

# The Influence of Body Position on Bioelectrical Impedance Spectroscopy Measurements in Young Children

**Jaz Lyons-Reid**

University of Auckland

**Leigh C Ward**

University of Queensland

**Mya-Thway Tint**

Agency for Science, Technology and Research

**Timothy Kenealy**

University of Auckland

**Keith M Godfrey**

University of Southampton

**Shiao-Yng Chan**

Agency for Science, Technology and Research

**Wayne S Cutfield** (✉ [w.cutfield@auckland.ac.nz](mailto:w.cutfield@auckland.ac.nz))

University of Auckland

---

## Research Article

**Keywords:** influence, bioelectrical, spectroscopy, young children

**Posted Date:** March 22nd, 2021

**DOI:** <https://doi.org/10.21203/rs.3.rs-310989/v1>

**License:** © ⓘ This work is licensed under a Creative Commons Attribution 4.0 International License.

[Read Full License](#)

---

# 1 **The influence of body position on bioelectrical impedance** 2 **spectroscopy measurements in young children**

3 **Jaz Lyons-Reid<sup>1</sup>, Leigh C Ward<sup>1,2</sup>, Mya-Thway Tint<sup>3,4</sup>, Timothy Kenealy<sup>1,5</sup>, Keith M**  
4 **Godfrey<sup>6,7</sup>, Shiao-Yng Chan<sup>3,4</sup>, Wayne S Cutfield<sup>1,8\*</sup>**

5

6 <sup>1</sup>Liggins Institute, University of Auckland, Auckland, New Zealand

7 <sup>2</sup>School of Chemistry and Molecular Biosciences, The University of Queensland, Brisbane, Australia

8 <sup>3</sup>Singapore Institute for Clinical Sciences, Agency for Science, Technology and Research (A\*STAR), Singapore

9 <sup>4</sup>Department of Obstetrics & Gynaecology, Yong Loo Lin School of Medicine, National University of Singapore,  
10 Singapore

11 <sup>5</sup>Department of Medicine and Department of General Practice and Primary Health Care, University of Auckland,  
12 Auckland, New Zealand

13 <sup>6</sup>MRC Lifecourse Epidemiology Unit, University of Southampton, Southampton, United Kingdom

14 <sup>7</sup>NIHR Southampton Biomedical Research Centre, University of Southampton and University Hospital  
15 Southampton NHS Foundation Trust, Southampton, United Kingdom

16 <sup>8</sup>A Better Start – National Science Challenge, University of Auckland, Auckland, New Zealand

17 \*w.cutfield@auckland.ac.nz

18

## 19 **ABSTRACT**

20 Bioelectrical impedance techniques are easy to use and portable tools for assessing body  
21 composition. While measurements vary according to standing vs supine position in adults,  
22 and fasting and bladder voiding have been proposed as additional important influences, these  
23 have not been assessed in young children. Therefore, the influence of position, fasting, and  
24 voiding on bioimpedance measurements was examined in children. Bioimpedance  
25 measurements (ImpediMed SFB7) were made in 50 children (3.5 years). Measurements were  
26 made when supine and twice when standing (immediately on standing and after four minutes).  
27 Impedance and body composition were compared between positions, and the effect of fasting  
28 and voiding was assessed. Impedance varied between positions, but body composition  
29 parameters other than fat mass (total body water, intra- and extra-cellular water, fat-free mass)  
30 differed by less than 5%. There were no differences according to time of last meal or void.  
31 Equations were developed to allow standing measurements of fat mass to be combined with  
32 supine measurements. In early childhood, it can be difficult to meet requirements for fasting,

33 voiding, and lying supine prior to measurement. This study provides evidence to enable  
34 standing and supine bioimpedance measurements to be combined in cohorts of young  
35 children.

