower body
84 muscle strength (Fraser et al., 2020), motor fitness (Henriksson et al., 2016),
85 and a healthier cardiovascular profile later in life, with a lower risk of premature
86 death (Ruiz et al., 2009). Thus, it seems reasonable to hypothesize that
87 physical activity or physical fitness might play a crucial role in the association
88 between fitness-lean or physical activity-lean in this age group.

89

90 Given the available evidence, it is apparent that studies examining the
91 relationship between physical activity levels, physical fitness, and body
92 composition, specifically LBM in children between 6 and 12 years of age, are
93 limited (Hao et al., 2019). Furthermore, most of the available studies have used
94 statistical methods such as analysis of covariance, multiple linear regression, or
95 logistic regression to adjust for confounding factors, statistical methods that
96 cannot distinguish the effect of mediating variables.

97 Furthermore, there are important differences between DXA, pQCT, and BIA. On
98 the one hand, DXA is an accurate device for assessing body composition of the
99 whole body and regions/segments (Laskey, 1996). At the same time, pQCT

100 allows assessment at the appendicular level (limb muscle cross-sectional
101 area)(Frank-Wilson et al., 2015). Still, they are not available to everyone due to
102 their high price and radiation exposure (Guglielmi et al., 2016). On the other
103 hand, BIA is an effective alternative to assess body composition of the whole
104 body, low price in the absence of radiation (Orsso et al., 2019). Therefore, it will
105 be important to use all three devices to assess LBM. In addition, the
106 normalization of body size should also be considered, since most of the
107 evidence available to date uses absolute or relative measures of LBM (kg
108 and%), which makes it difficult to make comparisons between individuals or
109 populations (Wells et al., 2002).

110

111 Therefore, in this study, we hypothesized that children with higher physical
112 activity or physical fitness would have higher LBM values, and physical activity
113 or physical fitness would be a mediator between the fitness-lean or physical
114 activity-lean associations. Its purposes are to (1) investigate the associations
115 between objectively assessed physical activity, physical fitness, and LBM
116 measured by dual-energy X-ray absorptiometry (DXA), bioimpedance analysis
117 (BIA), and peripheral quantitative computed tomography (pQCT); and (2)
118 evaluate whether these associations were mediated by total physical activity or
119 physical fitness or with handgrip strength in a Spanish cohort of children.

120

121 **2. Material and Methods**

122

123 *2.1 Study participants*

124

125 This longitudinal observational study evaluated a representative cohort of
126 children born in Spain between 2009-2010. The initial sample consisted of 1602
127 newborns, followed every month during the first year and then every year until
128 they turned 6.

129

130 In 2016 and 2017, the recruited families (n=952) from the baseline examination
131 were invited to participate in this follow-up study. They were invited to
132 participate in an additional body composition assessment in our laboratory.

133 From the 415 children who participated in this follow-up assessment, 136 were

134 excluded from the study for the following reasons: lack of data on body
135 composition and physical fitness (n=90), or lack of valid accelerometer data
136 (n=46). Finally, 279 children (150 boys and 129 girls), between 6 and 8 years,
137 with complete DXA, pQCT, BIA, physical activity, and physical fitness
138 examination data were included in this study.

139

140 *2.2 Ethics statement*

141

142 This study was conducted following the Declaration of Helsinki (Fortaleza 2013
143 review) ethical guidelines. In 2009, it was approved by the Ethics Committee in
144 Clinical Research. In 2016, it was again approved by the same committee for
145 the follow-up presented in this manuscript.

146

147 All participants were evaluated after parents' signed the informed consent and
148 the verbal assent of the children.

149

150 *2.3 Body composition*

151

152 *2.3.1 Weight, height, and bioimpedance analysis (BIA):*

153

154 The weight and bioelectrical impedance analysis were measured using a
155 TANITA BC 418 MA electronic scale (Tanita Europe BV, Amsterdam,
156 Netherlands) with a 0.1 kg precision and 0-200 kg range. Following the
157 manufacturer's instructions (Tanita Corporation - Japan, 2002), the children
158 were asked to stand barefoot and with as little clothing as possible on the
159 weighing platform, touching the electrodes, in a stable position, without bending
160 the knees, and with both hands grabbing the handles.

161

162 A SECA[®] 225 portable stadiometer (SECA[®] 225, Hamburg, Germany) with a
163 precision of 0.1 cm and a range of 70-220 cm was used to determine the
164 height. The body mass index (BMI) was calculated as the weight divided by the
165 squared height (kg/m²). The AnthroPlus software from the World Health
166 Organization (WHO) (World Health Organization, 2006) was used to calculate

167 BMI age and gender-specific z-scores. The FFMI was calculated relating the
168 FFM in kilograms divided by the squared height in meters.

169

170 *2.3.2 Dual-energy X-ray absorptiometry (DXA):*

171 Dual-energy X-ray absorptiometry was used to determine the LBM (kg), using
172 the DXA QDR-Explorer™ 4500 (Hologic Inc., Bedford, Massachusetts, USA).
173 All measurements we performed following manufacturer's instructions (National
174 Health and Nutrition Examination Survey (NHANES), 2012). The participants
175 were measured lightly clothed and without metal objects or jewelry. An
176 examination of the whole body was performed. The participants were placed
177 supine, with arms along the body, without touching the trunk (Crabtree et al.,
178 2007). Analyzes and regions of interest (ROI) were determined using Pediatric
179 Hologic Corp. software version 12.4. The whole-body scan intra-measures
180 coefficient variation of LBM in our laboratory was 1.9.

181

182 This study used the sum value of the fat, lean, and bone masses (Lean soft
183 tissue mass LSTM) = LBM - bone mineral content (BMC). The total LSTM index
184 (TLSTMI) and the appendicular LSTM index (ALSTMI) were calculated by
185 dividing both the LSTM (Kg) and the appendicular LSTM (Kg) by the height
186 squared.

187

188 **In this study, the DXA and BIA analyses were carried out between 4.30 pm and**
189 **6.30 pm, 2 -4 hours after the end of the previous meal.**

190

191 *2.3.3 Peripheral quantitative computed tomography (pQCT):*

192

193 Stratec XCT 2000 L (Stratec Medizintechnik, Pforzheim, Germany) was used to
194 measure the muscle cross-sectional area (MCSA) at 66% of the total length of
195 the left tibia (González-Agüero et al., 2013), a method that has proven to be
196 valid for estimating the lean mass (Córdoba-Rodríguez et al., 2021).

197

198 All the measurements were carried out after the calibration of the equipment.

199 The total length of the tibia was determined using a segmometer, measuring the

200 distance from the cleft of the knee's medial joint to the tibia's (leg) Sphyrion
201 tibial medial malleolus (Roggen et al., 2015).

202 For the definition of the reference line in the distal tibia, an exploratory scout
203 view was performed, and the reference line was placed at the medial end of the
204 distal epiphysis. The intra-measures coefficient of variation for MCSA using
205 pQCT was 1.69, as previously reported (Gómez-Bruton et al., 2014).

206 The muscle cross-sectional area index (MCSAI) was calculated by dividing the
207 muscle cross-sectional area (MCSA) by the squared height.

208

209 The pQCT and DXA images were evaluated visually by a technician to identify
210 motion artifacts. The images showing movement were subjected to a new
211 exploration or excluded from the data analysis. Moreover, an external evaluator
212 reviewed all the images to endorse the image quality.

213

214 *2.4 Physical activity*

215 Physical activity was evaluated using the ActiGraph wGT3x-BT triaxial
216 accelerometer (ActiGraph, Ft. Walton Beach, USA). Parents/caregivers and
217 children were asked to place the accelerometer under the child's clothing at the
218 level of the right iliac crest, during their waking day, for seven consecutive days
219 and to complete a diary to assess the minutes and reasons why accelerometers
220 were not being used (Bammann et al., 2011).

221 Additionally, children and parents / caregivers were advised not to use
222 accelerometers in water activities.

223 All the accelerometers were programmed to record data every 15 seconds
224 (epoch). Twenty minutes or more of consecutive zero counts were defined as
225 unused time.

226 For the information processing, a day in which the child used the accelerometer
227 for at least 8 hours was considered a valid day. A minimum of 3 days was
228 required, including at least one weekend day, as was previously used by other
229 authors in similar population groups (Moliner-Urdiales et al., 2010a)

230

231 Actilife software version 6.0 (ActiGraph, Pensacola, FL., USA) was used for
232 data processing. The cut-off points determined by Evenson were used as a
233 reference to define the time dedicated to sedentary and different intensity
234 physical activity (sedentary: 0–100, light: 101–2295, moderate: 2296–4011,
235 vigorous: 4012, and more accelerometer counts per minute [cpm]) (Evenson et
236 al., 2008).

237 The absolute amount of time in each intensity category determined from the
238 accelerometer data was used for the analyzes, taking as a reference that the
239 World Health Organization (WHO)(World Health Organization, 2020) frequently
240 uses the min/day metric to issue their physical activity recommendations.

241

242 Most of the children had complete actigraphic data of 7 days (n=126, 42.2%) or
243 6-5 days (n=118, 42.4%) of. The average follow-up included 6.2 days of
244 recordings.

245

246 *2.5 Upper limb strength - Handgrip strength test:*

247 A TKK-5401 (Takei Scientific Instruments Co., Ltd., Niigata, Japan) digital
248 handgrip dynamometer with a precision of 0.1kg and adjustable grip was used
249 according to the size of the hand derived from the following gender-specific
250 equations (España-Romero et al., 2008):

251 Boys: $Y = X/4 + 0.44 \text{ cm}$

252 Girls: $Y = 0.3X - 0.52 \text{ cm}$

253 Where Y = optimal grip and X = size of the wide-open hand, measured from the
254 tip of the thumb to the tip of the little finger.

255 During the measurement, participants stood, elbow extended, avoiding any
256 bodily contact with the dynamometer, except with the measured hand. The
257 children were asked to press as hard as possible for 3 to 5 seconds with each
258 hand; two attempts were made with each hand, three minutes of rest interval
259 between each of them. The dynamometer display was aligned to face the
260 examiner, providing blind measurements to the children. The final score was

261 calculated as the average of the best attempt obtained for the left and right
262 hands in kg (De Miguel-Etayo et al., 2014).

263 *2.6 Lower limb strength. Standing long jump test*

264 The long jump test was used to assess the explosive strength of the lower
265 extremities. The children were asked to stand, feet slightly apart, in front of a
266 starting line. Then, from a standing position, a forward jump was made using
267 the arms' impulse, being careful not to step on the starting line and pushing with
268 both feet at the same time. The measurement was made from the impulse line
269 to the heel closest to the start line and recorded in cm (López-Gil et al., 2020).
270 Two attempts were made, recording the highest value obtained.

271 *2.7 Covariates*

272 We recorded the following variables for all the children:

273 *2.7.1 Parental education*

274 *Highest level of education:* the parents were asked to report their highest
275 achieved level of education when they came to our lab in 2016-2017 (no
276 studies; basic-primary studies; intermediate studies [including modules
277 vocational training and secondary studies]; higher studies; and university
278 degrees). These were later coded according to the International Standard
279 Classification of Education (ISCED-2011) (Statistics, 2012) and categorized
280 again into low (0-2), medium (3-4), and high (5-8) educational levels (Unesco,
281 1997).

282 *2.7.2 Anthropometric data of the child at birth:*

283 At the beginning of the study, in the first visit (15 days after birth), the pediatric
284 and nursing staff conducting the *Programa de Salud Infantil* (Child Health
285 Program) collected the birth weight variable.

286 *2.7.3 Food frequency questionnaire and diet quality index (DQI):*

287 A previously validated, semi-quantitative food frequency questionnaire was
288 used to assess the dietary intake (Mouratidou et al., 2019), including 37 foods
289 and beverages. The responding parents were asked to classify each food's
290 consumption frequency using a 6-Point Likert scale ranging from never to every
291 day (never/less than once a month, 1 to 3 times per month, 1 day/week, 2 to 4
292 days/week, 5 to 6 days/week, and every day) and serving size.

293 The DQI was subsequently calculated. This index mainly assesses the
294 components of dietary diversity, dietary quality, and dietary balance. Dietary
295 diversity evaluated the daily consumption of at least one food serving from the
296 eight recommended food groups. This component's score ranged from 0 to 9 for
297 at least one serving from a recommended food group. Diet quality expressed
298 whether the children made optimal decisions on the food quality within a food
299 group. Dietary balance was calculated from the difference between the food
300 intake's suitability and its excess in the diet (Huybrechts et al., 2010; Vyncke et
301 al., 2013). These three components of the DQI were presented in percentages.
302 The categories' scores were summed and divided by 3 to compute the overall
303 DQI, resulting in scores ranging from -33% to 100%. The components' mean
304 were used to calculate the DQI, with the highest scores reflecting greater
305 compliance with the diet (Iglesia et al., 2020).

306 *2.8 Statistical analysis*

307 Statistical analyses were carried out using IBM SPSS Statistics® software,
308 version 25 (IBM Corp., Armonk, NY, USA). The variables studied were
309 presented as means (M) ±, standard deviations (SD), and median and
310 interquartile intervals (25th and 75th) in the case of non-normally distributed
311 variables. The distribution of variables was checked and verified using the
312 Kolmogorov-Smirnov test. The Student's t-test or the Mann-Whitney U test were
313 used to identify sex differences between each of the studied variables. Because
314 we observed a significant interaction effect between gender and physical
315 activity and physical fitness, all the analyses were performed separately for
316 boys and girls.

317 Multiple linear regression (Forced Entry) was used to study the association
318 between physical activity (total, light, moderate, vigorous, or MVPA) or physical
319 fitness (handgrip strength, standing long jump) with the body composition
320 outcomes, which included TLSTMI, MCSAI, and FFMI. These relationships
321 were analyzed in individual regression models (one for each physical activity
322 level or physical fitness variable) using educational level, DQI, and birth weight
323 as covariates. The assumptions of independence of errors were verified for all
324 the multiple regression models using the Durbin-Watson test. Equally,
325 collinearity diagnosis was carried out through the variance inflation factor (VIF).

326 For the significant models, we used a mediation analysis according to the
327 procedure proposed by Baron and Kenny (Baron & Kenny, 1986) using the
328 macro PROCESS developed by Hayes (Hayes, 2013) with a bootstrap
329 threshold of 10,000 and model 4 for Statistical Package for the Social Sciences
330 (SPSS) version 25.0 (IBM Corporation, New York, USA). The purpose of this
331 model was to examine the total effect (c) and the individual direct effects (a, b,
332 c') reflected by the non-standardized regression coefficient (β), as well as the
333 statistical significance of the relationship between each model's independent
334 and dependent variables. The model also examined the indirect effect obtained
335 from the coefficients' product ($a \times b$). It indicated the change in TLSTMI by the
336 change in vigorous physical activity and the change in TLSTMI by the handgrip
337 strength change, mediated by the proposed mediator (handgrip strength or
338 physical activity, respectively). If zero was not included in the estimate's 95%
339 confidence interval (CI), we concluded that the indirect effect (IE) was
340 statistically significant, as shown in Figure 1.

341 **3. Results**

342

343 *3.1 Descriptive statistics*

344

345 The characteristics of participants ranked by gender are shown in **Table 1**. Boys
346 showed higher levels of TLSTM (kg), TLSTMI (kg/m^2), ALSTMI (kg/m^2), MCSA
347 (mm^2), MCSAI (mm^2/m^2), FFM (Kg), FFMI (kg/m^2), total, light, moderate,

348 vigorous, and MVPA, handgrip strength and standing long jump than girls (all
349 $p < 0.05$).

350

351 *3.2 Association between physical activity or physical fitness with LBM outcomes*

352

353 Adjusted associations between physical activity and TLSTMI, MCSAI, and FFMI
354 for both boys and girls are shown in **Table 2**.

355

356 In girls, we observed that for Model 1 (physical activity [min/day]), there was an
357 association between total physical activity and TLSTMI ($\beta = 0.272$, $p = 0.004$). In
358 Model 2, after adding the DQI, the parent's educational level, and age, an
359 association was found between total physical activity ($\beta = 0.275$, $p = 0.004$) and
360 vigorous physical activity ($\beta = 0.159$, $p = 0.001$) with TLSTMI. Finally, once the
361 birth weight was added to create Model 3, slight decreases were found for the
362 significant association between total physical activity ($\beta = 0.247$, $p = 0.010$) and
363 vigorous physical activity ($\beta = 0.143$, $p = 0.001$) with TLSTMI.

364

365 In boys, Model 1 showed a positive association between total physical activity
366 ($\beta = 0.335$, $p < 0.001$), light ($\beta = 0.287$, $p = 0.001$), and vigorous physical activity
367 ($\beta = 0.203$, $p = 0.020$) with TLSTMI by DXA. After adding the DQI, the parents'
368 educational level, and the age to create Model 2, slight increases were found in
369 the significant associations between total physical activity ($\beta = 0.371$, $p < 0.001$),
370 light physical activity ($\beta = 0.302$, $p = 0.001$), vigorous ($\beta = 0.224$, $p = 0.013$). An
371 association was found between MVPA ($\beta = 0.189$, $p = 0.038$) with TLSTMI (Table
372 2). Once birth weight was added to create Model 3, slight decreases were
373 observed in the associations between total physical activity ($\beta = 0.337$, $p < 0.001$),
374 light ($\beta = 0.290$, $p = 0.001$) and vigorous physical activity ($\beta = 0.200$, $p = 0.023$) with
375 TLSTMI (Table 2). The association between MVPA and TLSTMI in this model
376 disappeared.

377

378 No associations were found between physical activity and MCSAI for girls or
379 boys. Regarding FFMI assessed with bioimpedance, no associations were
380 found between FFMI and physical activity in girls. For boys, total physical
381 activity significantly predicted FFMI.

382

383 The adjusted associations between handgrip strength and TLSTMI, MCSAI,
384 and FFMI both in boys and girls are shown in **Table 3**. For both genders,
385 handgrip strength was a significant predictor for all the models. For girls, in
386 Model 1, handgrip strength was positively associated with TLSTMI, MCSAI, and
387 FFMI ($\beta=0.363$, $p<0.001$; $\beta=0.262$, $p=0.002$; $\beta=0.236$, $p=0.004$, respectively). In
388 Model 2, significant associations remained for TLSTMI, MCSAI, and FFMI
389 ($\beta=0.350$, $p<0.001$; $\beta=0.282$, $p=0.001$; $\beta=0.218$, $p=0.008$, respectively).
390 However, they decreased slightly for TLSTMI and FFMI. Finally, once birth
391 weight was added to Model 3, the significant associations again decreased
392 slightly, but remained significant ($\beta=0.314$, $p<0.001$; $\beta=0.267$, $p=0.002$;
393 $\beta=0.196$, $p=0.017$, respectively).

394

395 In boys, in Model 1 handgrip strength was positively associated with TLSTMI,
396 MCSAI, and FFMI ($\beta=0.478$, $p<0.001$; $\beta=0.273$, $p<0.001$; $\beta=0.289$, $p<0.001$,
397 respectively). In Model 2, significant associations were maintained for TLSTMI,
398 MCSAI, and FFMI ($\beta=0.464$, $p<0.001$; $\beta=0.287$, $p<0.001$; $\beta=0.307$, $p<0.001$,
399 respectively) but decreased slightly for TLSTMI and increased for MCSAI and
400 BIA. Finally, once birth weight was added to Model 3, the significant
401 associations again decreased slightly for TLSTMI and increased for MCSAI and
402 BIA ($\beta=0.425$, $p<0.001$; $\beta=0.301$, $p<0.001$; $\beta=0.325$, $p=0.008$, respectively).

403

404 There was no association between standing long jump and TLSTMI, MCSAI,
405 and FFMI in both girls and boys (Table 3). This analysis was also performed
406 adjusting standing long jump by weight, obtaining similar results.

407

408 *3.3 Mediation analysis*

409

410 An analysis of the handgrip's strength mediation effect on the association of
411 total physical activity with TLSTMI values indicated an association between total
412 physical activity and handgrip strength ($\beta_1=0.0057$, 95% CI: 0.0000 to 0.0114, p
413 0.0484^*) in the first regression equation (a). In the second equation (b),
414 handgrip strength was positively associated with TLSTMI ($\beta_3=0.2188$, 95% CI:
415 0.1637 to 0.2739, $p<0.001$). The third equation (c') showed a positive

416 relationship between total physical activity and TLSTMI ($\beta_{dir}=0.0073$, 95% CI:
417 0.0045 to 0.0101 $p<0.001^*$). The relationship between total physical activity and
418 TLSTMI was not statistically significant when including handgrip strength in the
419 model (a * b), indicating that handgrip strength does not mediate in this
420 relationship ($\beta_{ind}=0.0013$, 95% CI: 0.0000 a 0.0026) (Figure 1).

421

422 **4. Discussion**

423

424 Our results indicate that this sample of children accumulated around 110 min of
425 MVPA. Studies available in the population of the United States (Troost et al.,
426 2002) and Europe (Riddoch et al., 2004), where physical activity data were
427 objectively measured, suggest that young children (schoolchildren) accumulate
428 more than 100 min of MVPA per day. Our results are comparable with these
429 findings. This could be because children are more active at these ages
430 (possibly through organized activities (physical exercise and youth sports) and
431 not organized (recess and unstructured activity (Wickel & Eisenmann, 2007),
432 and active transport (Carver et al., 2011))), to later decrease the physical
433 activity in adolescence, especially in girls.

434

435 Our main findings indicate that there is an association between total physical
436 activity and vigorous physical activity with TLSTMI in girls, while for boys, the
437 associations were found between total, light, vigorous, and MVPA with TLSTMI.
438 One possible explanation may be the role of certain hormones (growth
439 hormone, IGF-1, and gender steroids, such as testosterone and estradiol),
440 which play a fundamental role in developing skeletal muscle during infancy,
441 childhood, and adolescence, depending on gender (Veldhuis et al., 2005).

442

443 Additionally, our results indicate that upper limb strength (handgrip strength)
444 does not mediate the relationship between total physical activity and TLSTMI.
445 As far as we know, this is the first time that these results are shown in a broad
446 sample of young children using high precision objective methods.

447

448 In agreement with our results, a study involving 283 Chinese adolescent girls
449 also found a significant positive association between total physical activity level

450 assessed by questionnaire and LBM ($p < 0.001$), suggesting that higher physical
451 activity levels may reflect a higher LBM (Foo et al., 2007). Similarly, a study by
452 Jiménez-Pavón study using accelerometers found that total physical activity
453 was positively associated with FFM ($p < 0.05$) in 2,200 (1016 male, 1184
454 females) European adolescents of both sexes (Jiménez-Pavón et al., 2013). In
455 the same line, Deheeger et al. (Deheeger et al., 1997) found that total physical
456 activity was positively associated with the percentage of FFM ($r = 0.23$; $p = 0.03$) in
457 ten-year-old children ($n = 86$). Rennie et al. also showed that the level of physical
458 activity was positively associated with the lean mass index (LMI) in a study
459 carried out with 100 children aged 6 to 8 (Rennie et al., 2005). Our study is in
460 the same line as previously published studies, suggesting that total physical
461 activity positively affects non-adipose tissue in both female and male children
462 and adolescents.

463

464 Regarding vigorous physical activity and MVPA, a study by Jiménez-Pavón
465 found positive associations of vigorous physical activity with FFM and muscle
466 mass (all $p < 0.05$) in both sexes (Jiménez-Pavón et al., 2013). In this study,
467 skinfold thickness and bioimpedance were used to evaluate body composition,
468 accelerometry for physical activity. Similarly, a study by Hao et al. found a
469 positive association between MVPA and SMMI ($\beta = 0.20$, $p < 0.001$). It involved
470 640 adolescents and concerned fat-free soft tissue mass assessed by DXA,
471 subsequently determining the skeletal muscle mass index (SMMI) (Hao et al.,
472 2019). Additionally, in a longitudinal study conducted in Pelotas (Brazil)
473 (Ramires et al., 2016), a consistent moderate and vigorous physical activity
474 practice during adolescence was associated with a greater lean mass index in
475 both sexes.

476

477 Conversely, studies such as the one developed by Moliner-Urdiales et al.
478 (Moliner-Urdiales et al., 2010a) in a sample of 363 Spanish adolescents aged
479 12.5 to 17.5 did not observe an association between FFM and physical activity
480 levels. Heelan et al. (Heelan & Eisenmann, 2006), in 4- to 7-year-old children,
481 observed, only in girls, a negative correlation between MVPA and FFM ($r = -$
482 0.39 , $p < 0.05$).

483

484 Different factors could explain our contrasting results. They include the
485 individuals' age ranges included in each study (preschoolers (Leppänen et al.,
486 2016), adolescents (Foo et al., 2007; Hao et al., 2019; Jiménez-Pavón et al.,
487 2013), and schoolchildren (Deheeger et al., 1997; Rennie et al., 2005)) and
488 their maturation stage. Other factors include the methods for assessing body
489 composition (DXA (Foo et al., 2007; Hao et al., 2019; Heelan & Eisenmann,
490 2006), anthropometry (Deheeger et al., 1997; Jiménez-Pavón et al., 2013), BIA
491 (Jiménez-Pavón et al., 2013), isotopic dilution (Rennie et al., 2005), and air
492 displacement plethysmography (Leppänen et al., 2016)) and physical activity
493 (questionnaires (Deheeger et al., 1997; Foo et al., 2007; Ramires et al., 2016;
494 Rennie et al., 2005) and accelerometers (Jiménez-Pavón et al., 2013)). They
495 also include the different types of devices for assessing physical activity (
496 uniaxial (Hao et al., 2019; Heelan & Eisenmann, 2006; Moliner-Urdiales et al.,
497 2010b) or triaxial devices (Jiménez-Pavón et al., 2013; Leppänen et al., 2016),
498 as well as the different cutoff points to define physical activity intensities,
499 varying sampling intervals (epochs) (10s (Leppänen et al., 2016), 15 s
500 (Jiménez-Pavón et al., 2013) and 1min (Hao et al., 2019; Heelan & Eisenmann,
501 2006)), and sporadic or episodes of accumulated physical activity data.

502 Moreover, the presence of FFM (sum of lean mass and bone mass) or fat-free
503 soft tissue mass (FFM - bone mineral content) and the use of absolute values
504 (kg (Foo et al., 2007; Jiménez-Pavón et al., 2013)) or relative values (%
505 (Deheeger et al., 1997) or index (Hao et al., 2019; Leppänen et al., 2016;
506 Ramires et al., 2016; Rennie et al., 2005)) to express the value of the FFM/LBM
507 can make it difficult to compare individuals of different sizes appropriately
508 because FFM varies with height, weight, age. The percentage of FFM
509 automatically decreases in proportion to the increase in the % of body fat. The
510 last possible affecting factors are the covariates taken into account in the
511 analyzes (age and maturation stage, the mother's BMI and educational level,
512 the father's BMI and educational level, and the age at the time of measurement
513 and time of use awake).

514

515 Some of these factors were also reported in a systematic review carried out by
516 Poitrasl et al. (Poitras et al., 2016). Their purpose was to examine the
517 relationships between objectively measured physical activity and relevant

518 indicators (body composition, cardiometabolic biomarkers, and physical fitness,
519 among others) in 5- to 17-year-old children and adolescents.

520

521 Our findings can support emerging evidence suggesting that different physical
522 activity intensities, including light physical activity, may significantly affect health
523 outcomes. They are in line with Corson's findings in 1,731 adolescents aged 12
524 to 19, evaluated during the National Health and Nutrition Examination Survey
525 2003/2004 and 2005/2006 (Carson et al., 2013)—reinforcing the claim that
526 some activity is better than none, but that “more is better” (Tremblay et al.,
527 2011). However, we also found that the assessment method could influence
528 results. No significant associations were found when evaluating the
529 associations using the variables of the lean component determined with BIA or
530 pQCT. An explanation for this could be the fact that, although BIA can provide
531 information on the whole-body FFM status (like DXA), alterations in body water,
532 such as dehydration, can influence it, increasing resistance to electricity, and
533 subsequently underestimating the FFM (or overestimating body fat). Another
534 influence could be the time of the day. In this study, the DXA and BIA analyses
535 were carried out between 4.30 pm and 6.30 pm to ensure that no child was
536 fasting, influencing the results. Other influences could include food consumption
537 and recent activity (exercise) (Heymsfield et al., 2015).

538

539 On the other hand, pQCT quantifies the cross-sectional area of the peripheral
540 (appendicular)/limb muscle (MCSA) (Frank-Wilson et al., 2015) and not the
541 whole body, which could explain why we found no associations between
542 physical activity and MCSAI. MCSA can also be influenced by the
543 anthropometric characteristics of the subjects (body and limb size); therefore,
544 we used MCSAI. Meanwhile, DXA provides information on the LSTM
545 (Bazzocchi et al., 2016) of the whole body or regional/segmental zones
546 (Laskey, 1996), with the advantages of its high precision and reproducibility
547 (Guglielmi et al., 2016).

548

549 Our main findings also suggest a positive association between upper body
550 muscle strength (handgrip strength) and TLSTMI, MCSAI, and FFMI in both
551 boys and girls, after controlling for gender, education level, DQI, and birth

552 weight. The results were consistent regardless of the methodology used to
553 assess body FFM; however, the associations were weaker when BIA was used
554 to measure LSTM markers. Similar results were found in the MINISTOP (mobile
555 device-based intervention aimed at stopping obesity in pre-schoolers) study
556 (Henriksson et al., 2016), where PREFIT (physical fitness test battery in
557 preschool children) was used to measure physical fitness (Ortega et al., 2015)
558 in 303 4-year-old children. Its associations showed a higher FFMI in participants
559 with better upper body muscle strength (handgrip strength) ($\beta=0.39$, $p<0.001$).

560

561 Anthropometric variables such as body height, body mass, and BMI, which can
562 influence grip strength, may explain these findings. In the case of body height, it
563 is directly correlated with grip strength; this could partially explain its close
564 relation to LBM (Jürimäe et al., 2009). Both the MINISTOP study and the
565 present study made the respective height adjustment.

566

567 The present study found no association between standing long jump and
568 TLSTMI, MCSAI, and FFMI. However, another study by Vicente-Rodriguez et
569 al. (Vicente-Rodriguez et al., 2004) showed an increase in the FFM of 28
570 children (soccer players) followed for three years. They found no significant
571 differences in muscle strength after evaluation through vertical jumps and
572 maximal isometric force (MIF) during leg extension. Conversely, Henriksson et
573 al. (Henriksson et al., 2016) observed that higher FFMI was associated with
574 improved lower-body muscular strength ($\beta=0.22$, $p<0.001$). This disparity could
575 be explained by the different populations included in each of the studies
576 (preschoolers, schoolchildren, and soccer players), as well as their maturation
577 stage (preschoolers, schoolchildren, and pre-puberty), the body composition
578 evaluation methods (DXA and air displacement plethysmography) and the
579 measurement methodology for the forces generated during vertical jumps (force
580 plate versus standing long jump test). The lack of association between lower
581 limb strength and FFM may also be because this test is considered weight-
582 dependent, requiring propulsion or body elevation. Therefore, children and
583 adolescents with higher LBM may not display better performance in these tests
584 because they likely weigh differently. These results could suggest that higher

585 performance in some physical fitness tests may occur in underweight
586 individuals because of the lower load that they must move.

587

588 Finally, the relationship between physical activity and physical fitness (handgrip
589 strength) and TLSTMI in our study is mainly consistent with the existing
590 evidence showing positive associations independent of physical activity and
591 physical fitness in TLSTMI. Our research shows that the association of total
592 physical activity with TLSTMI is not mediated by physical fitness (handgrip
593 strength). Therefore, future actions seeking to improve TLSTM in boys and girls
594 should encourage activities that enhance physical activity levels and physical
595 fitness.

596

597 *4.1 Limitations and Strengths*

598

599 Within the limitations of the present study, there is the fact that the data cannot
600 be extrapolated to other populations. Studies with different age groups and
601 different ethnic groups should be developed in the future. Furthermore, due to
602 its cross-sectional design, the observed associations cannot be interpreted as
603 causal relationships.

604

605 Despite these limitations, the strengths of our study are many. To date, studies
606 on a homogeneous sample with such young children are limited. The use of
607 accelerometers allowed the objective assessment of physical activity, including
608 both weekdays and weekend days. LSTM and FFM were measured using two
609 different techniques, DXA and BIA, the former, a robust method accepted as
610 the gold standard method. MCSA was measured by pQCT, a lower-cost method
611 previously validated against DXA in this population. The DXA and pQCT
612 measurements and the respective image analysis were performed by two
613 trained researchers in each of the techniques. The TLSTM, ALSTM, MCSA, and
614 FFM results were adjusted for the height of the subjects. Different tests were
615 used to assess muscle strength (handgrip strength and standing long jump),
616 controlling for several confounders, including gender, age, birth weight, DQI,
617 and parents' education level. Furthermore, it appears to be the first study
618 evaluating handgrip strength as a potential mediator of the relationship between

619 physical activity and TLSTMI in children.

620

621 **5. Conclusions**

622

623 The present study suggests positive associations between total and vigorous
624 physical activity and TLSTMI in boys and girls aged 6 to 8. Its results indicate
625 that upper limb strength (handgrip strength) does not mediate the relationship
626 between total physical activity and TLSTMI.

627

628 The results also suggest an association between handgrip strength and
629 TLSTMI, MCSAI, and FFMI in 6- to 8-year-old children. More studies are
630 needed in the future to clarify the relationships between physical activity and
631 physical fitness with TLSTM, MCSA, and FFM better, taking into account other
632 confounding factors such as genetics factors, as well as physical activity
633 intensities. To date, most of the available studies focus on the relationship at
634 higher intensities of physical activity (i.e., MVPA and vigorous physical activity).

635

636 Public policies should encourage participation in all physical activities, including
637 light physical activity. As we have shown, it can be an effective substitute for
638 sedentary activities and expand the focus beyond the MVPA as a strategy to
639 promote our children's health. Likewise, exercises that improve upper extremity
640 strength and lean mass should be encouraged, which, as mentioned above, is
641 an important tissue in children.

642

643 **Disclosure Statement**

644

645 The authors report no conflict of interest.

646

647

648 **Bibliography**

649 Bammann, K., Sioen, I., Huybrechts, I., Casajús, J. A., Vicente-Rodríguez, G.,
650 Cuthill, R., Konstabel, K., Tubić, B., Wawro, N., Rayson, M., Westerterp, K.,
651 Mårild, S., Pitsiladis, Y. P., Reilly, J. J., Moreno, L. A., & De Henauw, S.

652 (2011). The IDEFICS validation study on field methods for assessing
653 physical activity and body composition in children: Design and data
654 collection. *International Journal of Obesity*, 35, S79–S87.
655 <https://doi.org/10.1038/ijo.2011.38>

656 Baron, R. M., & Kenny, D. A. (1986). The moderator–mediator variable
657 distinction in social psychological research: Conceptual, strategic, and
658 statistical considerations. *Journal of Personality and Social Psychology*,
659 51(6), 1173. <https://doi.org/https://doi.org/10.1037/0022-3514.51.6.1173>

660 Baxter-Jones, A. D. G., Eisenmann, J. C., Mirwald, R. L., Faulkner, R. A., &
661 Bailey, D. A. (2008). The influence of physical activity on lean mass accrual
662 during adolescence: a longitudinal analysis. *Journal of Applied Physiology*,
663 105(2), 734–741.
664 <https://doi.org/https://doi.org/10.1152/jappphysiol.00869.2007>

665 Bazzocchi, A., Ponti, F., Albisinni, U., Battista, G., & Guglielmi, G. (2016). DXA:
666 Technical aspects and application. *European Journal of Radiology*, 85(8),
667 1481–1492. <https://doi.org/10.1016/j.ejrad.2016.04.004>

668 Boisseau, N., & Delamarche, P. (2000). Metabolic and hormonal responses to
669 exercise in children and adolescents. *Sports Medicine*, 30(6), 405–422.
670 <https://doi.org/10.2165/00007256-200030060-00003>

671 Burrows, R., Correa-Burrows, P., Reyes, M., Blanco, E., Albala, C., & Gahagan,
672 S. (2017). Low muscle mass is associated with cardiometabolic risk
673 regardless of nutritional status in adolescents: A cross-sectional study in a
674 Chilean birth cohort. *Pediatric Diabetes*, 18(8), 895–902.
675 <https://doi.org/10.1111/pedi.12505>

676 Carson, V., Ridgers, N. D., Howard, B. J., Winkler, E. A. H., Healy, G. N.,
677 Owen, N., Dunstan, D. W., & Salmon, J. (2013). Light-Intensity Physical
678 Activity and Cardiometabolic Biomarkers in US Adolescents. *PLoS ONE*,
679 8(8). <https://doi.org/10.1371/journal.pone.0071417>

680 Carver, A., Timperio, A. F., Hesketh, K. D., Ridgers, N. D., Salmon, J. L., &
681 Crawford, D. A. (2011). How is active transport associated with children's
682 and adolescents' physical activity over time? *International Journal of*
683 *Behavioral Nutrition and Physical Activity*, 8, 1–6.

- 684 <https://doi.org/10.1186/1479-5868-8-126>
- 685 Córdoba-Rodríguez, D. P., Iglesia, I., Gomez-Bruton, A., Miguel-Berges, M. L.,
686 Flores-Barrantes, P., Casajús, J. A., Moreno, L. A., & Rodríguez, G. (2021).
687 Quantitative peripheral computed tomography to measure muscle area and
688 assess lean soft tissue mass in children. *Annals of Human Biology*, 1–27.
689 <https://doi.org/10.1080/03014460.2021.1877352>
- 690 Crabtree, N. J., Leonard, M. B., & Zemel, B. S. (2007). *Dual-Energy X-Ray*
691 *Absorptiometry BT - Bone Densitometry in Growing Patients: Guidelines*
692 *for Clinical Practice* (A. J. Sawyer, L. K. Bachrach, & E. B. Fung (eds.); pp.
693 41–57). Humana Press. https://doi.org/10.1007/978-1-59745-211-3_3
- 694 De Miguel-Etayo, P., Gracia-Marco, L., Ortega, F. B., Intemann, T., Foraita, R.,
695 Lissner, L., Oja, L., Barba, G., Michels, N., Tornaritis, M., Molnár, D.,
696 Pitsiladis, Y., Ahrens, W., & Moreno, L. A. (2014). Physical fitness
697 reference standards in European children: The IDEFICS study.
698 *International Journal of Obesity*, 38(September), S57–S66.
699 <https://doi.org/10.1038/ijo.2014.136>
- 700 Deheeger, M., Rolland-Cachera, M. F., & Fontvieille, A. M. (1997). Physical
701 activity and body composition in 10 year old French children: Linkages with
702 nutritional intake? *International Journal of Obesity*, 21(5), 372–379.
703 <https://doi.org/10.1038/sj.ijo.0800415>
- 704 Dorsey, K. B., Thornton, J. C., Heymsfield, S. B., & Gallagher, D. (2010).
705 Greater lean tissue and skeletal muscle mass are associated with higher
706 bone mineral content in children. *Nutrition and Metabolism*, 7, 1–11.
707 <https://doi.org/10.1186/1743-7075-7-41>
- 708 Dotan, R., Mitchell, C., Cohen, R., Klentrou, P., Gabriel, D., & Falk, B. (2012).
709 Child-adult differences in muscle activation - A review. *Pediatric Exercise*
710 *Science*, 24(1), 2–21. <https://doi.org/10.1123/pes.24.1.2>
- 711 España-Romero, V., Artero, E. G., Santaliestra-Pasias, A. M., Gutierrez, A.,
712 Castillo, M. J., & Ruiz, J. R. (2008). Hand Span Influences Optimal Grip
713 Span in Boys and Girls Aged 6 to 12 Years. *Journal of Hand Surgery*,
714 33(3), 378–384. <https://doi.org/10.1016/j.jhsa.2007.11.013>
- 715 Evenson, K. R., Catellier, D. J., Gill, K., Ondrak, K. S., & McMurray, R. G.