36

## 37 **Introduction**

38 Bioelectrical impedance techniques allow quick, easy measurement of body composition  
39 including, total body water (TBW), fat mass (FM), and fat-free mass (FFM). Multi-frequency  
40 techniques, including multi-frequency bioimpedance analysis (MF BIA) and bioimpedance  
41 spectroscopy (BIS), are further able to distinguish between intracellular (ICW) and  
42 extracellular fluids (ECW)<sup>1,2</sup>. Although not widely used in early childhood, bioimpedance  
43 techniques are easy to administer, are inexpensive, and require less co-operation from the  
44 child compared to other widely used methods, such as dual-energy X-ray absorptiometry  
45 (DXA). However, there are many factors that may influence bioimpedance measurements and  
46 thus require standardisation<sup>3</sup>. These factors may be amplified in infants and young children,  
47 where compliance is a particular challenge<sup>4</sup>. One such factor is the requirement for children  
48 to lie supine for extended periods prior to measurement.

49

50 Brantlov et al.<sup>5</sup> reported that of 71 studies identified which used bioelectrical impedance  
51 analysis to estimate body composition in populations of healthy children, authors did not  
52 consistently report in what body position (i.e., standing or supine) measurements were  
53 obtained. Of concern, only 21% reported how long the child was in the position prior to  
54 measurement. In adults, it has been shown that standing and supine measurements are not  
55 interchangeable, and that it takes approximately 5 minutes for fluid stabilisation to occur to  
56 allow measurement of TBW<sup>6</sup>, and extended periods to establish ECW and ICW stabilisation<sup>6,7</sup>,  
57 which correspond to changes in impedance values<sup>8</sup>. As such, adult guidelines recommend  
58 that bioimpedance measurements be made in the supine position after 4 to 10 minutes have  
59 elapsed<sup>9,10</sup>. However, no guidelines exist for paediatric populations.

60  
61  
62  
63  
64  
65  
66  
67  
68  
69  
70  
71

While studies in young children have explored some of the factors which influence impedance measurements, such as movement<sup>11</sup> and electrode placement<sup>11-13</sup>, no study has evaluated the effect of body position. In young children, it may be more feasible to obtain bioimpedance measurements while the child is standing; however, it is unclear whether measurements taken in alternate body positions are interchangeable. In addition to recommendations about body position, adult guidelines state that bioimpedance measurements should be made when the subject is fasted and has voided their bladder<sup>9,10</sup>; however, it is unclear what effect, if any, these factors may have on measurements in young children. Therefore, the aim of this study was to determine whether BIS measurements obtained in different body positions can be used interchangeably, and whether fasting and bladder voiding influence associations.

## 72 **Methods**

### 73 **Subjects**

74 A convenience sample of children aged 3.5 years was selected from the Auckland site of the  
75 Nutritional Intervention Preconception and During Pregnancy to Maintain Healthy Glucose  
76 Metabolism and Offspring Health (“NiPPeR”) study<sup>14</sup>. Data were obtained from 50 children  
77 selected based on compliance with the NiPPeR BIS protocol (i.e., the child laid supine for □4  
78 minutes prior to the initial measurement).

79

### 80 **Ethics**

81 The NiPPeR trial was registered on 16 July 2015 with ClinicalTrials.gov (NCT02509988,  
82 Universal Trial Number U1111-1171-8056); ethics approval was granted by the Northern A  
83 Health and Disability Ethics Committee (15/NTA/21/AM20). Written informed consent was  
84 obtained from the parents/guardians of the study subjects. All procedures in this study were  
85 conducted according to the ethical principles and guidelines laid down in the Declaration of  
86 Helsinki<sup>15</sup>.

87

**88 Bioelectrical impedance spectroscopy**

89 Bioimpedance measurements were made with the ImpediMed SFB7 device (ImpediMed,  
90 Brisbane, Australia). This device measures bioimpedance parameters over a frequency range  
91 of 3 to 1000 kHz, resulting in 256 measurements per assessment<sup>16</sup>. Instrument calibration  
92 was checked daily prior to use using a test cell provided by the manufacturer. ImpediMed  
93 single-tab gel electrodes (25 × 23 mm) were used to attach sense leads to the left or right  
94 dorsum wrist and ankle, and the source leads to the palm at the metacarpal heads and the  
95 sole at the metatarsal heads on the same side of the body<sup>17</sup>. No differences in impedance  
96 parameters were observed between measurement sides (all  $p > 0.05$ ). Prior to careful  
97 application of the electrodes, the skin was cleaned with 70% isopropyl alcohol wipes and  
98 allowed to dry. Any clothing with metal (e.g., clips or buckles) was removed prior to  
99 measurement to avoid electrical interference. Otherwise, clothing was only removed to access  
100 electrode sites. For each body position, measurements were made in triplicate using the  
101 “continuous” setting of the device. Cole plots were examined to ensure data quality and  
102 measurements were repeated if movement occurred.