- 716 (2008). Calibration of two objective measures of physical activity for
717 children. *Journal of Sports Sciences*, 26(14), 1557–1565.
718 <https://doi.org/10.1080/02640410802334196>
- 719 Foo, L. H., Zhang, Q., Zhu, K., Ma, G., Greenfield, H., & Fraser, D. R. (2007).
720 *Influence of body composition , muscle strength , diet and physical activity*
721 *on total body and forearm bone mass in Chinese adolescent girls*. 1281–
722 1287. <https://doi.org/10.1017/S0007114507787421>
- 723 Frank-Wilson, A. W., Johnston, J. D., Olszynski, W. P., & Kontulainen, S. A.
724 (2015). Measurement of muscle and fat in postmenopausal women:
725 precision of previously reported pQCT imaging methods. *Bone*, 75, 49–54.
726 <https://doi.org/10.1016/J.BONE.2015.01.016>
- 727 Fraser, B. J., Blizzard, L., Cleland, V., Schmidt, M. D., Smith, K. J., Gall, S. L.,
728 Dwyer, T., Venn, A. J., & Magnussen, C. G. (2020). Factors associated
729 with muscular fitness phenotypes in Australian children: A cross-sectional
730 study. *Journal of Sports Sciences*, 38(1), 38–45.
731 <https://doi.org/10.1080/02640414.2019.1679575>
- 732 Gómez-Bruton, A., Gonzalez-Agüero, A., Casajús, J. A., & Rodríguez, G. V.
733 (2014). Swimming training repercussion on metabolic and structural bone
734 development; benefits of the incorporation of whole body vibration or
735 pilometric training; the RENACIMIENTO project. *Nutricion Hospitalaria*,
736 30(2), 399–409. <https://doi.org/10.3305/nh.2014.30.2.7603>
- 737 González-Agüero, A., Vicente-Rodríguez, G., Gómez-Cabello, A., & Casajús, J.
738 A. (2013). Cortical and trabecular bone at the radius and tibia in male and
739 female adolescents with Down syndrome: a peripheral quantitative
740 computed tomography (pQCT) study. *Osteoporosis International : A*
741 *Journal Established as Result of Cooperation between the European*
742 *Foundation for Osteoporosis and the National Osteoporosis Foundation of*
743 *the USA*, 24(3), 1035—1044. <https://doi.org/10.1007/s00198-012-2041-7>
- 744 Gracia-Marco, L., Moreno, L. A., Ruiz, J. R., Ortega, F. B., de Moraes, A. C. F.,
745 Gottrand, F., Roccaldo, R., Marcos, A., Gómez-Martínez, S., Dallongeville,
746 J., Kafatos, A., Molnar, D., Bueno, G., de Henauw, S., Widhalm, K., &
747 Wells, J. C. (2016). Body Composition Indices and Single and Clustered

- 748 Cardiovascular Disease Risk Factors in Adolescents: Providing Clinical-
749 Based Cut-Points. *Progress in Cardiovascular Diseases*, 58(5), 555–564.
750 <https://doi.org/10.1016/j.pcad.2015.11.002>
- 751 Guglielmi, G., Ponti, F., Agostini, M., Amadori, M., Battista, G., & Bazzocchi, A.
752 (2016). The role of DXA in sarcopenia. *Aging Clin Exp Res*, 28(6), 1047–
753 1060. <https://doi.org/10.1007/s40520-016-0589-3>
- 754 Hao, G., Pollock, N. K., Harris, R. A., Gutin, B., Su, S., & Wang, X. (2019).
755 Associations between muscle mass, physical activity and dietary behaviour
756 in adolescents. *Pediatric Obesity*, 14(3), 1–8.
757 <https://doi.org/10.1111/ijpo.12471>
- 758 Hayes, A. F. (2013). Mediation, moderation, and conditional process analysis.
759 Introduction to mediation, moderation, and conditional process analysis: A
760 regression-based approach. *New York: Guilford Publications*, 1–20.
- 761 Heelan, K. A., & Eisenmann, J. C. (2006). Physical activity, media time, and
762 body composition in young children. *Journal of Physical Activity and Health*,
763 3(2), 200–209. <https://doi.org/10.1123/jpah.3.2.200>
- 764 Henriksson, P., Cadenas-Sanchez, C., Leppänen, M. H., Nyström, C. D.,
765 Ortega, F. B., Pomeroy, J., Ruiz, J. R., & Löf, M. (2016). Associations of fat
766 mass and fat-free mass with physical fitness in 4-year-old children: Results
767 from the MINISTOP trial. *Nutrients*, 8(8), 1–11.
768 <https://doi.org/10.3390/nu8080473>
- 769 Heymsfield, S. B., Gonzalez, M. C., Lu, J., Jia, G., & Zheng, J. (2015). Skeletal
770 muscle mass and quality: Evolution of modern measurement concepts in
771 the context of sarcopenia. *Proceedings of the Nutrition Society*, 74(4), 355–
772 366. <https://doi.org/10.1017/S0029665115000129>
- 773 Huybrechts, I., Vereecken, C., De Bacquer, D., Vandevijvere, S., Van Oyen, H.,
774 Maes, L., Vanhauwaert, E., Temme, L., De Backer, G., & De Henauw, S.
775 (2010). Reproducibility and validity of a diet quality index for children
776 assessed using a FFQ. *British Journal of Nutrition*, 104(1), 135–144.
777 <https://doi.org/10.1017/S0007114510000231>
- 778 Iglesia, I., Intemann, T., De Miguel-Etayo, P., Pala, V., Hebestreit, A., Wolters,
779 M., Russo, P., Veidebaum, T., Papoutsou, S., Nagy, P., Eiben, G., Rise, P.,

- 780 De Henauw, S., & Moreno, L. A. (2020). Dairy consumption at snack meal
781 occasions and the overall quality of diet during childhood. Prospective and
782 cross-sectional analyses from the idefics/i.family cohort. *Nutrients*, *12*(3),
783 1–20. <https://doi.org/10.3390/nu12030642>
- 784 Isganaitis, E. (2019). Developmental Programming of Body Composition:
785 Update on Evidence and Mechanisms. *Current Diabetes Reports*, *19*(8), 1–
786 20. <https://doi.org/10.1007/s11892-019-1170-1>
- 787 Jiménez-Pavón, D., Fernández-Vázquez, A., Alexy, U., Pedrero, R., Cuenca-
788 García, M., Polito, A., Vanhelst, J., Manios, Y., Kafatos, A., Molnar, D.,
789 Sjöström, M., & Moreno, L. A. (2013). Association of objectively measured
790 physical activity with body components in European adolescents. *BMC*
791 *Public Health*, *13*(1). <https://doi.org/10.1186/1471-2458-13-667>
- 792 Jürimäe, T., Hurbo, T., & Jürimäe, J. (2009). Relationship of handgrip strength
793 with anthropometric and body composition variables in prepubertal
794 children. *HOMO- Journal of Comparative Human Biology*, *60*(3), 225–238.
795 <https://doi.org/10.1016/j.jchb.2008.05.004>
- 796 Kim, J. H., & Park, Y. S. (2016). Low muscle mass is associated with metabolic
797 syndrome in Korean adolescents: the Korea National Health and Nutrition
798 Examination Survey 2009-2011. *Nutrition Research*, *36*(12), 1423–1428.
799 <https://doi.org/10.1016/j.nutres.2016.09.013>
- 800 Kim, K., Hong, S., & Kim, E. Y. (2016). Reference values of skeletal muscle
801 mass for Korean children and adolescents using data from the Korean
802 national health and nutrition examination survey 2009-2011. *PLoS ONE*,
803 *11*(4), 1–10. <https://doi.org/10.1371/journal.pone.0153383>
- 804 Kim, S., & Valdez, R. (2015). Metabolic risk factors in U.S. youth with low
805 relative muscle mass. *Obesity Research and Clinical Practice*, *9*(2), 125–
806 132. <https://doi.org/10.1016/j.orcp.2014.05.002>
- 807 Kulkarni, B., Hills, A. P., & Byrne, N. M. (2014). Nutritional influences over the
808 life course on lean body mass of individuals in developing countries.
809 *Nutrition Reviews*, *72*(3), 190–204. <https://doi.org/10.1111/nure.12097>
- 810 Kuriyan, R. (2018). Body composition techniques. *Indian Journal of Medical*
811 *Research*, *148*(5), 648–658. https://doi.org/10.4103/ijmr.IJMR_1777_18

812 Labayen, I., Moreno, L. A., Blay, M. G., Blay, V. A., Mesana, M. I., González-
813 Gross, M., Bueno, G., Sarría, A., & Bueno, M. (2006). Early programming
814 of body composition and fat distribution in adolescents. *Journal of Nutrition*,
815 136(1), 147–152. <https://doi.org/10.1093/jn/136.1.147>

816 Larqué, E., Labayen, I., Flodmark, C. E., Lissau, I., Czernin, S., Moreno, L. A.,
817 Pietrobelli, A., & Widhalm, K. (2019). From conception to infancy — early
818 risk factors for childhood obesity. *Nature Reviews Endocrinology*, 15(8),
819 456–478. <https://doi.org/10.1038/s41574-019-0219-1>

820 Laskey, M. A. (1996). Dual-energy X-ray absorptiometry and body composition.
821 *Nutrition*, 12(1), 45–51.
822 <https://doi.org/10.1097/01.mco.0000165010.31826.3d>

823 Leppänen, M., Henriksson, P., Delisle Nyström, C., Henriksson, H., Ortega, F.
824 B., Pomeroy, J., Ruiz, J. R., Cadenas-Sanchez, C., & Löf, M. (2017).
825 Longitudinal physical activity, body composition, and physical fitness in
826 preschoolers. In *Medicine and Science in Sports and Exercise* (Vol. 49,
827 Issue 10). <https://doi.org/10.1249/MSS.0000000000001313>

828 Leppänen, M., Nyström, C., Henriksson, P., Pomeroy, J., Ruiz, J. R., Ortega, F.,
829 Cadenas-Sánchez, C., & Löf, M. (2016). Physical activity intensity,
830 sedentary behavior, body composition and physical fitness in 4-year-old
831 children: Results from the ministop trial. *International Journal of Obesity*,
832 40(7), 1126–1133. <https://doi.org/10.1038/ijo.2016.54>

833 Liu, J., Yan, Y., Xi, B., Huang, G., & Mi, J. (2019). Skeletal muscle reference for
834 Chinese children and adolescents. *Journal of Cachexia, Sarcopenia and*
835 *Muscle*, 10(1), 155–164. <https://doi.org/10.1002/jcsm.12361>

836 López-Gil, J. F., Brazo-Sayavera, J., García-Hermoso, A., & Yuste Lucas, J. L.
837 (2020). Adherence to Mediterranean Diet Related with Physical Fitness and
838 Physical Activity in Schoolchildren Aged 6–13. *Nutrients*, 12(2), 567.
839 <https://doi.org/10.3390/nu12020567>

840 Moliner-Urdiales, D., Ortega, F. B., Vicente-Rodriguez, G., Rey-Lopez, J. P.,
841 Gracia-Marco, L., Widhalm, K., Sjöström, M., Moreno, L. A., Castillo, M. J.,
842 & Ruiz, J. R. (2010a). Association of physical activity with muscular
843 strength and fat-free mass in adolescents: The HELENA study. *European*

844 *Journal of Applied Physiology*, 109(6), 1119–1127.
845 <https://doi.org/10.1007/s00421-010-1457-z>

846 Moliner-Urdiales, D., Ortega, F. B., Vicente-Rodriguez, G., Rey-Lopez, J. P.,
847 Gracia-Marco, L., Widhalm, K., Sjöström, M., Moreno, L. A., Castillo, M. J.,
848 & Ruiz, J. R. (2010b). Association of physical activity with muscular
849 strength and fat-free mass in adolescents: The HELENA study. *European*
850 *Journal of Applied Physiology*, 109(6), 1119–1127.
851 <https://doi.org/10.1007/s00421-010-1457-z>

852 Mouratidou, T., Mesana Graffe, M. I., Huybrechts, I., De Decker, E., De
853 Craemer, M., Androutsos, O., Manios, Y., Galcheva, S., Lateva, M.,
854 Gurzkowska, B., Kułaga, Z., Birnbaum, J., Koletzko, B., & Moreno, L. A.
855 (2019). Reproducibility and relative validity of a semiquantitative food
856 frequency questionnaire in European preschoolers: The ToyBox study.
857 *Nutrition*, 65, 60–67. <https://doi.org/10.1016/j.nut.2019.03.003>

858 National Health and Nutrition Examination Survey (NHANES). (2012). *Body*
859 *Composition Procedures Manual*. July, 1–125.

860 Orsso, C. E., Tibaes, J. R. B., Oliveira, C. L. P., Rubin, D. A., Field, C. J.,
861 Heymsfield, S. B., Prado, C. M., & Haqq, A. M. (2019). Low muscle mass
862 and strength in pediatrics patients: Why should we care? *Clinical Nutrition*,
863 38(5), 2002–2015. <https://doi.org/10.1016/j.clnu.2019.04.012>

864 Ortega, F. B., Cadenas-Sánchez, C., Sánchez-Delgado, G., Mora-González, J.,
865 Martínez-Téllez, B., Artero, E. G., Castro-Piñero, J., Labayen, I., Chillón,
866 P., Löf, M., & Ruiz, J. R. (2015). Systematic Review and Proposal of a
867 Field-Based Physical Fitness-Test Battery in Preschool Children: The
868 PREFIT Battery. *Sports Medicine*, 45(4), 533–555.
869 <https://doi.org/10.1007/s40279-014-0281-8>

870 Poitras, V. J., Gray, C. E., Borghese, M. M., Carson, V., Chaput, J.-P., Janssen,
871 I., Katzmarzyk, P. T., Pate, R. R., Connor Gorber, S., & Kho, M. E. (2016).
872 Systematic review of the relationships between objectively measured
873 physical activity and health indicators in school-aged children and youth.
874 *Applied Physiology, Nutrition, and Metabolism*, 41(6), S197–S239.
875 <https://doi.org/10.1139/apnm-2015-0663>

- 876 Ramires, V. V., Dumith, S. C., Wehrmeister, F. C., Hallal, P. C., Menezes, A. M.
877 B., & Gonçalves, H. (2016). Physical activity throughout adolescence and
878 body composition at 18 years: 1993 Pelotas (Brazil) birth cohort study.
879 *International Journal of Behavioral Nutrition and Physical Activity*, 13(1), 1–
880 13. <https://doi.org/10.1186/s12966-016-0430-6>
- 881 Rennie, K. L., Livingstone, M. B. E., Wells, J. C. K., Mcgloin, A., Coward, W. A.,
882 & Prentice, A. M. (2005). *Association of physical activity with body-*
883 *composition indexes in children aged 6 – 8 y at varied risk of obesity 1 – 3.*
884 13–20. <https://doi.org/https://doi.org/10.1093/ajcn/82.1.13>
- 885 Riddoch, C. J., Andersen, L. B., Wedderkopp, N., Harro, M., Klasson-Heggebø,
886 L., Sardinha, L. B., Cooper, A. R., & Ekelund, U. (2004). Physical Activity
887 Levels and Patterns of 9- and 15-yr-Old European Children. *Medicine and*
888 *Science in Sports and Exercise*, 36(1), 86–92.
889 <https://doi.org/10.1249/01.MSS.0000106174.43932.92>
- 890 Roggen, I., Roelants, M., Sioen, I., Vandewalle, S., De Henauw, S., Goemaere,
891 S., Kaufman, J.-M., & De Schepper, J. (2015). Pediatric Reference Values
892 for Tibial Trabecular Bone Mineral Density and Bone Geometry Parameters
893 Using Peripheral Quantitative Computed Tomography. *Calcified Tissue*
894 *International*, 96(6), 527–533. <https://doi.org/10.1007/s00223-015-9988-2>
- 895 Ruiz, J. R., Castro-Piñero, J., Artero, E. G., Ortega, F. B., Sjöström, M., Suni, J.,
896 & Castillo, M. J. (2009). Predictive validity of health-related fitness in youth:
897 a systematic review. *British Journal of Sports Medicine*, 43(12), 909 LP –
898 923. <https://doi.org/10.1136/bjism.2008.056499>
- 899 Santos, D. A., Magalhaes, J. P., Judice, P. B., Correia, I. R., Minderico, C. S.,
900 Ekelund, U., & Sardinha, L. B. (2019). Fitness Mediates Activity and
901 Sedentary Patterns Associations with Adiposity in Youth. *Medicine &*
902 *Science in Sports & Exercise*, 51(2), 323–329.
903 <https://doi.org/10.1249/MSS.0000000000001785>
- 904 Scheurer, J. M., Zhang, L., Plummer, E. A., Hultgren, S. A., Demerath, E. W., &
905 Ramel, S. E. (2018). Body Composition Changes from Infancy to 4 Years
906 and Associations with Early Childhood Cognition in Preterm and Full-Term
907 Children. *Neonatology*, 114(2), 169–176.

908 <https://doi.org/10.1159/000487915>

909 Singhal, A., Wells, J., Cole, T. J., Fewtrell, M., & Lucas, A. (2003). Programming
910 of lean body mass: A link between birth weight, obesity, and cardiovascular
911 disease? *American Journal of Clinical Nutrition*, *77*(3), 726–730.
912 <https://doi.org/10.1093/ajcn/77.3.726>

913 Sioen, I., Lust, E., De Henauw, S., Moreno, L. A., & Jiménez-Pavón, D. (2016).
914 Associations Between Body Composition and Bone Health in Children and
915 Adolescents: A Systematic Review. *Calcified Tissue International*, *99*(6),
916 557–577. <https://doi.org/10.1007/s00223-016-0183-x>

917 Statistics, U. I. for. (2012). *International standard classification of education:*
918 *ISCED 2011*. UNESCO Institute for Statistics Montreal.