103

104 Data was analysed using Biolmp software version 5.4.0.3 (ImpediMed), using the default  
105 settings [frequency range 5–500 kHz, automatic time delay (Td) correction on, no data  
106 rejection limit]. The impedance values of interest were as follows:

107 1. Resistance at zero frequency,  $R_0$

108 At low frequencies, the cell membrane acts as an imperfect capacitor and current  
109 cannot be passed, and therefore the resistance measured is from ECW only.

110

111 2. Resistance at infinite frequency,  $R_\infty$

112 This value is indicative of TBW (ECW + ICW) as at high frequencies the electrical  
113 current can pass across the cell membrane and ICW as well as ECW can be  
114 measured.

115

116 3. Impedance at 50 kHz,  $Z_{50}$

117 Most SFBIA devices measure impedance at this frequency to predict TBW and FFM.

118 At this frequency both ECW and ICW are represented, although ECW still  
119 predominates

120

121 4. Resistance at 50 kHz,  $R_{50}$

122 Resistance is the component of  $Z_{50}$  that is related to TBW.

123

124 5. Reactance at 50 kHz,  $X_{C50}$

125 Reactance is the component of  $Z_{50}$  that is related to cell membrane capacitance.

126

127 6. Impedance at the characteristic frequency,  $Z_c$

128 The characteristic frequency ( $f_c$ ) is the frequency where reactance is maximal in an  
129 individual. At this frequency, the ratio of current flow through extra- and intracellular  
130 paths is independent of the membrane capacitance<sup>18</sup>.  $Z_c$  has therefore been suggested  
131 to be an appropriate predictor of TBW<sup>1</sup>.

132

### 133 **Assessment of body composition**

134 Standing height was measured in triplicate to the nearest 0.1 cm using a calibrated SECA 213  
135 portable stadiometer (SECA, Hamburg, Germany), with median height being used in analyses,  
136 while a single weight measurement was obtained to the nearest 100 g using calibrated SECA  
137 899 scales. Along with sex, these values were used to compute body composition measures  
138 using two methods: mixture theory in combination with Cole modelling [i.e., the SFB7's default

139 equations and constants: resistivity of ECW ( $\rho_{ECW}$ ) and ICW ( $\rho_{ICW}$ ) – females 235.5 and  
 140 894.2  $\Omega/cm$ , and males 273.9 and 937.2  $\Omega/cm$ , respectively; body density (Db) 1.05 g/L; body  
 141 proportion factor (Kb) 4.30; and hydration factor (HF) 0.732]<sup>16,19,20</sup>, and an empirically-derived  
 142 regression equation<sup>21</sup>.

143

144 The SFB7 provides the following body composition values: TBW, ECW, ICW, FFM, and FM.

145

146 The empirically derived regression equation for FFM ( $FFM_{Rush}$ ) was developed using DXA  
 147 among a cohort of New Zealand 2-year-olds<sup>21</sup>. The reported equation is as follows:

$$148 \quad FFM_{Rush} \text{ (kg)} = 0.367 \frac{\text{height (cm)}^2}{\text{resistance}} + 0.188 \text{ weight (kg)} + 0.077 \text{ height (cm)} + 0.273 \text{ sex (male}$$

$$149 \quad \quad \quad = 1, \text{ female} = 0)$$

150

151 FM ( $FM_{Rush}$ ) was computed from FFM considering a two-compartment model of body  
 152 composition<sup>22</sup> and the following equation:

$$153 \quad \quad \quad FM_{Rush} \text{ (kg)} = \text{Weight (kg)} - FFM_{Rush} \text{ (kg)}$$