919 Tanita Corporation - Japan. (2002). *Body Composition Analyzer BC-418*
920 *Instruction Manual*. 1–43.

921 Tremblay, M. S., Warburton, D. E. R., Janssen, I., Paterson, D. H., Latimer, A.
922 E., Rhodes, R. E., Kho, M. E., Hicks, A., LeBlanc, A. G., Zehr, L.,
923 Murumets, K., & Duggan, M. (2011). New Canadian physical activity
924 guidelines. *Applied Physiology, Nutrition and Metabolism*, *36*(1), 36–46.
925 <https://doi.org/10.1139/H11-009>

926 Trost, S. G., Pate, R. R., Sallis, J. F., Freedson, P. S., Taylor, W. C., Dowda,
927 M., & Sirard, J. (2002). Age and gender differences in objectively measured
928 physical activity in youth. *Medicine and Science in Sports and Exercise*,
929 *34*(2), 350–355. <https://doi.org/10.1097/00005768-200202000-00025>

930 Unesco. (1997). *International Standard Classification of Education-ISCED*
931 *1997: November 1997*. Unesco.

932 Veldhuis, J. D., Roemmich, J. N., Richmond, E. J., Rogol, A. D., Lovejoy, J. C.,
933 Sheffield-Moore, M., Mauras, N., & Bowers, C. Y. (2005). Endocrine control
934 of body composition in infancy, childhood, and puberty. *Endocrine*
935 *Reviews*, *26*(1), 114–146. <https://doi.org/10.1210/er.2003-0038>

936 Vicente-Rodriguez, G., Ara, I., Perez-Gomez, J., Serrano-Sanchez, J. A.,
937 Dorado, C., & Calbet, J. A. L. (2004). High Femoral Bone Mineral Density
938 Accretion in Prepubertal Soccer Players. *Medicine & Science in Sports &*

939 *Exercise*, 36(10). <https://journals.lww.com/acsm->
940 [msse/Fulltext/2004/10000/High_Femoral_Bone_Mineral_Density_Accretion](https://journals.lww.com/acsm-)
941 [_in.19.aspx](https://journals.lww.com/acsm-)

942 Vyncke, K., Cruz Fernandez, E., Fajó-Pascual, M., Cuenca-García, M., De
943 Keyzer, W., Gonzalez-Gross, M., Moreno, L. A., Beghin, L., Breidenassel,
944 C., Kersting, M., Albers, U., Diethelm, K., Mouratidou, T., Grammatikaki, E.,
945 De Vriendt, T., Marcos, A., Bammann, K., Börnhorst, C., Leclercq, C., ...
946 Huybrechts, I. (2013). Validation of the Diet Quality Index for Adolescents
947 by comparison with biomarkers, nutrient and food intakes: The HELENA
948 study. *British Journal of Nutrition*, 109(11), 2067–2078.
949 <https://doi.org/10.1017/S000711451200414X>

950 Wells, J. C. K. (2000). A Hattori chart analysis of body mass index in infants and
951 children. *International Journal of Obesity*, 24(3), 325–329.
952 <https://doi.org/10.1038/sj.ijo.0801132>

953 Wells, J. C. K., Cole, T. J., & team, A. study. (2002). Adjustment of fat-free
954 mass and fat mass for height in children aged 8 y. *International Journal Of*
955 *Obesity*, 26, 947. <https://doi.org/10.1038/sj.ijo.0802027>

956 Westerterp, K. R., Yamada, Y., Sagayama, H., Ainslie, P. N., Andersen, L. F., &
957 Anderson, L. J. (2021). *Physical activity and fat-free mass during growth*
958 *and in later life*. 1–7.

959 Wickel, E. E., & Eisenmann, J. C. (2007). Contribution of youth sport to total
960 daily physical activity among 6- to 12-yr-old boys. *Medicine and Science in*
961 *Sports and Exercise*, 39(9), 1493–1500.
962 <https://doi.org/10.1249/mss.0b013e318093f56a>

963 World Health Organization. (2006). *WHO child growth standards: length/height-*
964 *for-age, weight-for-age, weight-for-length, weight-for-height and body mass*
965 *index-for-age: methods and development*.

966 World Health Organization. (2020). WHO Guidelines on physical activity and
967 sedentary behaviour. In *World Health Organization*.
968 <https://apps.who.int/iris/bitstream/handle/10665/325147/WHO-NMH-PND->
969 [2019.4-](https://apps.who.int/iris/bitstream/handle/10665/325147/WHO-NMH-PND-)
970 [eng.pdf?sequence=1&isAllowed=y%0Ahttp://www.who.int/iris/handle/1066](https://apps.who.int/iris/bitstream/handle/10665/325147/WHO-NMH-PND-2019.4-eng.pdf?sequence=1&isAllowed=y%0Ahttp://www.who.int/iris/handle/1066)

971 5/311664%0Ahttps://apps.who.int/iris/handle/10665/325147
972 Wyszyńska, J., Matłosz, P., Szybisty, A., Lenik, P., Dereń, K., Mazur, A., &
973 Herbert, J. (2020). Obesity and Body Composition in Preschool Children
974 with Different Levels of Actigraphy-Derived Physical Activity—A Cross-
975 Sectional Study. *Journal of Clinical Medicine*, 9(4), 1210.
976 <https://doi.org/10.3390/jcm9041210>
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978

Table 1. Main characteristics of the participating children (n= 279).

Characteristics	All n= 279 (M±SD)	Girls n= 129 (M±SD)	Boys n= 150 (M±SD)	P-value
Age (y)	7.5 ± 0.3	7.6 ± 0.4	7.5 ± 0.3	0.41
Anthropometric measurements				
Height (cm)	126.1 ± 5.9	125.6 ± 5.6	125.5 ± 6.0	0.12
Weight (kg) ^a	26.4 (23.6 - 30.0)	26.2 (23.6 - 30.0)	26.7 (23.7 - 30.8)	0.59
BMI (kg/m ²) ^a	16.6 (15.4 - 18.3)	16.7 (15.5 - 18.3)	16.6 (15.4 - 18.5)	0.74
BMI z-score [†]	0.84 ± 1.2	0.76 ± 1.1	0.87 ± 1.3	0.48
BIA				
FFM (kg) ^a	20.5 (18.7 - 22.5)	19.9 (18.5 - 21.8)	20.8 (19.1 - 23.4)	<0.01*
FFMI (kg/m ²)	13.1 ± 1.4	12.7 (12.1 - 13.4)	13.2 (12.4 - 14.2)	<0.01*
DXA				
TLSTM (kg) ^a	18.1 (16.6 – 20.0)	17.5 (16.2 - 18.7)	18.9 (17.0 - 21.0)	<0.01*
TLSTMI (kg/m ²)	11.6 ± 1.1	11.2 ± 1.0	11.9 ± 1.1	<0.01*
ALSTMI (Kg/m ²)	4.7± 0.5	4.5± 0.5	4.8± 0.5	<0.01*
pQCT				
Tibia length (mm)	274.3 ± 18.1	275.8 ±17.3	273.1 ±18.8	0.14

MCSA (mm ²)	3275.3 ± 496.0	3136.5 ± 473.2	3389.1 ± 488.3	<0.01*
MCSAI (mm ² /m ²)	2057.1 ± 263.1	1996.3 ± 257.6	2112.1 ± 256.5	<0.01*
Physical activity				
Light (min/day)	41.3 ± 11.1	37.0 ± 9.9	45.0 ± 10.8	<0.01*
Moderate (min/day) ^a	29.2 (21.1 - 36.2)	24.3 (18.1 - 32.1)	31.9 (24.9 - 41.1)	<0.01*
Vigorous (min/day) ^a	69.0 (55.6 - 85.0)	59.8 (50.0 - 75.7)	77.5 (64.2 - 92.6)	<0.01*
MVPA (min/day) ^a	97.3 (77.4 - 121.4)	85.9 (67.8 - 108.4)	110.8 (88.0 - 134.0)	<0.01*
Total PA (min/day)	269.6 (240.3 - 299.7)	254.0 (231.3 - 285.2)	282.5 (245.9 - 312.7)	<0.01*
Physical fitness				
Handgrip strength (kg)	10.5 ± 2.2	10.1 ± 2.0	10.9 ± 2.1	<0.01*
Standing long jump (cm)	102.7 ± 17.8	98.9 ± 17.0	107.6 ± 17.7	<0.01*

Abbreviation: ALSTMI= Appendicular lean soft tissue mass index; BIA= Bioelectrical impedance; BMI= Body mass index; MCSA= Muscle cross-sectional area; DXA= Dual-energy X-ray absorptiometry; FFM= Fat-free mass; FFMI= Fat-free mass index; MVPA= Moderate-vigorous physical activity; pQCT= quantitative peripheral computed tomography; TLSTM= Total lean soft tissue mass; TLSTMI= Total lean soft tissue mass index

Normally distributed variables are shown as mean ± SD (Student t-test)

^a Non-normally distributed variables are shown as median and interquartile intervals (25th and 75th, U Mann–Whitney)

†BMI z-scores were calculated according to the World Health Organization (WHO)

* Significant differences by gender

Significance was set at 0.05 level.

Table 2. Associations between physical activity and total lean soft tissue mass index, muscle cross-sectional area index, and fat-free mass index (n= 279)

Predictors Physical activity	Girls										Boys										
	Total		Light		Moderate		Vigorous		MVPA		Total		Light		Moderate		Vigorous		MVPA		
	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	
TLSTMI by DXA																					
<i>Model 1</i>	.272	.004*	.154	.109	.123	.201	.151	.117	.144	.137	.335	.000*	.287	.001*	.093	.289	.203	.020*	.166	.058	
<i>Model 2</i>	.275	.004*	.170	.078	.120	.219	.159	.001*	.148	.130	.371	.000*	.302	.001*	.116	.206	.224	.013*	.189	.038*	
<i>Model 3</i>	.247	.010*	.151	.111	.111	.248	.143	.001*	.134	.162	.337	.000*	.290	.001*	.081	.367	.200	.023*	.160	.072	
MCSAI by pQCT																					
<i>Model 1</i>	.148	.131	.112	.253	-.003	.974	.057	.564	.036	.717	.091	.314	.074	.410	.052	.560	.068	.447	.064	.480	
<i>Model 2</i>	.124	.223	.093	.359	-.034	.742	.030	.768	.007	.944	.114	.219	.083	.364	.053	.569	.074	.426	.068	.468	
<i>Model 3</i>	.097	.347	.077	.448	-.040	.692	.018	.863	-.004	.972	.110	.245	.081	.381	.049	.607	.070	.454	.063	.500	
FFMI by BIA																					
<i>Model 1</i>	.125	.194	.132	.173	-.047	.626	.042	.664	.010	.918	.260	.003*	.161	.064	-.010	.908	.078	.373	.047	.594	
<i>Model 2</i>	.185	.053	.149	.122	-.060	.539	.045	.642	.007	.939	.308	.000*	.175	.050	-.026	.781	.078	.389	.042	.648	
<i>Model 3</i>	.149	.097	.137	.154	-.066	.496	.035	.717	-.001	.988	.299	.001*	.175	.051	-.026	.780	.079	.388	.042	.647	

β : standardized regression coefficient

Abbreviation: BIA= Bioimpedance analysis; DXA= Dual-energy X-ray absorptiometry; FFMI= Fat-free mass index; MCSAI= Muscle cross-sectional area index; pQCT= Peripheral quantitative computed tomography; TLSTMI= Total lean soft tissue mass index.

Model 1 Physical activity (basic model without adjustments)

Model 2 *Model 1* + diet quality index, parents' highest education level and age.

Model 3 *Model 2* + birth weight

*Significance was set at 0.05 level.

Table 3. Associations between physical fitness and total lean soft tissue mass index, muscle cross-sectional area index, and fat-free mass index (n= 279)

Predictors Physical fitness	Girls				Boys				
	Handgrip strength		Standing long jump		Handgrip strength		Standing long jump		
	β	p	β	p	β	p	β	p	
TLSTMI by DXA									
<i>Model 1</i>	.363	.000*	.055	.512	.478	.000*	-.019	.839	
<i>Model 2</i>	.350	.000*	.070	.403	.464	.000*	-.013	.893	
<i>Model 3</i>	.314	.000*	.034	.672	.425	.000*	-.036	.690	
MCSAI by pQCT									
<i>Model 1</i>	.262	.002*	.056	.520	.273	.000*	-.064	.508	
<i>Model 2</i>	.282	.001*	.074	.404	.287	.000*	-.060	.540	
<i>Model 3</i>	.267	.002*	.051	.561	.301	.000*	-.059	.547	
FFMI by BIA									
<i>Model 1</i>	.236	.004*	-.082	.332	.289	.000*	-.084	.367	
<i>Model 2</i>	.218	.008*	-.065	.430	.307	.000*	-.083	.367	
<i>Model 3</i>	.196	.017*	-.088	.281	.325	.000*	-.082	.384	

β : standardized regression coefficient

Abbreviation: BIA= Bioimpedance analysis, DXA= Dual-energy X-ray absorptiometry, FFMI= Fat-free mass index, MCSAI= Muscle cross-sectional area index, pQCT= Peripheral quantitative computed tomography and TLSTMI= Total lean soft tissue mass index.

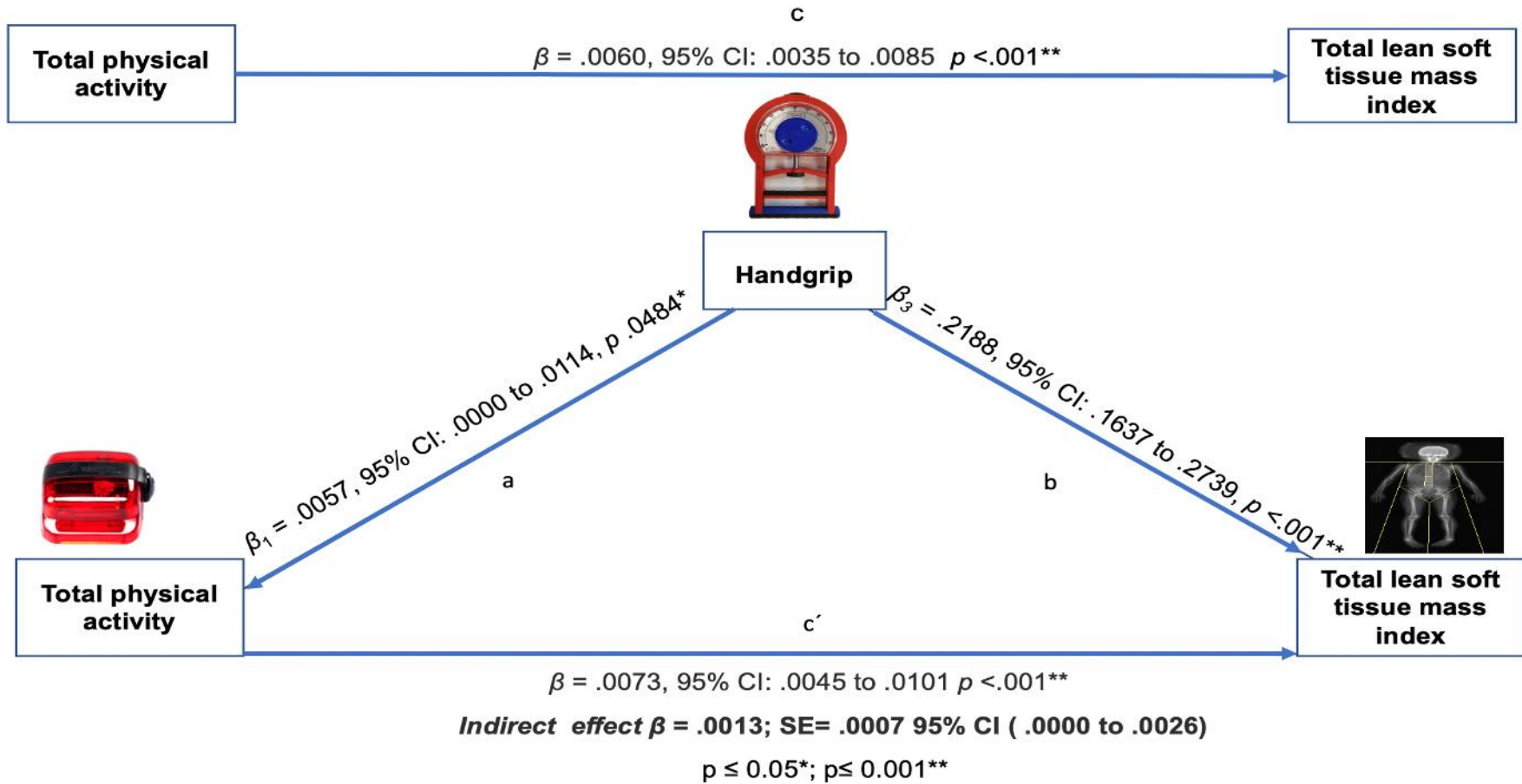
Model 1 physical fitness (basic model without adjustments)

Model 2 *Model 1* + diet quality index, parents' highest education level and age.

Model 3 *Model 2* + birth weight

*Significance was set at 0.05 level.

Figure 1. Serial multiple mediation model of the association between total physical activity and total lean soft tissue mass index, using handgrip strength as mediator, controlling for gender.



1 **Associations between Spanish children's physical activity and physical**
2 **fitness with lean body mass: The CALINA Study**

3
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39

40

41 **Abstract**

42
43 The aim of the present study is to investigate the associations between physical
44 activity (PA) and physical fitness (PF) with lean body mass (LBM) and to assess
45 whether PA mediates the association between PF and LBM. 279 children (150
46 boys) aged 7.5 ± 0.3 years participated in the study. PA was assessed by
47 accelerometry and PF with handgrip and the standing long jump test. Total lean
48 soft tissue mass index (TLSTMI), muscle cross-sectional area index (MCSAI),
49 and fat-free mass index (FFMI) were evaluated using dual-energy X-ray
50 absorptiometry, peripheral quantitative computed
51 tomography, and bioimpedance analysis, respectively.

52
53 Total ($\beta=0.247$) and vigorous PA ($\beta=0.143$) were associated with TLSTMI in
54 girls. In boys, total ($\beta=0.337$), light ($\beta=0.290$), vigorous ($\beta=0.200$), and
55 moderate-vigorous PA ($\beta=0.189$) were associated with TLSTMI. Only total PA
56 was associated with FFMI ($\beta=0.299$). Handgrip strength does not mediate the
57 relationship between total PA and TLSTMI. Positive associations were found
58 between handgrip strength and TLSTMI, MCSAI, and FFMI in both girls and
59 boys.

60
61 In children, there is a positive association between total and vigorous PA with
62 TLSTMI. Handgrip strength does not mediate the relationship between total PA
63 and TLSTMI. It was associated with TLSTMI, MCSAI, and FFMI.

64 **Key words:** lean body mass, muscle cross-sectional area, physical activity,
65 physical fitness, schoolchildren

66 67 **1. Introduction**

68
69 Lean body mass (LBM) is mainly constituted by muscle mass, internal organ
70 non-adipose components, and extracellular fluid (Kuriyan, 2018). In recent
71 years, LBM has been considered to play an essential role in growth
72 maintenance, normal development, and systemic glucose metabolism in

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73 children (Liu et al., 2019). It has also been associated with the risk of
74 cardiovascular disease (S. Kim & Valdez, 2015), affecting bone health (bone
75 mineral density and structure in both sexes during childhood) (Dorsey et al.,
76 2010; Sioen et al., 2016), and cognitive development (Scheurer et al., 2018),
77 among others. Studies on children and adolescents with low lean mass showed
78 a higher cardiometabolic risk (S. Kim & Valdez, 2015), related to significantly
79 higher waist circumference, blood pressure, triglycerides, and total
80 cholesterol/high-density lipoprotein cholesterol values (Burrows et al., 2017;
81 Luis Gracia-Marco et al., 2016). Other studies have shown an increased risk of
82 metabolic syndrome (Burrows et al., 2017; J. H. Kim & Park, 2016; K. Kim et al.,
83 2016). In this regard, the literature revealed that those presenting a phenotype
84 combining low lean mass and obesity had the most unfavorable
85 cardiometabolic risk profile (Burrows et al., 2017). Therefore, low levels of lean
86 mass in children and adolescents may represent a public health problem and a
87 burden on the health system for future stages in life.

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89 Currently, it is accepted that several factors can influence the development of
90 lean mass/muscle mass throughout the life cycle, including fetal programming
91 (Isganaitis, 2019; Labayen et al., 2006; Larqué et al., 2019), early nutritional
92 status (Singhal et al., 2003), age, gender (Wells, 2000), hormones (Veldhuis et
93 al., 2005), diet, and physical activity and exercise (Kulkarni et al., 2014;
94 Westerterp et al., 2021)

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96 Physical activity and exercise play an essential role in the development of both
97 the size of muscle fibers and the recruitment of motor units, thus, developing
98 strength (Dotan et al., 2012) and metabolic adaptations (Boisseau &
99 Delamarche, 2000).

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101 A cross-sectional study, in preschool-age children between 5 and 6, showed
102 that a level of MVPA below the World Health Organization recommendation was
103 significantly associated with a lower content of muscle mass and FFM (0.8%
104 and 1.19%, respectively); these differences were more evident in boys than in
105 girls (Wyszyńska et al., 2020).

107 A longitudinal study (Leppänen et al., 2016, 2017) in 4.5-year-old children
108 demonstrated that children with higher and moderate to vigorous physical
109 activity (MVPA) levels had a higher fat-free mass index (FFMI) after a 12-month
110 follow-up. Similar results have been reported in preschool children (Henriksson
111 et al., 2016) and adolescents (Baxter-Jones et al., 2008; Ramires et al., 2016).

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114 The relationship between physical activity levels, physical fitness, and body
115 composition with fat mass has been widely studied in children and adolescents
116 (Henriksson et al., 2016; Santos et al., 2019). Regarding its relationship with
117 LBM, Baxter-Jones et al. (Baxter-Jones et al., 2008) reported that daily physical
118 activity had a significant independent influence on LBM development during
119 adolescence (after controlling for biological maturity and stature). In addition,
120 studies in children have shown that higher fat-free mass (FFM) values are
121 associated with better cardiorespiratory fitness, improved upper and lower body
122 muscle strength (Fraser et al., 2020), motor fitness (Henriksson et al., 2016),
123 and a healthier cardiovascular profile later in life, with a lower risk of premature
124 death (Ruiz et al., 2009). Thus, it seems reasonable to hypothesize that
125 physical activity or physical fitness might play a crucial role in the association
126 between fitness-lean or physical activity-lean in this age group.

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128 Given the available evidence, it is apparent that studies examining the
129 relationship between physical activity levels, physical fitness, and body
130 composition, specifically LBM in children between 6 and 12 years of age, are
131 limited (Hao et al., 2019). Furthermore, most of the available studies have used
132 statistical methods such as analysis of covariance, multiple linear regression, or
133 logistic regression to adjust for confounding factors, statistical methods that
134 cannot distinguish the effect of mediating variables.

135 Furthermore, there are important differences between DXA, pQCT, and BIA. On
136 the one hand, DXA is an accurate device for assessing body composition of the
137 whole body and regions/segments (Laskey, 1996). At the same time, pQCT
138 allows assessment at the appendicular level (limb muscle cross-sectional
139 area)(Frank-Wilson et al., 2015). Still, they are not available to everyone due to
140 their high price and radiation exposure (Guglielmi et al., 2016). On the other

141 hand, BIA is an effective alternative to assess body composition of the whole
142 body, low price in the absence of radiation (Orsso et al., 2019). Therefore, it will
143 be important to use all three devices to assess LBM. In addition, the
144 normalization of body size should also be considered, since most of the
145 evidence available to date uses absolute or relative measures of LBM (kg
146 and%), which makes it difficult to make comparisons between individuals or
147 populations (Wells et al., 2002).

148
149 Therefore, in this study, we hypothesized that children with higher physical
150 activity or physical fitness would have higher LBM values, and physical activity
151 or physical fitness would be a mediator between the fitness-lean or physical
152 activity-lean associations. Its purposes are to (1) investigate the associations
153 between objectively assessed physical activity, physical fitness, and LBM
154 measured by dual-energy X-ray absorptiometry (DXA), bioimpedance analysis
155 (BIA), and peripheral quantitative computed tomography (pQCT); and (2)
156 evaluate whether these associations were mediated by total physical activity or
157 physical fitness or with handgrip strength in a Spanish cohort of children.

158 159 **2. Material and Methods**

160 161 *2.1 Study participants*

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163 This study's participants were involved in the CALINA study (Growth and
164 Feeding during Breastfeeding and Early Childhood in Children from Aragón).
165 This longitudinal observational study evaluated a representative cohort of
166 children born in Aragon (Spain) between 2009-2010. The initial sample
167 consisted of 1602 newborns (Oves Suárez et al., 2014), followed every month
168 during the first year and then every year until they turned 6.

169
170 In 2016 and 2017, the recruited families (n=952) from the baseline examination
171 were invited to participate in this follow-up study in Zaragoza, the biggest city in
172 Aragón (Spain). They were invited to participate in an additional body
173 composition assessment in our laboratory at the University of Zaragoza. From
174 the 415 children who participated in this follow-up assessment, 136 were

175 excluded from the study for the following reasons: lack of data on body
176 composition and physical fitness (n=90), or lack of valid accelerometer data
177 (n=46). Finally, 279 children (150 boys and 129 girls), between 6 and 8 years,
178 with complete DXA, pQCT, BIA, physical activity, and physical fitness
179 examination data were included in this study.

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181 *2.2 Ethics statement*

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183 This study was conducted following the Declaration of Helsinki (Fortaleza 2013
184 review) ethical guidelines. In 2009, it was approved by the Ethics Committee in
185 Clinical Research of the Government of Aragon (ref. PI ICS108/0088, Spain). In
186 2016, it was again approved by the same committee for the follow-up presented
187 in this manuscript (Ref. CPPI13/00105, Spain).

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189 All participants were evaluated after parents' signed the informed consent and
190 the verbal assent of the children.

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192 *2.3 Body composition*

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194 *2.3.1 Weight, height, and bioimpedance analysis (BIA):*

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196 The weight and bioelectrical impedance analysis were measured using a
197 TANITA BC 418 MA electronic scale (Tanita Europe BV, Amsterdam,
198 Netherlands) with a 0.1 kg precision and 0-200 kg range. Following the
199 manufacturer's instructions (Tanita Corporation - Japan, 2002), the children
200 were asked to stand barefoot and with as little clothing as possible on the
201 weighing platform, touching the electrodes, in a stable position, without bending
202 the knees, and with both hands grabbing the handles.

203

204 A SECA[®] 225 portable stadiometer (SECA[®] 225, Hamburg, Germany) with a
205 precision of 0.1 cm and a range of 70-220 cm was used to determine the
206 height. The body mass index (BMI) was calculated as the weight divided by the
207 squared height (kg/m²). The AnthroPlus software from the World Health
208 Organization (WHO) (World Health Organization, 2006) was used to calculate

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209 BMI age and gender-specific z-scores. The FFMI was calculated relating the
210 FFM in kilograms divided by the squared height in meters.

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212 2.3.2 Dual-energy X-ray absorptiometry (DXA):

213 Dual-energy X-ray absorptiometry was used to determine the LBM (kg), using
214 the DXA QDR-Explorer™ 4500 (Hologic Inc., Bedford, Massachusetts, USA).
215 All measurements we performed following manufacturer's instructions (National
216 Health and Nutrition Examination Survey (NHANES), 2012). The participants
217 were measured lightly clothed and without metal objects or jewelry. An
218 examination of the whole body was performed. The participants were placed
219 supine, with arms along the body, without touching the trunk (Crabtree et al.,
220 2007). Analyzes and regions of interest (ROI) were determined using Pediatric
221 Hologic Corp. software version 12.4. The whole-body scan intra-measures
222 coefficient variation of LBM in our laboratory was 1.9; a result elsewhere
223 described (L Gracia-Marco et al., 2012).

224

225 This study used the sum value of the fat, lean, and bone masses (Lean soft
226 tissue mass LSTM) = LBM - bone mineral content (BMC). The total LSTM index
227 (TLSTMI) and the appendicular LSTM index (ALSTMI) were calculated by
228 dividing both the LSTM (Kg) and the appendicular LSTM (Kg) by the height
229 squared.

230

231 In this study, the DXA and BIA analyses were carried out between 4.30 pm and
232 6.30 pm, 2 -4 hours after the end of the previous meal.

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234 2.3.3 Peripheral quantitative computed tomography (pQCT):

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236 Stratec XCT 2000 L (Stratec Medizintechnik, Pforzheim, Germany) was used to
237 measure the muscle cross-sectional area (MCSA) at 66% of the total length of
238 the left tibia (González-Agüero et al., 2013), a method that has proven to be
239 valid for estimating the lean mass (Córdoba-Rodríguez et al., 2021).

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241 All the measurements were carried out after the calibration of the equipment.

242 The total length of the tibia was determined using a segmometer, measuring the

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243 distance from the cleft of the knee's medial joint to the tibia's (leg) Sphyrion
244 tibial medial malleolus (Roggen et al., 2015).

245 For the definition of the reference line in the distal tibia, an exploratory scout
246 view was performed, and the reference line was placed at the medial end of the
247 distal epiphysis. The intra-measures coefficient of variation for MCSA using
248 pQCT was 1.69, as previously reported (Gómez-Bruton et al., 2014).

249 The muscle cross-sectional area index (MCSAI) was calculated by dividing the
250 muscle cross-sectional area (MCSA) by the squared height.

251
252 The pQCT and DXA images were evaluated visually by a technician to identify
253 motion artifacts. The images showing movement were subjected to a new
254 exploration or excluded from the data analysis. Moreover, an external evaluator
255 reviewed all the images to endorse the image quality.

256 257 *2.4 Physical activity*

258 Physical activity was evaluated using the ActiGraph wGT3x-BT triaxial
259 accelerometer (ActiGraph, Ft. Walton Beach, USA). Parents/caregivers and
260 children were asked to place the accelerometer under the child's clothing at the
261 level of the right iliac crest, during their waking day, for seven consecutive days
262 and to complete a diary to assess the minutes and reasons why accelerometers
263 were not being used (Bammann et al., 2011).

264 Additionally, children and parents / caregivers were advised not to use
265 accelerometers in water activities.

266 All the accelerometers were programmed to record data every 15 seconds
267 (epoch). Twenty minutes or more of consecutive zero counts were defined as
268 unused time.

269 For the information processing, a day in which the child used the accelerometer
270 for at least 8 hours was considered a valid day. A minimum of 3 days was
271 required, including at least one weekend day, as was previously used by other
272 authors in similar population groups (Moliner-Urdiales et al., 2010a)

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274 Actilife software version 6.0 (ActiGraph, Pensacola, FL., USA) was used for
275 data processing. The cut-off points determined by Evenson were used as a
276 reference to define the time dedicated to sedentary and different intensity
277 physical activity (sedentary: 0–100, light: 101–2295, moderate: 2296–4011,
278 vigorous: 4012, and more accelerometer counts per minute [cpm]) (Evenson et
279 al., 2008).

280 The absolute amount of time in each intensity category determined from the
281 accelerometer data was used for the analyzes, taking as a reference that the
282 World Health Organization (WHO)(World Health Organization, 2020) frequently
283 uses the min/day metric to issue their physical activity recommendations.

284

285 Most of the children had complete actigraphic data of 7 days (n=126, 42.2%) or
286 6-5 days (n=118, 42.4%) of. The average follow-up included 6.2 days of
287 recordings.

288

289 *2.5 Upper limb strength - Handgrip strength test:*

290 A TKK-5401 (Takei Scientific Instruments Co., Ltd., Niigata, Japan) digital
291 handgrip dynamometer with a precision of 0.1kg and adjustable grip was used
292 according to the size of the hand derived from the following gender-specific
293 equations (España-Romero et al., 2008):

294 Boys: $Y = X/4 + 0.44$ cm

295 Girls: $Y = 0.3X - 0.52$ cm

296 Where Y = optimal grip and X = size of the wide-open hand, measured from the
297 tip of the thumb to the tip of the little finger.

298 During the measurement, participants stood, elbow extended, avoiding any
299 bodily contact with the dynamometer, except with the measured hand. The
300 children were asked to press as hard as possible for 3 to 5 seconds with each
301 hand; two attempts were made with each hand, three minutes of rest interval
302 between each of them. The dynamometer display was aligned to face the
303 examiner, providing blind measurements to the children. The final score was

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304 calculated as the average of the best attempt obtained for the left and right
305 hands in kg (De Miguel-Etayo et al., 2014).

306 *2.6 Lower limb strength. Standing long jump test*

307 The long jump test was used to assess the explosive strength of the lower
308 extremities. The children were asked to stand, feet slightly apart, in front of a
309 starting line. Then, from a standing position, a forward jump was made using
310 the arms' impulse, being careful not to step on the starting line and pushing with
311 both feet at the same time. The measurement was made from the impulse line
312 to the heel closest to the start line and recorded in cm (López-Gil et al., 2020).
313 Two attempts were made, recording the highest value obtained.

314 *2.7 Covariates*

315 We recorded the following variables for all the children:

316 *2.7.1 Parental education*

317 *Highest level of education:* the parents were asked to report their highest
318 achieved level of education when they came to our lab in 2016-2017 (no
319 studies; basic-primary studies; intermediate studies [including modules
320 vocational training and secondary studies]; higher studies; and university
321 degrees). These were later coded according to the International Standard
322 Classification of Education (ISCED-2011) (Statistics, 2012) and categorized
323 again into low (0-2), medium (3-4), and high (5-8) educational levels (Unesco,
324 1997).

325 *2.7.2 Anthropometric data of the child at birth:*

326 At the beginning of the CALINA study, in the first visit (15 days after birth), the
327 pediatric and nursing staff conducting the *Programa de Salud Infantil* (Child
328 Health Program) collected the birth weight variable.

329 *2.7.3 Food frequency questionnaire and diet quality index (DQI):*

330 A previously validated, semi-quantitative food frequency questionnaire was
331 used to assess the dietary intake (Mouratidou et al., 2019), including 37 foods
332 and beverages. The responding parents were asked to classify each food's
333 consumption frequency using a 6-Point Likert scale ranging from never to every
334 day (never/less than once a month, 1 to 3 times per month, 1 day/week, 2 to 4
335 days/week, 5 to 6 days/week, and every day) and serving size.

336 The DQI was subsequently calculated. This index mainly assesses the
337 components of dietary diversity, dietary quality, and dietary balance. Dietary
338 diversity evaluated the daily consumption of at least one food serving from the
339 eight recommended food groups. This component's score ranged from 0 to 9 for
340 at least one serving from a recommended food group. Diet quality expressed
341 whether the children made optimal decisions on the food quality within a food
342 group. Dietary balance was calculated from the difference between the food
343 intake's suitability and its excess in the diet (Huybrechts et al., 2010; Vyncke et
344 al., 2013). These three components of the DQI were presented in percentages.
345 The categories' scores were summed and divided by 3 to compute the overall
346 DQI, resulting in scores ranging from -33% to 100%. The components' mean
347 were used to calculate the DQI, with the highest scores reflecting greater
348 compliance with the diet (Iglesia et al., 2020).

349 *2.8 Statistical analysis*

350 Statistical analyses were carried out using IBM SPSS Statistics® software,
351 version 25 (IBM Corp., Armonk, NY, USA). The variables studied were
352 presented as means (M) ±, standard deviations (SD), and median and
353 interquartile intervals (25th and 75th) in the case of non-normally distributed
354 variables. The distribution of variables was checked and verified using the
355 Kolmogorov-Smirnov test. The Student's t-test or the Mann-Whitney U test were
356 used to identify sex differences between each of the studied variables. Because
357 we observed a significant interaction effect between gender and physical
358 activity and physical fitness, all the analyses were performed separately for
359 boys and girls.

360 Multiple linear regression (Forced Entry) was used to study the association
361 between physical activity (total, light, moderate, vigorous, or MVPA) or physical
362 fitness (handgrip strength, standing long jump) with the body composition
363 outcomes, which included TLSTMI, MCSAI, and FFMI. These relationships
364 were analyzed in individual regression models (one for each physical activity
365 level or physical fitness variable) using educational level, DQI, and birth weight
366 as covariates. The assumptions of independence of errors were verified for all
367 the multiple regression models using the Durbin-Watson test. Equally,
368 collinearity diagnosis was carried out through the variance inflation factor (VIF).

369 For the significant models, we used a mediation analysis according to the
370 procedure proposed by Baron and Kenny (Baron & Kenny, 1986) using the
371 macro PROCESS developed by Hayes (Hayes, 2013) with a bootstrap
372 threshold of 10,000 and model 4 for Statistical Package for the Social Sciences
373 (SPSS) version 25.0 (IBM Corporation, New York, USA). The purpose of this
374 model was to examine the total effect (c) and the individual direct effects (a, b,
375 c') reflected by the non-standardized regression coefficient (β), as well as the
376 statistical significance of the relationship between each model's independent
377 and dependent variables. The model also examined the indirect effect obtained
378 from the coefficients' product ($a \times b$). It indicated the change in TLSTMI by the
379 change in vigorous physical activity and the change in TLSTMI by the handgrip
380 strength change, mediated by the proposed mediator (handgrip strength or
381 physical activity, respectively). If zero was not included in the estimate's 95%
382 confidence interval (CI), we concluded that the indirect effect (IE) was
383 statistically significant, as shown in Figure 1.

384 **3. Results**

385 386 *3.1 Descriptive statistics*

387
388 The characteristics of participants ranked by gender are shown in **Table 1**. Boys
389 showed higher levels of TLSTM (kg), TLSTMI (kg/m^2), ALSTMI (kg/m^2), MCSA
390 (mm^2), MCSAI (mm^2/m^2), FFM (Kg), FFMI (kg/m^2), total, light, moderate,

391 vigorous, and MVPA, handgrip strength and standing long jump than girls (all
392 $p < 0.05$).

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394 *3.2 Association between physical activity or physical fitness with LBM outcomes*

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396 Adjusted associations between physical activity and TLSTMI, MCSAI, and FFMI
397 for both boys and girls are shown in **Table 2**.

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399 In girls, we observed that for Model 1 (physical activity [min/day]), there was an
400 association between total physical activity and TLSTMI ($\beta = 0.272$, $p = 0.004$). In
401 Model 2, after adding the DQI, the parent's educational level, and age, an
402 association was found between total physical activity ($\beta = 0.275$, $p = 0.004$) and
403 vigorous physical activity ($\beta = 0.159$, $p = 0.001$) with TLSTMI. Finally, once the
404 birth weight was added to create Model 3, slight decreases were found for the
405 significant association between total physical activity ($\beta = 0.247$, $p = 0.010$) and
406 vigorous physical activity ($\beta = 0.143$, $p = 0.001$) with TLSTMI.

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408 In boys, Model 1 showed a positive association between total physical activity
409 ($\beta = 0.335$, $p < 0.001$), light ($\beta = 0.287$, $p = 0.001$), and vigorous physical activity
410 ($\beta = 0.203$, $p = 0.020$) with TLSTMI by DXA. After adding the DQI, the parents'
411 educational level, and the age to create Model 2, slight increases were found in
412 the significant associations between total physical activity ($\beta = 0.371$, $p < 0.001$),
413 light physical activity ($\beta = 0.302$, $p = 0.001$), vigorous ($\beta = 0.224$, $p = 0.013$). An
414 association was found between MVPA ($\beta = 0.189$, $p = 0.038$) with TLSTMI (Table
415 2). Once birth weight was added to create Model 3, slight decreases were
416 observed in the associations between total physical activity ($\beta = 0.337$, $p < 0.001$),
417 light ($\beta = 0.290$, $p = 0.001$) and vigorous physical activity ($\beta = 0.200$, $p = 0.023$) with
418 TLSTMI (Table 2). The association between MVPA and TLSTMI in this model
419 disappeared.