154

## 155 **Experimental design**

156 Children were measured in three body positions. First, as per adult guidelines<sup>9,10</sup>, children  
 157 were measured supine on non-conductive examination tables with the legs separated and  
 158 arms by their sides without skin-to-skin contact between arms and the trunk, after at least four  
 159 minutes had elapsed (thus allowing fluid stabilisation). Second, the children were measured  
 160 immediately (within one minute) on standing (from being supine) while maintaining correct  
 161 abduction of the arms and legs. Finally, children were measured in the same standing position  
 162 after at least four minutes had elapsed. During this period, children were required to remain  
 163 upright (standing or seated). For each body position electrode placement remained the same  
 164 and it was ensured that the leads were not tangled or touching any metal surfaces. It was not

165 possible to ensure that the leads were not touching the ground during the standing  
166 measurements due to the placement of the electrodes.

167 In addition, whether the child had fasted or voided their bladder was recorded. The effect of  
168 consumption of food or drink on impedance measurements has not been explored in  
169 preschool aged children. Evidence from infancy suggests that it is time after consumption,  
170 rather than volume, that is important<sup>11</sup>. Thus, time of last meal or drink (>2 hr ago, 1–2 hr ago,  
171 30 min – 1 hr ago, or  $\leq$ 30 min ago) was recorded, as was time of last void. Time of last void  
172 was categorised according to whether or not the child had voided their bladder within half an  
173 hour of measurement. If the child consumed any food or fluid, or voided their bladder between  
174 measurement positions, this was recorded. These children were excluded from analyses  
175 evaluating the effect of fasting and voiding on differences in impedance between body  
176 positions ( $n = 5$ ).

177

## 178 **Statistical methods**

179 Mean (SD) bioimpedance parameters ( $R_{\infty}$ ,  $R_0$ ,  $Z_c$ ,  $R_{50}$ ,  $Z_{50}$ , and  $Xc_{50}$ ) and body composition  
180 values ( $TBW_{SFB7}$ ,  $ECW_{SFB7}$ ,  $ICW_{SFB7}$ ,  $FFM_{SFB7}$ ,  $FM_{SFB7}$ ,  $FFM_{Rush}$ , and  $FM_{Rush}$ ) were assessed  
181 in each of the body positions (supine, standing  $\leq$ 1 min, and standing  $\leq$ 4 min), with sex  
182 differences in impedance parameters being explored using independent samples  $t$  tests.  
183 Differences in impedance and body composition between supine and both standing positions  
184 was assessed using repeated measures ANOVA with Bonferroni post hoc testing, with  
185 differences in body composition values between supine and standing ( $\leq$ 4 min) positions being  
186 presented as percentage differences. The effect of fasting and bladder voiding on differences  
187 in impedance measurements was assessed using one-way ANOVA and independent samples  
188  $t$  tests.

189

190 In order to develop equations to allow adjustment of bioimpedance parameters obtained while  
191 standing, thus allowing their use in equations where supine body position is indicated, the  
192 cohort was split into development (70%) and validation cohorts (30%) using a random number  
193 generator within SPSS version 26 (IBM Corp, Armonk, NY, USA). Among the development  
194 cohort ( $n = 35$ ), for each impedance parameter simple linear regression was used to develop  
195 an equation to adjust impedance values obtained while standing ( $\square 4$  min) to be comparable  
196 to those obtained while supine. These resulting equations were then applied to the validation  
197 cohort ( $n = 15$ ). The equations were also applied to standing ( $\square 1$  min) measurements among  
198 the validation cohort to further elucidate the importance of time spent standing. Impedance  
199 parameters from supine measurements were compared to the adjusted standing  
200 measurements using paired samples  $t$  tests and Bland-Altman's methods<sup>23</sup>. All tests were two-  
201 tailed and were performed within SPSS.  $P$  values  $\leq 0.05$  were considered statistically  
202 significant.

203

## 204 **Results**

### 205 **Demographics**

206 The sample comprised 50 children, 20 of whom were male and 30 female. On average, the  
207 children were 3.38 years old, with boys being somewhat taller and heavier than girls (Table  
208 1).