420

421 No associations were found between physical activity and MCSAI for girls or
422 boys. Regarding FFMI assessed with bioimpedance, no associations were
423 found between FFMI and physical activity in girls. For boys, total physical
424 activity significantly predicted FFMI.

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2 426 The adjusted associations between handgrip strength and TLSTMI, MCSAI,
3 427 and FFMI both in boys and girls are shown in **Table 3**. For both genders,
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5 428 handgrip strength was a significant predictor for all the models. For girls, in
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7 429 Model 1, handgrip strength was positively associated with TLSTMI, MCSAI, and
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9 430 FFMI ($\beta=0.363$, $p<0.001$; $\beta=0.262$, $p=0.002$; $\beta=0.236$, $p=0.004$, respectively). In
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11 431 Model 2, significant associations remained for TLSTMI, MCSAI, and FFMI
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13 432 ($\beta=0.350$, $p<0.001$; $\beta=0.282$, $p=0.001$; $\beta=0.218$, $p=0.008$, respectively).
14
15 433 However, they decreased slightly for TLSTMI and FFMI. Finally, once birth
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17 434 weight was added to Model 3, the significant associations again decreased
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19 435 slightly, but remained significant ($\beta=0.314$, $p<0.001$; $\beta=0.267$, $p=0.002$;
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21 436 $\beta=0.196$, $p=0.017$, respectively).

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23 438 In boys, in Model 1 handgrip strength was positively associated with TLSTMI,
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25 439 MCSAI, and FFMI ($\beta=0.478$, $p<0.001$; $\beta=0.273$, $p<0.001$; $\beta=0.289$, $p<0.001$,
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27 440 respectively). In Model 2, significant associations were maintained for TLSTMI,
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29 441 MCSAI, and FFMI ($\beta=0.464$, $p<0.001$; $\beta=0.287$, $p<0.001$; $\beta=0.307$, $p<0.001$,
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31 442 respectively) but decreased slightly for TLSTMI and increased for MCSAI and
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33 443 BIA. Finally, once birth weight was added to Model 3, the significant
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35 444 associations again decreased slightly for TLSTMI and increased for MCSAI and
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37 445 BIA ($\beta=0.425$, $p<0.001$; $\beta=0.301$, $p<0.001$; $\beta=0.325$, $p=0.008$, respectively).

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40 447 There was no association between standing long jump and TLSTMI, MCSAI,
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42 448 and FFMI in both girls and boys (Table 3). This analysis was also performed
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44 449 adjusting standing long jump by weight, obtaining similar results.

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47 451 *3.3 Mediation analysis*

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51 453 An analysis of the handgrip's strength mediation effect on the association of
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53 454 total physical activity with TLSTMI values indicated an association between total
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55 455 physical activity and handgrip strength ($\beta_1=0.0057$, 95% CI: 0.0000 to 0.0114, p
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57 456 0.0484*) in the first regression equation (a). In the second equation (b),
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59 457 handgrip strength was positively associated with TLSTMI ($\beta_3=0.2188$, 95% CI:
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61 458 0.1637 to 0.2739, $p<0.001$). The third equation (c') showed a positive

1 459 relationship between total physical activity and TLSTMI ($\beta_{dir}=0.0073$, 95% CI:
2 460 0.0045 to 0.0101 $p<0.001^*$). The relationship between total physical activity and
3 461 TLSTMI was not statistically significant when including handgrip strength in the
4 462 model (a * b), indicating that handgrip strength does not mediate in this
5 463 relationship ($\beta_{ind}=0.0013$, 95% CI: 0.0000 a 0.0026) (Figure 1).
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10 465 **4. Discussion**

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13 467 Our results indicate that this sample of children accumulated around 110 min of
14 468 MVPA. Studies available in the population of the United States (Troost et al.,
15 469 2002) and Europe (Riddoch et al., 2004), where physical activity data were
16 470 objectively measured, suggest that young children (schoolchildren) accumulate
17 471 more than 100 min of MVPA per day. Our results are comparable with these
18 472 findings. This could be because children are more active at these ages
19 473 (possibly through organized activities (physical exercise and youth sports) and
20 474 not organized (recess and unstructured activity (Wickel & Eisenmann, 2007),
21 475 and active transport (Carver et al., 2011))), to later decrease the physical
22 476 activity in adolescence, especially in girls.
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36 479 Our main findings indicate that there is an association between total physical
37 480 activity and vigorous physical activity with TLSTMI in girls, while for boys, the
38 481 associations were found between total, light, vigorous, and MVPA with TLSTMI.
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40 482 One possible explanation may be the role of certain hormones (growth
41 483 hormone, IGF-1, and gender steroids, such as testosterone and estradiol),
42 484 which play a fundamental role in developing skeletal muscle during infancy,
43 485 childhood, and adolescence, depending on gender (Veldhuis et al., 2005).
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51 487 Additionally, our results indicate that upper limb strength (handgrip strength)
52 488 does not mediate the relationship between total physical activity and TLSTMI.
53 489 As far as we know, this is the first time that these results are shown in a broad
54 490 sample of young children using high precision objective methods.
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60 492 In agreement with our results, a study involving 283 Chinese adolescent girls
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493 also found a significant positive association between total physical activity level
494 assessed by questionnaire and LBM ($p < 0.001$), suggesting that higher physical
495 activity levels may reflect a higher LBM (Foo et al., 2007). Similarly, a study by
496 Jiménez-Pavón study using accelerometers found that total physical activity
497 was positively associated with FFM ($p < 0.05$) in 2,200 (1016 male, 1184
498 females) European adolescents of both sexes (Jiménez-Pavón et al., 2013). In
499 the same line, Deheeger et al. (Deheeger et al., 1997) found that total physical
500 activity was positively associated with the percentage of FFM ($r = 0.23$; $p = 0.03$) in
501 ten-year-old children ($n = 86$). Rennie et al. also showed that the level of physical
502 activity was positively associated with the lean mass index (LMI) in a study
503 carried out with 100 children aged 6 to 8 (Rennie et al., 2005). Our study is in
504 the same line as previously published studies, suggesting that total physical
505 activity positively affects non-adipose tissue in both female and male children
506 and adolescents.

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508 Regarding vigorous physical activity and MVPA, a study by Jiménez-Pavón
509 found positive associations of vigorous physical activity with FFM and muscle
510 mass (all $p < 0.05$) in both sexes (Jiménez-Pavón et al., 2013). In this study,
511 skinfold thickness and bioimpedance were used to evaluate body composition,
512 accelerometry for physical activity. Similarly, a study by Hao et al. found a
513 positive association between MVPA and SMMI ($\beta = 0.20$, $p < 0.001$). It involved
514 640 adolescents and concerned fat-free soft tissue mass assessed by DXA,
515 subsequently determining the skeletal muscle mass index (SMMI) (Hao et al.,
516 2019). Additionally, in a longitudinal study conducted in Pelotas (Brazil)
517 (Ramires et al., 2016), a consistent moderate and vigorous physical activity
518 practice during adolescence was associated with a greater lean mass index in
519 both sexes.

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521 Conversely, studies such as the one developed by Moliner-Urdiales et al.
522 (Moliner-Urdiales et al., 2010a) in a sample of 363 Spanish adolescents aged
523 12.5 to 17.5 did not observe an association between FFM and physical activity
524 levels. Heelan et al. (Heelan & Eisenmann, 2006), in 4- to 7-year-old children,
525 observed, only in girls, a negative correlation between MVPA and FFM ($r = -$
526 0.39 , $p < 0.05$).

527

528 Different factors could explain our contrasting results. They include the
529 individuals' age ranges included in each study (preschoolers (Leppänen et al.,
530 2016), adolescents (Foo et al., 2007; Hao et al., 2019; Jiménez-Pavón et al.,
531 2013), and schoolchildren (Deheeger et al., 1997; Rennie et al., 2005)) and
532 their maturation stage. Other factors include the methods for assessing body
533 composition (DXA (Foo et al., 2007; Hao et al., 2019; Heelan & Eisenmann,
534 2006), anthropometry (Deheeger et al., 1997; Jiménez-Pavón et al., 2013), BIA
535 (Jiménez-Pavón et al., 2013), isotopic dilution (Rennie et al., 2005), and air
536 displacement plethysmography (Leppänen et al., 2016)) and physical activity
537 (questionnaires (Deheeger et al., 1997; Foo et al., 2007; Ramires et al., 2016;
538 Rennie et al., 2005) and accelerometers (Jiménez-Pavón et al., 2013)). They
539 also include the different types of devices for assessing physical activity (
540 uniaxial (Hao et al., 2019; Heelan & Eisenmann, 2006; Moliner-Urdiales et al.,
541 2010b) or triaxial devices (Jiménez-Pavón et al., 2013; Leppänen et al., 2016),
542 as well as the different cutoff points to define physical activity intensities,
543 varying sampling intervals (epochs) (10s (Leppänen et al., 2016), 15 s
544 (Jiménez-Pavón et al., 2013) and 1min (Hao et al., 2019; Heelan & Eisenmann,
545 2006)), and sporadic or episodes of accumulated physical activity data.
546 Moreover, the presence of FFM (sum of lean mass and bone mass) or fat-free
547 soft tissue mass (FFM - bone mineral content) and the use of absolute values
548 (kg (Foo et al., 2007; Jiménez-Pavón et al., 2013)) or relative values (%
549 (Deheeger et al., 1997) or index (Hao et al., 2019; Leppänen et al., 2016;
550 Ramires et al., 2016; Rennie et al., 2005)) to express the value of the FFM/LBM
551 can make it difficult to compare individuals of different sizes appropriately
552 because FFM varies with height, weight, age. The percentage of FFM
553 automatically decreases in proportion to the increase in the % of body fat. The
554 last possible affecting factors are the covariates taken into account in the
555 analyzes (age and maturation stage, the mother's BMI and educational level,
556 the father's BMI and educational level, and the age at the time of measurement
557 and time of use awake).

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559 Some of these factors were also reported in a systematic review carried out by
560 Poitrasl et al. (Poitras et al., 2016). Their purpose was to examine the

1 561 relationships between objectively measured physical activity and relevant
2 562 indicators (body composition, cardiometabolic biomarkers, and physical fitness,
3 563 among others) in 5- to 17-year-old children and adolescents.
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7 565 Our findings can support emerging evidence suggesting that different physical
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9 566 activity intensities, including light physical activity, may significantly affect health
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11 567 outcomes. They are in line with Corson's findings in 1,731 adolescents aged 12
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13 568 to 19, evaluated during the National Health and Nutrition Examination Survey
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15 569 2003/2004 and 2005/2006 (Carson et al., 2013)—reinforcing the claim that
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17 570 some activity is better than none, but that “more is better” (Tremblay et al.,
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19 571 2011). However, we also found that the assessment method could influence
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21 572 results. No significant associations were found when evaluating the
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23 573 associations using the variables of the lean component determined with BIA or
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25 574 pQCT. An explanation for this could be the fact that, although BIA can provide
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27 575 information on the whole-body FFM status (like DXA), alterations in body water,
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29 576 such as dehydration, can influence it, increasing resistance to electricity, and
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31 577 subsequently underestimating the FFM (or overestimating body fat). Another
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33 578 influence could be the time of the day. In this study, the DXA and BIA analyses
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35 579 were carried out between 4.30 pm and 6.30 pm to ensure that no child was
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37 580 fasting, influencing the results. Other influences could include food consumption
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39 581 and recent activity (exercise) (Heymsfield et al., 2015).
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43 583 On the other hand, pQCT quantifies the cross-sectional area of the peripheral
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45 584 (appendicular)/limb muscle (MCSA)(Frank-Wilson et al., 2015) and not the
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47 585 whole body, which could explain why we found no associations between
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49 586 physical activity and MCSAI. MCSA can also be influenced by the
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51 587 anthropometric characteristics of the subjects (body and limb size); therefore,
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53 588 we used MCSAI. Meanwhile, DXA provides information on the LSTM
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55 589 (Bazzocchi et al., 2016) of the whole body or regional/segmental zones
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57 590 (Laskey, 1996), with the advantages of its high precision and
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59 591 reproducibility(Guglielmi et al., 2016).
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63 593 Our main findings also suggest a positive association between upper body
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65 594 muscle strength (handgrip strength) and TLSTMI, MCSAI, and FFMI in both

1 595 boys and girls, after controlling for gender, education level, DQI, and birth
2 596 weight. The results were consistent regardless of the methodology used to
3 597 assess body FFM; however, the associations were weaker when BIA was used
4 598 to measure LSTM markers. Similar results were found in the MINISTOP (mobile
5 599 device-based intervention aimed at stopping obesity in pre-schoolers) study
6 600 (Henriksson et al., 2016), where PREFIT (physical fitness test battery in
7 601 preschool children) was used to measure physical fitness (Ortega et al., 2015)
8 602 in 303 4-year-old children. Its associations showed a higher FFMI in participants
9 603 with better upper body muscle strength (handgrip strength) ($\beta=0.39$, $p<0.001$).
10 604

11 605 Anthropometric variables such as body height, body mass, and BMI, which can
12 606 influence grip strength, may explain these findings. In the case of body height, it
13 607 is directly correlated with grip strength; this could partially explain its close
14 608 relation to LBM (Jürimäe et al., 2009). Both the MINISTOP study and the
15 609 present study made the respective height adjustment.
16 610

17 611 The present study found no association between standing long jump and
18 612 TLSTMI, MCSAI, and FFMI. However, another study by Vicente-Rodriguez et
19 613 al. (Vicente-Rodriguez et al., 2004) showed an increase in the FFM of 28
20 614 children (soccer players) followed for three years. They found no significant
21 615 differences in muscle strength after evaluation through vertical jumps and
22 616 maximal isometric force (MIF) during leg extension. Conversely, Henriksson et
23 617 al. (Henriksson et al., 2016) observed that higher FFMI was associated with
24 618 improved lower-body muscular strength ($\beta=0.22$, $p<0.001$). This disparity could
25 619 be explained by the different populations included in each of the studies
26 620 (preschoolers, schoolchildren, and soccer players), as well as their maturation
27 621 stage (preschoolers, schoolchildren, and pre-puberty), the body composition
28 622 evaluation methods (DXA and air displacement plethysmography) and the
29 623 measurement methodology for the forces generated during vertical jumps (force
30 624 plate versus standing long jump test). The lack of association between lower
31 625 limb strength and FFM may also be because this test is considered weight-
32 626 dependent, requiring propulsion or body elevation. Therefore, children and
33 627 adolescents with higher LBM may not display better performance in these tests
34 628 because they likely weigh differently. These results could suggest that higher
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629 performance in some physical fitness tests may occur in underweight
630 individuals because of the lower load that they must move.

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632 Finally, the relationship between physical activity and physical fitness (handgrip
633 strength) and TLSTMI in our study is mainly consistent with the existing
634 evidence showing positive associations independent of physical activity and
635 physical fitness in TLSTMI. Our research shows that the association of total
636 physical activity with TLSTMI is not mediated by physical fitness (handgrip
637 strength). Therefore, future actions seeking to improve TLSTM in boys and girls
638 should encourage activities that enhance physical activity levels and physical
639 fitness.

640

641 *4.1 Limitations and Strengths*

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643 Within the limitations of the present study, there is the fact that the data cannot
644 be extrapolated to other populations. Studies with different age groups and
645 different ethnic groups should be developed in the future. Furthermore, due to
646 its cross-sectional design, the observed associations cannot be interpreted as
647 causal relationships.

648

649 Despite these limitations, the strengths of our study are many. To date, studies
650 on a homogeneous sample with such young children are limited. The use of
651 accelerometers allowed the objective assessment of physical activity, including
652 both weekdays and weekend days. LSTM and FFM were measured using two
653 different techniques, DXA and BIA, the former, a robust method accepted as
654 the gold standard method. MCSA was measured by pQCT, a lower-cost method
655 previously validated against DXA in this population. The DXA and pQCT
656 measurements and the respective image analysis were performed by two
657 trained researchers in each of the techniques. The TLSTM, ALSTM, MCSA, and
658 FFM results were adjusted for the height of the subjects. Different tests were
659 used to assess muscle strength (handgrip strength and standing long jump),
660 controlling for several confounders, including gender, age, birth weight, DQI,
661 and parents' education level. Furthermore, it appears to be the first study
662 evaluating handgrip strength as a potential mediator of the relationship between

663 physical activity and TLSTMI in children.

664

665 **5. Conclusions**

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667 The present study suggests positive associations between total and vigorous
668 physical activity and TLSTMI in boys and girls aged 6 to 8. Its results indicate
669 that upper limb strength (handgrip strength) does not mediate the relationship
670 between total physical activity and TLSTMI.

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672 The results also suggest an association between handgrip strength and
673 TLSTMI, MCSAI, and FFMI in 6- to 8-year-old children. More studies are
674 needed in the future to clarify the relationships between physical activity and
675 physical fitness with TLSTM, MCSA, and FFM better, taking into account other
676 confounding factors such as genetics factors, as well as physical activity
677 intensities. To date, most of the available studies focus on the relationship at
678 higher intensities of physical activity (i.e., MVPA and vigorous physical activity).

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680 Public policies should encourage participation in all physical activities, including
681 light physical activity. As we have shown, it can be an effective substitute for
682 sedentary activities and expand the focus beyond the MVPA as a strategy to
683 promote our children's health. Likewise, exercises that improve upper extremity
684 strength and lean mass should be encouraged, which, as mentioned above, is
685 an important tissue in children.

686

687 **Acknowledgments**

688 The authors would like to thank the parents and children who participated in this
689 research.

690

691 The CALINA study was supported by three grants from the Carlos III Health
692 Institute: the PI08/0559 Aragon Health Sciences Institute for the Growth and
693 Feeding in Infants from Aragon (CALINA) project, the PI13/02359
694 Environmental factors influencing early development of obesity during childhood
695 and body composition programming, and the RD16/0022 Maternal, Child
696 Health, and Development Network (Retic SAMID) RETICS, funded by the PN

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697 I+D+I 2008-2011 (Spain), ISCIII- Sub-Directorate General for Research
698 Assessment and Promotion, and the European Regional Development Fund
699 (ERDF). Additionally, we thank the Carolina Foundation Ph.D. Grants for
700 supporting first author in producing this manuscript.

701

702 **Authors' Contributions**

703 DPCR analyzed the data and drafted the manuscript. IIA conducted the data
704 collection and drafted the manuscript. AGB drafted the manuscript. MLMB
705 conducted the data collection. PFB conducted the data collection. JACS and
706 LMA drafted the manuscript. GR conceived the study and participated in its
707 design and coordination and helped draft the manuscript.

708 All the authors have read and approved this manuscript's final version. They
709 agree with the authors' order of presentation.