209

### 210 **Sex effects**

211 Mean impedance parameters were larger among girls than boys in each of the body positions.  
212 These differences were significant, with the exception of standing ( $\square 4$  minutes) mean  
213 reactance at 50 kHz ( $p = 0.065$ ). In contrast, the means of the differences in impedance  
214 parameters between supine and standing ( $\square 4$  minutes) positions were not significantly  
215 different between sexes, with the exception of reactance at 50 kHz ( $p < 0.001$ ). Given the  
216 similarity in all other mean differences, further comparisons were made using the entire cohort.

217

**218 Differences between standing and supine**

219 Mean impedance parameters for supine and standing (<1 minute and □4 minutes)  
220 measurements are presented in Table 2. There were significant differences between body  
221 positions in all impedance parameters ( $p < 0.001$ ). Post-hoc comparisons revealed that there  
222 were differences between impedance parameters obtained when supine compared to both  
223 standing positions (all  $p < 0.001$ ), with supine values larger than those obtained when standing.  
224 Impedance parameters were generally higher when obtained standing immediately from  
225 supine (<1 minute) compared to standing (□4 minutes), with the exception of reactance at 50  
226 kHz where the reverse was true, but these differences were not statistically significant. There  
227 were also significant differences ( $p < 0.001$ ) in all body composition parameters between  
228 supine and both standing positions (Table 3). However, these differences were probably of  
229 little clinical significance, with percentage differences of less than five percent, with the  
230 exception of FM, which exhibited both greater percentage differences and greater variability.

231

**232 Effect of fasting and voiding**

233 Among children who did not eat, drink, or void between measurements ( $n = 45$ ), there was no  
234 clear pattern (i.e., increasing or decreasing across categories) in mean impedance values  
235 according to category of last meal (<30 min ago, 30 min – 1 hr ago, 1–2 hr ago, or >2 hr ago).  
236 Furthermore, differences in impedance between standing (□4 minutes) and supine  
237 measurements (i.e., mean differences) did not vary significantly according to category of last  
238 meal ( $p$  values:  $R_0 = 0.94$ ,  $R_\infty = 0.30$ ,  $Z_c = 0.64$ ,  $Z_{50} = 0.80$ ,  $R_{50} = 0.79$ ,  $X_{C_{50}} = 0.59$ ). However,  
239 most of the children consumed food within half an hour of measurement, therefore, the groups  
240 30 min to 1 hr ( $n = 7$ ), 1 to 2 hr ( $n = 9$ ), and over 2 hr ( $n = 5$ ) were collapsed, and differences  
241 were assessed using an independent samples  $t$  test. Although there was a trend for greater  
242 impedance and resistance, but reduced reactance among those who had not eaten within half  
243 an hour of measurement, there remained no significant differences in mean impedance

244 parameters (all  $p > 0.10$ ), or in mean differences in impedance parameters between supine  
245 and standing ( $\approx 4$  min) positions ( $p$  values:  $R_0 = 0.70$ ,  $R_\infty = 0.86$ ,  $Z_c = 0.74$ ,  $Z_{50} = 0.58$ ,  $R_{50} =$   
246  $0.58$ ,  $X_{c50} = 0.83$ ).

247

248 Mean impedance parameters were higher among those who had not voided within half an  
249 hour of measurement, compared to those who had; however, these differences were not  
250 statistically significant (all  $p > 0.50$ ). Likewise, there were no significant variations in the mean  
251 differences of impedance parameters according to whether or not the child had voided ( $p$   
252 values:  $R_0 = 0.55$ ,  $R_\infty = 0.16$ ,  $Z_c = 0.71$ ,  $Z_{50} = 0.84$ ,  $R_{50} = 0.86$ ). Although, there was a  
253 borderline significant difference in reactance at 50 kHz, with mean differences being higher  
254 among those who had not voided, compared to those who had ( $p = 0.062$ ).