710

711 **Disclosure Statement**

712

713 The authors report no conflict of interest.

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716 **Bibliography**

717 Bammann, K., Sioen, I., Huybrechts, I., Casajús, J. A., Vicente-Rodríguez, G.,
718 Cuthill, R., Konstabel, K., Tubić, B., Wawro, N., Rayson, M., Westerterp, K.,
719 Mårild, S., Pitsiladis, Y. P., Reilly, J. J., Moreno, L. A., & De Henauw, S.
720 (2011). The IDEFICS validation study on field methods for assessing
721 physical activity and body composition in children: Design and data
722 collection. *International Journal of Obesity*, 35, S79–S87.
723 <https://doi.org/10.1038/ijo.2011.38>

724 Baron, R. M., & Kenny, D. A. (1986). The moderator–mediator variable
725 distinction in social psychological research: Conceptual, strategic, and
726 statistical considerations. *Journal of Personality and Social Psychology*,
727 51(6), 1173. <https://doi.org/https://doi.org/10.1037/0022-3514.51.6.1173>

728 Baxter-Jones, A. D. G., Eisenmann, J. C., Mirwald, R. L., Faulkner, R. A., &
729 Bailey, D. A. (2008). The influence of physical activity on lean mass accrual

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- 1 730 during adolescence: a longitudinal analysis. *Journal of Applied Physiology*,
2 731 105(2), 734–741.
3
4 732 <https://doi.org/https://doi.org/10.1152/jappphysiol.00869.2007>
- 5
6 733 Bazzocchi, A., Ponti, F., Albisinni, U., Battista, G., & Guglielmi, G. (2016). DXA:
7 734 Technical aspects and application. *European Journal of Radiology*, 85(8),
8 735 1481–1492. <https://doi.org/10.1016/j.ejrad.2016.04.004>
- 9
10
11 736 Boisseau, N., & Delamarche, P. (2000). Metabolic and hormonal responses to
12 737 exercise in children and adolescents. *Sports Medicine*, 30(6), 405–422.
13 738 <https://doi.org/10.2165/00007256-200030060-00003>
- 14
15
16
17 739 Burrows, R., Correa-Burrows, P., Reyes, M., Blanco, E., Albala, C., & Gahagan,
18 740 S. (2017). Low muscle mass is associated with cardiometabolic risk
19 741 regardless of nutritional status in adolescents: A cross-sectional study in a
20 742 Chilean birth cohort. *Pediatric Diabetes*, 18(8), 895–902.
21 743 <https://doi.org/10.1111/pedi.12505>
- 22
23
24
25 744 Carson, V., Ridgers, N. D., Howard, B. J., Winkler, E. A. H., Healy, G. N.,
26 745 Owen, N., Dunstan, D. W., & Salmon, J. (2013). Light-Intensity Physical
27 746 Activity and Cardiometabolic Biomarkers in US Adolescents. *PLoS ONE*,
28 747 8(8). <https://doi.org/10.1371/journal.pone.0071417>
- 29
30
31
32 748 Carver, A., Timperio, A. F., Hesketh, K. D., Ridgers, N. D., Salmon, J. L., &
33 749 Crawford, D. A. (2011). How is active transport associated with children's
34 750 and adolescents' physical activity over time? *International Journal of*
35 751 *Behavioral Nutrition and Physical Activity*, 8, 1–6.
36 752 <https://doi.org/10.1186/1479-5868-8-126>
- 37
38
39
40 753 Córdoba-Rodríguez, D. P., Iglesia, I., Gomez-Bruton, A., Miguel-Berges, M. L.,
41 754 Flores-Barrantes, P., Casajús, J. A., Moreno, L. A., & Rodríguez, G. (2021).
42 755 Quantitative peripheral computed tomography to measure muscle area and
43 756 assess lean soft tissue mass in children. *Annals of Human Biology*, 1–27.
44 757 <https://doi.org/10.1080/03014460.2021.1877352>
- 45
46
47
48 758 Crabtree, N. J., Leonard, M. B., & Zemel, B. S. (2007). *Dual-Energy X-Ray*
49 759 *Absorptiometry BT - Bone Densitometry in Growing Patients: Guidelines*
50 760 *for Clinical Practice* (A. J. Sawyer, L. K. Bachrach, & E. B. Fung (eds.); pp.
51 761 41–57). Humana Press. https://doi.org/10.1007/978-1-59745-211-3_3
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56
57
58
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60
61
62
63
64
65

762 De Miguel-Etayo, P., Gracia-Marco, L., Ortega, F. B., Intemann, T., Foraita, R.,
763 Lissner, L., Oja, L., Barba, G., Michels, N., Tornaritis, M., Molnár, D.,
764 Pitsiladis, Y., Ahrens, W., & Moreno, L. A. (2014). Physical fitness
765 reference standards in European children: The IDEFICS study.
766 *International Journal of Obesity*, 38(September), S57–S66.
767 <https://doi.org/10.1038/ijo.2014.136>

768 Deheeger, M., Rolland-Cachera, M. F., & Fontvieille, A. M. (1997). Physical
769 activity and body composition in 10 year old French children: Linkages with
770 nutritional intake? *International Journal of Obesity*, 21(5), 372–379.
771 <https://doi.org/10.1038/sj.ijo.0800415>

772 Dorsey, K. B., Thornton, J. C., Heymsfield, S. B., & Gallagher, D. (2010).
773 Greater lean tissue and skeletal muscle mass are associated with higher
774 bone mineral content in children. *Nutrition and Metabolism*, 7, 1–11.
775 <https://doi.org/10.1186/1743-7075-7-41>

776 Dotan, R., Mitchell, C., Cohen, R., Klentrou, P., Gabriel, D., & Falk, B. (2012).
777 Child-adult differences in muscle activation - A review. *Pediatric Exercise
778 Science*, 24(1), 2–21. <https://doi.org/10.1123/pes.24.1.2>

779 España-Romero, V., Artero, E. G., Santaliestra-Pasias, A. M., Gutierrez, A.,
780 Castillo, M. J., & Ruiz, J. R. (2008). Hand Span Influences Optimal Grip
781 Span in Boys and Girls Aged 6 to 12 Years. *Journal of Hand Surgery*,
782 33(3), 378–384. <https://doi.org/10.1016/j.jhsa.2007.11.013>

783 Evenson, K. R., Catellier, D. J., Gill, K., Ondrak, K. S., & McMurray, R. G.
784 (2008). Calibration of two objective measures of physical activity for
785 children. *Journal of Sports Sciences*, 26(14), 1557–1565.
786 <https://doi.org/10.1080/02640410802334196>

787 Foo, L. H., Zhang, Q., Zhu, K., Ma, G., Greenfield, H., & Fraser, D. R. (2007).
788 *Influence of body composition , muscle strength , diet and physical activity
789 on total body and forearm bone mass in Chinese adolescent girls*. 1281–
790 1287. <https://doi.org/10.1017/S0007114507787421>

791 Frank-Wilson, A. W., Johnston, J. D., Olszynski, W. P., & Kontulainen, S. A.
792 (2015). Measurement of muscle and fat in postmenopausal women:
793 precision of previously reported pQCT imaging methods. *Bone*, 75, 49–54.

- 794 <https://doi.org/10.1016/J.BONE.2015.01.016>
- 1
2 795 Fraser, B. J., Blizzard, L., Cleland, V., Schmidt, M. D., Smith, K. J., Gall, S. L.,
3
4 796 Dwyer, T., Venn, A. J., & Magnussen, C. G. (2020). Factors associated
5
6 797 with muscular fitness phenotypes in Australian children: A cross-sectional
7
8 798 study. *Journal of Sports Sciences*, 38(1), 38–45.
9
10 799 <https://doi.org/10.1080/02640414.2019.1679575>
- 11 800 Gómez-Bruton, A., Gonzalez-Agüero, A., Casajús, J. A., & Rodríguez, G. V.
12
13 801 (2014). Swimming training repercussion on metabolic and structural bone
14
15 802 development; benefits of the incorporation of whole body vibration or
16
17 803 pilometric training; the RENACIMIENTO project. *Nutricion Hospitalaria*,
18
19 804 30(2), 399–409. <https://doi.org/10.3305/nh.2014.30.2.7603>
- 20
21 805 González-Agüero, A., Vicente-Rodríguez, G., Gómez-Cabello, A., & Casajús, J.
22
23 806 A. (2013). Cortical and trabecular bone at the radius and tibia in male and
24
25 807 female adolescents with Down syndrome: a peripheral quantitative
26
27 808 computed tomography (pQCT) study. *Osteoporosis International: A*
28
29 809 *Journal Established as Result of Cooperation between the European*
30
31 810 *Foundation for Osteoporosis and the National Osteoporosis Foundation of*
32
33 811 *the USA*, 24(3), 1035—1044. <https://doi.org/10.1007/s00198-012-2041-7>
- 34
35 812 Gracia-Marco, L., Ortega, F. B., Jiménez-Pavón, D., Rodríguez, G., Castillo, M.
36
37 813 J., Vicente-Rodríguez, G., & Moreno, L. A. (2012). Adiposity and bone
38
39 814 health in Spanish adolescents. The HELENA study. *Osteoporosis*
40
41 815 *International*, 23(3), 937–947. <https://doi.org/10.1007/s00198-011-1649-3>
- 42
43 816 Gracia-Marco, Luis, Moreno, L. A., Ruiz, J. R., Ortega, F. B., de Moraes, A. C.
44
45 817 F., Gottrand, F., Roccaldo, R., Marcos, A., Gómez-Martínez, S.,
46
47 818 Dallongeville, J., Kafatos, A., Molnar, D., Bueno, G., de Henauw, S.,
48
49 819 Widhalm, K., & Wells, J. C. (2016). Body Composition Indices and Single
50
51 820 and Clustered Cardiovascular Disease Risk Factors in Adolescents:
52
53 821 Providing Clinical-Based Cut-Points. *Progress in Cardiovascular Diseases*,
54
55 822 58(5), 555–564. <https://doi.org/10.1016/j.pcad.2015.11.002>
- 56
57 823 Guglielmi, G., Ponti, F., Agostini, M., Amadori, M., Battista, G., & Bazzocchi, A.
58
59 824 (2016). The role of DXA in sarcopenia. *Aging Clin Exp Res*, 28(6), 1047–
60
61 825 1060. <https://doi.org/10.1007/s40520-016-0589-3>
- 62
63
64
65

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2
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52
53
54
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56
57
58
59
60
61
62
63
64
65

826 Hao, G., Pollock, N. K., Harris, R. A., Gutin, B., Su, S., & Wang, X. (2019).
827 Associations between muscle mass, physical activity and dietary behaviour
828 in adolescents. *Pediatric Obesity*, *14*(3), 1–8.
829 <https://doi.org/10.1111/ijpo.12471>

830 Hayes, A. F. (2013). Mediation, moderation, and conditional process analysis.
831 Introduction to mediation, moderation, and conditional process analysis: A
832 regression-based approach. *New York: Guilford Publications*, 1–20.

833 Heelan, K. A., & Eisenmann, J. C. (2006). Physical activity, media time, and
834 body composition in young children. *Journal of Physical Activity and Health*,
835 *3*(2), 200–209. <https://doi.org/10.1123/jpah.3.2.200>

836 Henriksson, P., Cadenas-Sanchez, C., Leppänen, M. H., Nyström, C. D.,
837 Ortega, F. B., Pomeroy, J., Ruiz, J. R., & Löf, M. (2016). Associations of fat
838 mass and fat-free mass with physical fitness in 4-year-old children: Results
839 from the MINISTOP trial. *Nutrients*, *8*(8), 1–11.
840 <https://doi.org/10.3390/nu8080473>

841 Heymsfield, S. B., Gonzalez, M. C., Lu, J., Jia, G., & Zheng, J. (2015). Skeletal
842 muscle mass and quality: Evolution of modern measurement concepts in
843 the context of sarcopenia. *Proceedings of the Nutrition Society*, *74*(4), 355–
844 366. <https://doi.org/10.1017/S0029665115000129>

845 Huybrechts, I., Vereecken, C., De Bacquer, D., Vandevijvere, S., Van Oyen, H.,
846 Maes, L., Vanhauwaert, E., Temme, L., De Backer, G., & De Henauw, S.
847 (2010). Reproducibility and validity of a diet quality index for children
848 assessed using a FFQ. *British Journal of Nutrition*, *104*(1), 135–144.
849 <https://doi.org/10.1017/S0007114510000231>

850 Iglesia, I., Intemann, T., De Miguel-Etayo, P., Pala, V., Hebestreit, A., Wolters,
851 M., Russo, P., Veidebaum, T., Papoutsou, S., Nagy, P., Eiben, G., Rise, P.,
852 De Henauw, S., & Moreno, L. A. (2020). Dairy consumption at snack meal
853 occasions and the overall quality of diet during childhood. Prospective and
854 cross-sectional analyses from the idefics/i.family cohort. *Nutrients*, *12*(3),
855 1–20. <https://doi.org/10.3390/nu12030642>

856 Isganaitis, E. (2019). Developmental Programming of Body Composition:
857 Update on Evidence and Mechanisms. *Current Diabetes Reports*, *19*(8), 1–

- 1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
20
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47
48
49
50
51
52
53
54
55
56
57
58
59
60
61
62
63
64
65
- 858 20. <https://doi.org/10.1007/s11892-019-1170-1>
- 859 Jiménez-Pavón, D., Fernández-Vázquez, A., Alexy, U., Pedrero, R., Cuenca-
860 García, M., Polito, A., Vanhelst, J., Manios, Y., Kafatos, A., Molnar, D.,
861 Sjöström, M., & Moreno, L. A. (2013). Association of objectively measured
862 physical activity with body components in European adolescents. *BMC*
863 *Public Health*, 13(1). <https://doi.org/10.1186/1471-2458-13-667>
- 864 Jürimäe, T., Hurbo, T., & Jürimäe, J. (2009). Relationship of handgrip strength
865 with anthropometric and body composition variables in prepubertal
866 children. *HOMO- Journal of Comparative Human Biology*, 60(3), 225–238.
867 <https://doi.org/10.1016/j.jchb.2008.05.004>
- 868 Kim, J. H., & Park, Y. S. (2016). Low muscle mass is associated with metabolic
869 syndrome in Korean adolescents: the Korea National Health and Nutrition
870 Examination Survey 2009-2011. *Nutrition Research*, 36(12), 1423–1428.
871 <https://doi.org/10.1016/j.nutres.2016.09.013>
- 872 Kim, K., Hong, S., & Kim, E. Y. (2016). Reference values of skeletal muscle
873 mass for Korean children and adolescents using data from the Korean
874 national health and nutrition examination survey 2009-2011. *PLoS ONE*,
875 11(4), 1–10. <https://doi.org/10.1371/journal.pone.0153383>
- 876 Kim, S., & Valdez, R. (2015). Metabolic risk factors in U.S. youth with low
877 relative muscle mass. *Obesity Research and Clinical Practice*, 9(2), 125–
878 132. <https://doi.org/10.1016/j.orcp.2014.05.002>
- 879 Kulkarni, B., Hills, A. P., & Byrne, N. M. (2014). Nutritional influences over the
880 life course on lean body mass of individuals in developing countries.
881 *Nutrition Reviews*, 72(3), 190–204. <https://doi.org/10.1111/nure.12097>
- 882 Kuriyan, R. (2018). Body composition techniques. *Indian Journal of Medical*
883 *Research*, 148(5), 648–658. https://doi.org/10.4103/ijmr.IJMR_1777_18
- 884 Labayen, I., Moreno, L. A., Blay, M. G., Blay, V. A., Mesana, M. I., González-
885 Gross, M., Bueno, G., Sarría, A., & Bueno, M. (2006). Early programming
886 of body composition and fat distribution in adolescents. *Journal of Nutrition*,
887 136(1), 147–152. <https://doi.org/10.1093/jn/136.1.147>
- 888 Larqué, E., Labayen, I., Flodmark, C. E., Lissau, I., Czernin, S., Moreno, L. A.,

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
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19
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45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
61
62
63
64
65

889 Pietrobelli, A., & Widhalm, K. (2019). From conception to infancy — early
890 risk factors for childhood obesity. *Nature Reviews Endocrinology*, *15*(8),
891 456–478. <https://doi.org/10.1038/s41574-019-0219-1>

892 Laskey, M. A. (1996). Dual-energy X-ray absorptiometry and body composition.
893 *Nutrition*, *12*(1), 45–51.
894 <https://doi.org/10.1097/01.mco.0000165010.31826.3d>

895 Leppänen, M., Henriksson, P., Delisle Nyström, C., Henriksson, H., Ortega, F.
896 B., Pomeroy, J., Ruiz, J. R., Cadenas-Sanchez, C., & Löf, M. (2017).
897 Longitudinal physical activity, body composition, and physical fitness in
898 preschoolers. In *Medicine and Science in Sports and Exercise* (Vol. 49,
899 Issue 10). <https://doi.org/10.1249/MSS.0000000000001313>

900 Leppänen, M., Nyström, C., Henriksson, P., Pomeroy, J., Ruiz, J. R., Ortega, F.,
901 Cadenas-Sánchez, C., & Löf, M. (2016). Physical activity intensity,
902 sedentary behavior, body composition and physical fitness in 4-year-old
903 children: Results from the ministop trial. *International Journal of Obesity*,
904 *40*(7), 1126–1133. <https://doi.org/10.1038/ijo.2016.54>

905 Liu, J., Yan, Y., Xi, B., Huang, G., & Mi, J. (2019). Skeletal muscle reference for
906 Chinese children and adolescents. *Journal of Cachexia, Sarcopenia and*
907 *Muscle*, *10*(1), 155–164. <https://doi.org/10.1002/jcsm.12361>

908 López-Gil, J. F., Brazo-Sayavera, J., García-Hermoso, A., & Yuste Lucas, J. L.
909 (2020). Adherence to Mediterranean Diet Related with Physical Fitness and
910 Physical Activity in Schoolchildren Aged 6–13. *Nutrients*, *12*(2), 567.
911 <https://doi.org/10.3390/nu12020567>

912 Moliner-Urdiales, D., Ortega, F. B., Vicente-Rodriguez, G., Rey-Lopez, J. P.,
913 Gracia-Marco, L., Widhalm, K., Sjöström, M., Moreno, L. A., Castillo, M. J.,
914 & Ruiz, J. R. (2010a). Association of physical activity with muscular
915 strength and fat-free mass in adolescents: The HELENA study. *European*
916 *Journal of Applied Physiology*, *109*(6), 1119–1127.
917 <https://doi.org/10.1007/s00421-010-1457-z>

918 Moliner-Urdiales, D., Ortega, F. B., Vicente-Rodriguez, G., Rey-Lopez, J. P.,
919 Gracia-Marco, L., Widhalm, K., Sjöström, M., Moreno, L. A., Castillo, M. J.,
920 & Ruiz, J. R. (2010b). Association of physical activity with muscular

1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
17
18
19
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49
50
51
52
53
54
55
56
57
58
59
60
61
62
63
64
65

921 strength and fat-free mass in adolescents: The HELENA study. *European*
922 *Journal of Applied Physiology*, 109(6), 1119–1127.
923 <https://doi.org/10.1007/s00421-010-1457-z>

924 Mouratidou, T., Mesana Graffe, M. I., Huybrechts, I., De Decker, E., De
925 Craemer, M., Androustos, O., Manios, Y., Galcheva, S., Lateva, M.,
926 Gurzkowska, B., Kułaga, Z., Birnbaum, J., Koletzko, B., & Moreno, L. A.
927 (2019). Reproducibility and relative validity of a semiquantitative food
928 frequency questionnaire in European preschoolers: The ToyBox study.
929 *Nutrition*, 65, 60–67. <https://doi.org/10.1016/j.nut.2019.03.003>

930 National Health and Nutrition Examination Survey (NHANES). (2012). *Body*
931 *Composition Procedures Manual*. July, 1–125.

932 Orsso, C. E., Tibaes, J. R. B., Oliveira, C. L. P., Rubin, D. A., Field, C. J.,
933 Heymsfield, S. B., Prado, C. M., & Haqq, A. M. (2019). Low muscle mass
934 and strength in pediatrics patients: Why should we care? *Clinical Nutrition*,
935 38(5), 2002–2015. <https://doi.org/10.1016/j.clnu.2019.04.012>

936 Ortega, F. B., Cadenas-Sánchez, C., Sánchez-Delgado, G., Mora-González, J.,
937 Martínez-Téllez, B., Artero, E. G., Castro-Piñero, J., Labayen, I., Chillón,
938 P., Löf, M., & Ruiz, J. R. (2015). Systematic Review and Proposal of a
939 Field-Based Physical Fitness-Test Battery in Preschool Children: The
940 PREFIT Battery. *Sports Medicine*, 45(4), 533–555.
941 <https://doi.org/10.1007/s40279-014-0281-8>

942 Oves Suárez, B., Escartín Madurga, L., Samper Villagrasa, M. P., Cuadrón
943 Andrés, L., Álvarez Sauras, M. L., Lasarte Velillas, J. J., Moreno Aznar, L.
944 A., & Rodríguez Martínez, G. (2014). Inmigración y factores asociados con
945 la lactancia materna. Estudio CALINA. *Anales de Pediatría*, 81, 32–38.
946 <https://doi.org/10.1016/j.anpedi.2013.09.008>

947 Poitras, V. J., Gray, C. E., Borghese, M. M., Carson, V., Chaput, J.-P., Janssen,
948 I., Katzmarzyk, P. T., Pate, R. R., Connor Gorber, S., & Kho, M. E. (2016).
949 Systematic review of the relationships between objectively measured
950 physical activity and health indicators in school-aged children and youth.
951 *Applied Physiology, Nutrition, and Metabolism*, 41(6), S197–S239.
952 <https://doi.org/10.1139/apnm-2015-0663>

1
2
3
4
5
6
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11
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48
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52
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54
55
56
57
58
59
60
61
62
63
64
65

953 Ramires, V. V., Dumith, S. C., Wehrmeister, F. C., Hallal, P. C., Menezes, A. M.
954 B., & Gonçalves, H. (2016). Physical activity throughout adolescence and
955 body composition at 18 years: 1993 Pelotas (Brazil) birth cohort study.
956 *International Journal of Behavioral Nutrition and Physical Activity*, 13(1), 1–
957 13. <https://doi.org/10.1186/s12966-016-0430-6>

958 Rennie, K. L., Livingstone, M. B. E., Wells, J. C. K., Mcgloin, A., Coward, W. A.,
959 & Prentice, A. M. (2005). *Association of physical activity with body-*
960 *composition indexes in children aged 6 – 8 y at varied risk of obesity 1 – 3.*
961 13–20. <https://doi.org/https://doi.org/10.1093/ajcn/82.1.13>

962 Riddoch, C. J., Andersen, L. B., Wedderkopp, N., Harro, M., Klasson-Heggebø,
963 L., Sardinha, L. B., Cooper, A. R., & Ekelund, U. (2004). Physical Activity
964 Levels and Patterns of 9- and 15-yr-Old European Children. *Medicine and*
965 *Science in Sports and Exercise*, 36(1), 86–92.
966 <https://doi.org/10.1249/01.MSS.0000106174.43932.92>

967 Roggen, I., Roelants, M., Sioen, I., Vandewalle, S., De Henauw, S., Goemaere,
968 S., Kaufman, J.-M., & De Schepper, J. (2015). Pediatric Reference Values
969 for Tibial Trabecular Bone Mineral Density and Bone Geometry Parameters
970 Using Peripheral Quantitative Computed Tomography. *Calcified Tissue*
971 *International*, 96(6), 527–533. <https://doi.org/10.1007/s00223-015-9988-2>

972 Ruiz, J. R., Castro-Piñero, J., Artero, E. G., Ortega, F. B., Sjöström, M., Suni, J.,
973 & Castillo, M. J. (2009). Predictive validity of health-related fitness in youth:
974 a systematic review. *British Journal of Sports Medicine*, 43(12), 909 LP –
975 923. <https://doi.org/10.1136/bjism.2008.056499>

976 Santos, D. A., Magalhaes, J. P., Judice, P. B., Correia, I. R., Minderico, C. S.,
977 Ekelund, U., & Sardinha, L. B. (2019). Fitness Mediates Activity and
978 Sedentary Patterns Associations with Adiposity in Youth. *Medicine &*
979 *Science in Sports & Exercise*, 51(2), 323–329.
980 <https://doi.org/10.1249/MSS.0000000000001785>

981 Scheurer, J. M., Zhang, L., Plummer, E. A., Hultgren, S. A., Demerath, E. W., &
982 Ramel, S. E. (2018). Body Composition Changes from Infancy to 4 Years
983 and Associations with Early Childhood Cognition in Preterm and Full-Term
984 Children. *Neonatology*, 114(2), 169–176.

- 1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
16
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54
55
56
57
58
59
60
61
62
63
64
65
- 985 <https://doi.org/10.1159/000487915>
- 986 Singhal, A., Wells, J., Cole, T. J., Fewtrell, M., & Lucas, A. (2003). Programming
987 of lean body mass: A link between birth weight, obesity, and cardiovascular
988 disease? *American Journal of Clinical Nutrition*, 77(3), 726–730.
989 <https://doi.org/10.1093/ajcn/77.3.726>
- 990 Sioen, I., Lust, E., De Henauw, S., Moreno, L. A., & Jiménez-Pavón, D. (2016).
991 Associations Between Body Composition and Bone Health in Children and
992 Adolescents: A Systematic Review. *Calcified Tissue International*, 99(6),
993 557–577. <https://doi.org/10.1007/s00223-016-0183-x>
- 994 Statistics, U. I. for. (2012). *International standard classification of education:*
995 *ISCED 2011*. UNESCO Institute for Statistics Montreal.
- 996 Tanita Corporation - Japan. (2002). *Body Composition Analyzer BC-418*
997 *Instruction Manual*. 1–43.
- 998 Tremblay, M. S., Warburton, D. E. R., Janssen, I., Paterson, D. H., Latimer, A.
999 E., Rhodes, R. E., Kho, M. E., Hicks, A., LeBlanc, A. G., Zehr, L.,
1000 Murumets, K., & Duggan, M. (2011). New Canadian physical activity
1001 guidelines. *Applied Physiology, Nutrition and Metabolism*, 36(1), 36–46.
1002 <https://doi.org/10.1139/H11-009>
- 1003 Trost, S. G., Pate, R. R., Sallis, J. F., Freedson, P. S., Taylor, W. C., Dowda,
1004 M., & Sirard, J. (2002). Age and gender differences in objectively measured
1005 physical activity in youth. *Medicine and Science in Sports and Exercise*,
1006 34(2), 350–355. <https://doi.org/10.1097/00005768-200202000-00025>
- 1007 Unesco. (1997). *International Standard Classification of Education-ISCED*
1008 *1997: November 1997*. Unesco.
- 1009 Veldhuis, J. D., Roemmich, J. N., Richmond, E. J., Rogol, A. D., Lovejoy, J. C.,
1010 Sheffield-Moore, M., Mauras, N., & Bowers, C. Y. (2005). Endocrine control
1011 of body composition in infancy, childhood, and puberty. *Endocrine*
1012 *Reviews*, 26(1), 114–146. <https://doi.org/10.1210/er.2003-0038>
- 1013 Vicente-Rodriguez, G., Ara, I., Perez-Gomez, J., Serrano-Sanchez, J. A.,
1014 Dorado, C., & Calbet, J. A. L. (2004). High Femoral Bone Mineral Density
1015 Accretion in Prepubertal Soccer Players. *Medicine & Science in Sports &*

- 1016 *Exercise*, 36(10). <https://journals.lww.com/acsm->
1017 [msse/Fulltext/2004/10000/High_Femoral_Bone_Mineral_Density_Accretion](https://journals.lww.com/acsm-)
1018 [_in.19.aspx](https://journals.lww.com/acsm-)
- 1019 Vyncke, K., Cruz Fernandez, E., Fajó-Pascual, M., Cuenca-García, M., De
1020 Keyzer, W., Gonzalez-Gross, M., Moreno, L. A., Beghin, L., Breidenassel,
1021 C., Kersting, M., Albers, U., Diethelm, K., Mouratidou, T., Grammatikaki, E.,
1022 De Vriendt, T., Marcos, A., Bammann, K., Börnhorst, C., Leclercq, C., ...
1023 Huybrechts, I. (2013). Validation of the Diet Quality Index for Adolescents
1024 by comparison with biomarkers, nutrient and food intakes: The HELENA
1025 study. *British Journal of Nutrition*, 109(11), 2067–2078.
1026 <https://doi.org/10.1017/S000711451200414X>
- 1027 Wells, J. C. K. (2000). A Hattori chart analysis of body mass index in infants and
1028 children. *International Journal of Obesity*, 24(3), 325–329.
1029 <https://doi.org/10.1038/sj.ijo.0801132>
- 1030 Wells, J. C. K., Cole, T. J., & team, A. study. (2002). Adjustment of fat-free
1031 mass and fat mass for height in children aged 8 y. *International Journal Of*
1032 *Obesity*, 26, 947. <https://doi.org/10.1038/sj.ijo.0802027>
- 1033 Westerterp, K. R., Yamada, Y., Sagayama, H., Ainslie, P. N., Andersen, L. F., &
1034 Anderson, L. J. (2021). *Physical activity and fat-free mass during growth*
1035 *and in later life*. 1–7.
- 1036 Wickel, E. E., & Eisenmann, J. C. (2007). Contribution of youth sport to total
1037 daily physical activity among 6- to 12-yr-old boys. *Medicine and Science in*
1038 *Sports and Exercise*, 39(9), 1493–1500.
1039 <https://doi.org/10.1249/mss.0b013e318093f56a>
- 1040 World Health Organization. (2006). *WHO child growth standards: length/height-*
1041 *for-age, weight-for-age, weight-for-length, weight-for-height and body mass*
1042 *index-for-age: methods and development*.
- 1043 World Health Organization. (2020). WHO Guidelines on physical activity and
1044 sedentary behaviour. In *World Health Organization*.
1045 [https://apps.who.int/iris/bitstream/handle/10665/325147/WHO-NMH-PND-](https://apps.who.int/iris/bitstream/handle/10665/325147/WHO-NMH-PND-2019.4-eng.pdf?sequence=1&isAllowed=y%0Ahttp://www.who.int/iris/handle/1066)
1046 [2019.4-](https://apps.who.int/iris/bitstream/handle/10665/325147/WHO-NMH-PND-2019.4-eng.pdf?sequence=1&isAllowed=y%0Ahttp://www.who.int/iris/handle/1066)
1047 [eng.pdf?sequence=1&isAllowed=y%0Ahttp://www.who.int/iris/handle/1066](https://apps.who.int/iris/bitstream/handle/10665/325147/WHO-NMH-PND-2019.4-eng.pdf?sequence=1&isAllowed=y%0Ahttp://www.who.int/iris/handle/1066)

1048 5/311664%0Ahttps://apps.who.int/iris/handle/10665/325147
1
2 1049 Wszyńska, J., Matłosz, P., Szybisty, A., Lenik, P., Dereń, K., Mazur, A., &
3
4 1050 Herbert, J. (2020). Obesity and Body Composition in Preschool Children
5
6 1051 with Different Levels of Actigraphy-Derived Physical Activity—A Cross-
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8 1052 Sectional Study. *Journal of Clinical Medicine*, 9(4), 1210.
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10 1053 <https://doi.org/10.3390/jcm9041210>

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Table 1. Main characteristics of the participating children (n= 279).

Characteristics	All n= 279 (M±SD)	Girls n= 129 (M±SD)	Boys n= 150 (M±SD)	P-value
Age (y)	7.5 ± 0.3	7.6 ± 0.4	7.5 ± 0.3	0.41
Anthropometric measurements				
Height (cm)	126.1 ± 5.9	125.6 ± 5.6	125.5 ± 6.0	0.12
Weight (kg) ^a	26.4 (23.6 - 30.0)	26.2 (23.6 - 30.0)	26.7 (23.7 - 30.8)	0.59
BMI (kg/m ²) ^a	16.6 (15.4 - 18.3)	16.7 (15.5 - 18.3)	16.6 (15.4 - 18.5)	0.74
BMI z-score [†]	0.84 ± 1.2	0.76 ± 1.1	0.87 ± 1.3	0.48
BIA				
FFM (kg) ^a	20.5 (18.7 - 22.5)	19.9 (18.5 - 21.8)	20.8 (19.1 - 23.4)	<0.01*
FFMI (kg/m ²)	13.1 ± 1.4	12.7 (12.1 - 13.4)	13.2 (12.4 - 14.2)	<0.01*
DXA				
TLSTM (kg) ^a	18.1 (16.6 – 20.0)	17.5 (16.2 - 18.7)	18.9 (17.0 - 21.0)	<0.01*
TLSTMI (kg/m ²)	11.6 ± 1.1	11.2 ± 1.0	11.9 ± 1.1	<0.01*
ALSTMI (Kg/m ²)	4.7± 0.5	4.5± 0.5	4.8± 0.5	<0.01*
pQCT				
Tibia length (mm)	274.3 ± 18.1	275.8 ±17.3	273.1 ±18.8	0.14

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MCSA (mm ²)	3275.3 ± 496.0	3136.5 ± 473.2	3389.1 ± 488.3	<0.01*
MCSAI (mm ² /m ²)	2057.1 ± 263.1	1996.3 ± 257.6	2112.1 ± 256.5	<0.01*
Physical activity				
Light (min/day)	41.3 ± 11.1	37.0 ± 9.9	45.0 ± 10.8	<0.01*
Moderate (min/day) ^a	29.2 (21.1 - 36.2)	24.3 (18.1 - 32.1)	31.9 (24.9 - 41.1)	<0.01*
Vigorous (min/day) ^a	69.0 (55.6 - 85.0)	59.8 (50.0 - 75.7)	77.5 (64.2 - 92.6)	<0.01*
MVPA (min/day) ^a	97.3 (77.4 - 121.4)	85.9 (67.8 – 108.4)	110.8 (88.0 -134.0)	<0.01*
Total PA (min/day)	269.6 (240.3 – 299.7)	254.0 (231.3 – 285.2)	282.5 (245.9 – 312.7)	<0.01*
Physical fitness				
Handgrip strength (kg)	10.5 ± 2.2	10.1 ± 2.0	10.9 ± 2.1	<0.01*
Standing long jump (cm)	102.7 ± 17.8	98.9 ± 17.0	107.6 ± 17.7	<0.01*

Abbreviation: ALSTMI= Appendicular lean soft tissue mass index; BIA= Bioelectrical impedance; BMI= Body mass index; MCSA= Muscle cross-sectional area; DXA= Dual-energy X-ray absorptiometry; FFM= Fat-free mass; FFMI= Fat-free mass index; MVPA= Moderate-vigorous physical activity; pQCT= quantitative peripheral computed tomography; TLSTM= Total lean soft tissue mass; TLSTMI= Total lean soft tissue mass index

Normally distributed variables are shown as mean ± SD (Student t-test)

^a Non-normally distributed variables are shown as median and interquartile intervals (25th and 75th, U Mann–Whitney)

[†]BMI z-scores were calculated according to the World Health Organization (WHO)

* Significant differences by gender

Significance was set at 0.05 level.

Table 2. Associations between physical activity and total lean soft tissue mass index, muscle cross-sectional area index, and fat-free mass index (n= 279)

Predictors	Girls										Boys									
	Total		Light		Moderate		Vigorous		MVPA		Total		Light		Moderate		Vigorous		MVPA	
	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>	β	<i>p</i>
TLSTMI by DXA																				
Model 1	.272	.004*	.154	.109	.123	.201	.151	.117	.144	.137	.335	.000*	.287	.001*	.093	.289	.203	.020*	.166	.058
Model 2	.275	.004*	.170	.078	.120	.219	.159	.001*	.148	.130	.371	.000*	.302	.001*	.116	.206	.224	.013*	.189	.038*
Model 3	.247	.010*	.151	.111	.111	.248	.143	.001*	.134	.162	.337	.000*	.290	.001*	.081	.367	.200	.023*	.160	.072
MCSAI by pQCT																				
Model 1	.148	.131	.112	.253	-.003	.974	.057	.564	.036	.717	.091	.314	.074	.410	.052	.560	.068	.447	.064	.480
Model 2	.124	.223	.093	.359	-.034	.742	.030	.768	.007	.944	.114	.219	.083	.364	.053	.569	.074	.426	.068	.468
Model 3	.097	.347	.077	.448	-.040	.692	.018	.863	-.004	.972	.110	.245	.081	.381	.049	.607	.070	.454	.063	.500
FFMI by BIA																				
Model 1	.125	.194	.132	.173	-.047	.626	.042	.664	.010	.918	.260	.003*	.161	.064	-.010	.908	.078	.373	.047	.594
Model 2	.185	.053	.149	.122	-.060	.539	.045	.642	.007	.939	.308	.000*	.175	.050	-.026	.781	.078	.389	.042	.648
Model 3	.149	.097	.137	.154	-.066	.496	.035	.717	-.001	.988	.299	.001*	.175	.051	-.026	.780	.079	.388	.042	.647

β : standardized regression coefficient

Abbreviation: BIA= Bioimpedance analysis; DXA= Dual-energy X-ray absorptiometry; FFMI= Fat-free mass index; MCSAI= Muscle cross-sectional area index; pQCT= Peripheral quantitative tomography; TLSTMI= Total lean soft tissue mass index.

Model 1 Physical activity (basic model without adjustments)

Model 2 Model 1 + diet quality index, parents' highest education level and age.

Model 3 Model 2 + birth weight

*Significance was set at 0.05 level.

Table 3. Associations between physical fitness and total lean soft tissue mass index, muscle cross-sectional area index, and fat-free mass index (n= 279)

Predictors Physical fitness	Girls				Boys				
	Handgrip strength		Standing long jump		Handgrip strength		Standing long jump		
	β	p	β	p	β	p	β	p	
TLSTMI by DXA									
<i>Model 1</i>	.363	.000*	.055	.512	.478	.000*	-.019	.839	
<i>Model 2</i>	.350	.000*	.070	.403	.464	.000*	-.013	.893	
<i>Model 3</i>	.314	.000*	.034	.672	.425	.000*	-.036	.690	
MCSAI by pQCT									
<i>Model 1</i>	.262	.002*	.056	.520	.273	.000*	-.064	.508	
<i>Model 2</i>	.282	.001*	.074	.404	.287	.000*	-.060	.540	
<i>Model 3</i>	.267	.002*	.051	.561	.301	.000*	-.059	.547	
FFMI by BIA									
<i>Model 1</i>	.236	.004*	-.082	.332	.289	.000*	-.084	.367	
<i>Model 2</i>	.218	.008*	-.065	.430	.307	.000*	-.083	.367	
<i>Model 3</i>	.196	.017*	-.088	.281	.325	.000*	-.082	.384	

β : standardized regression coefficient

Abbreviation: BIA= Bioimpedance analysis, DXA= Dual-energy X-ray absorptiometry, FFMI= Fat-free mass index, MCSAI= Muscle cross-sectional area index, pQCT= Peripheral quantitative computed tomography and TLSTMI= Total lean soft tissue mass index.

Model 1 physical fitness (basic model without adjustments)

Model 2 *Model 1* + diet quality index, parents' highest education level and age.

Model 3 *Model 2* + birth weight

*Significance was set at 0.05 level.

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Figure 1. Serial multiple mediation model of the association between total physical activity and total lean soft tissue mass index, using handgrip strength as mediator, controlling for gender.