255

### 256 **Adjustment equations**

257 As there were statistically significant differences between supine and standing positions,  
258 equations were developed to allow impedance measurements obtained when standing to be  
259 adjusted to be comparable to those obtained while supine (Table 4). The development cohort  
260 ( $n = 35$ ) was not different from the validation cohort ( $n = 15$ ) in age, sex, height, weight, or BMI  
261  $z$  score (all  $p > 0.05$ ).

262

263 When the adjustment equations were applied to the validation cohort, there were no significant  
264 differences in mean impedance values between supine and adjusted standing measurements  
265 (all  $p > 0.05$ ). Bland-Altman analysis revealed small biases and narrow limits of agreement.  
266 These are expressed as absolute values and as percentages of mean supine impedance  
267 values (Table 5). The equations were subsequently applied to standing ( $\leq 1$  minute)  
268 measurements, and there were no significant differences between the adjusted and supine  
269 values (all  $p > 0.05$ ). Bias was larger, but was still less than 1% of mean supine impedance;  
270 however, limits of agreement were marginally narrower (supplementary Table 1).

271

## 272 Discussion

273 Although adult guidelines dictate that BIA measurements be made supine after at least 4 min  
274 have elapsed<sup>9,10</sup>, it is not always feasible in infants and young children. In our study of 50  
275 young children, impedance measurements differed between body positions, with higher  
276 derived TBW, ECW, ICW, and FFM, and lower FM in the standing body position; most of the  
277 body composition values differed by less than 5%, with the exception of FM ( $FM_{\text{SFB7}}$  13.75%  
278 lower and  $FM_{\text{Rush}}$  9.12% lower).

279

280 A recent study evaluated the effect of body position on phase angle in a cohort of 1298  
281 Mexican children and adolescents aged 4 to 20 years<sup>24</sup>. Phase angle was higher when  
282 measured supine than standing, with differences between body positions increasing with  
283 increased phase angle, age, and height. However, the children were measured with two  
284 different BIA devices, which had different electrode types (metal and adhesive), and thus are  
285 not directly comparable.

286

287 Another study examined differences in body fluid according to measurement position  
288 (standing and supine) in a cohort of 23 boys (6–14 years) and 26 men (23–82 years)<sup>25</sup>.  
289 Significant impedance differences were also observed (at 50 and 100 kHz in boys, and at 100  
290 kHz in men). No significant differences were seen in TBW, FFM, FM, or percentage of body  
291 fat (%BF), but body water shifted so that ECW increased and ICW decreased when standing.  
292 This is in contrast to our study, where differences were observed in all body composition  
293 values, and both ECW and ICW increased when standing. In adults, Gibson et al.<sup>6</sup> found that  
294 ECW decreased and ICW increased while supine. When standing, although ECW increased  
295 incrementally, decreases in ICW were not significant. It has been suggested that it takes  
296 extended periods to achieve fluid stabilisation<sup>6,7</sup>, which may explain this observed  
297 discrepancy.

298

299 We were unable to explore time-course changes in impedance values, however, previous  
300 research has suggested that changes in impedance are greatest immediately on recumbence/  
301 standing, and changes thereafter are gradual<sup>26,27</sup>. Furthermore, we observed no significant  
302 differences in impedance values when measured immediately on standing, compared to after  
303 at least four minutes had elapsed.

304

305 Regression equations were developed to allow adjustment of standing BIA measurements to  
306 be comparable to measurements obtained while supine, irrespective of the amount of time  
307 spent standing (Tables 5 and S1). Previously, regression equations have been developed  
308 among adults to allow measurements made while sitting upright in a wheelchair to be  
309 comparable to measurements made while supine<sup>28</sup>. Similarly, Rush et al.<sup>29</sup> developed  
310 adjustment factors to convert standing measurement to equate supine in children and adults  
311 (categorised: 5–14 years, 15–30 years, 31–59 years, and 60+ years). Our equations may be  
312 of benefit in studies in young children that wish to use a previously published prediction  
313 equation where supine body position is dictated, but where this may not be achievable.

314

315 To our knowledge, this study is the first to explore the influence of body position on  
316 bioimpedance measurements in young children (<5 years). At this age, children are often non-  
317 compliant, and it is not feasible to obtain BIA measurements after extended periods of lying.  
318 It may be of benefit to take measurements while the child is in an alternative body position, for  
319 example, while standing. A limitation of our study was that electrode placement meant that the  
320 leads were touching the ground. Although, only the external insulating plastic sheath was in  
321 contact and the leads are actively shielded against electrical interference. Furthermore, the  
322 placement used was necessary to maintain adequate separation of the electrodes<sup>9,10</sup>. This  
323 methodology meant that use of two different BIA devices was avoided. Although some studies  
324 have evaluated the effect of body position using different BIA devices<sup>24,30</sup>, ample evidence  
325 suggests that BIA device types are not interchangeable<sup>31-33</sup>. Jensen et al.<sup>24</sup> used two differing  
326 BIA devices in their study, and concluded that electrode type explained approximately half of

327 the observed differences in phase angle between body positions when they conducted  
328 additional analyses in a cohort of adults. However, this is likely related to the differing electrode  
329 positions, in addition to the electrode type (metal vs adhesive)

330

331 The effect of fasting has not previously been evaluated in preschool aged children, however,  
332 evidence from infancy suggests that impedance parameters do not change significantly when  
333 measured pre- and post-feed<sup>11,34</sup>. Although, Sesmero et al.<sup>11</sup> did observe a general trend for  
334 increasing  $R_0$  with increasing time after feed, but this was only significant among their 1-week-  
335 old infants. The effect of bladder voiding has not been evaluated in any paediatric population.  
336 In adults, bladder voiding has been associated with a small measurement error of 1.0%<sup>35</sup>. In  
337 this study, time of last meal or bladder void were often estimated; however, there were no  
338 significant differences in impedance between body positions according to fasting or voiding.  
339 Nonetheless, half an hour may not be a sufficient difference in time to evaluate the effect of  
340 fasting and voiding. However, it would be not be feasible nor ethical to request young children  
341 to refrain from eating or voiding for extended periods to evaluate this further, though a larger  
342 study group may provide more clarity on this issue.

343

344 Other limitations of this research include that the equations used to estimate body composition  
345 might not be appropriate for this cohort, as evidenced by the wide standard deviations for FM.  
346 However, the aim of the study was not to accurately estimate body composition; rather, body  
347 composition values were used to ascertain if clinically significant differences were apparent  
348 between body positions. Nonetheless, we used two different methods for estimating body  
349 composition (Rush et al.<sup>21</sup> and SFB7 equations), and the resulting percentage differences  
350 between body positions were comparable. In addition, we did not randomise the order of  
351 measurements as inclusion into this sub-study was based on compliance with the NiPPeR  
352 protocol. Studies in adults have suggested that position order is not important<sup>6,29,36</sup>. For  
353 example, among children and adults assessed both standing prior to lying supine and standing

354 following a supine measurement, the second standing measurement was lower than the first  
355 by only approximately 1 ohm<sup>29</sup>.

356

357 This study provides the first evidence to describe the influence of body position on  
358 bioimpedance measurements in young children. This study suggests that researchers and  
359 clinicians can take bioimpedance measurements without requiring the child to meet various  
360 requirements for fasting, voiding, and lying supine for extended periods. Future research is  
361 required to confirm these findings, and to further evaluate the effect of fasting and voiding on  
362 bioimpedance measurements in young children.

363

#### 364 **Data availability**

365 The data required to reproduce these findings cannot be shared at this time as the data also  
366 forms part of an ongoing study.

## References

- 1 Cornish, B. H., Thomas, B. J. & Ward, L. C. Improved prediction of extracellular and total body water using impedance loci generated by multiple frequency bioelectrical impedance analysis. *Phys Med Biol* **38**, 337-346, doi:10.1088/0031-9155/38/3/001 (1993).
- 2 De Lorenzo, A., Andreoli, A., Matthie, J. & Withers, P. Predicting body cell mass with bioimpedance by using theoretical methods: a technological review. *J Appl Physiol* **82**, 1542-1558, doi:10.1152/jappl.1997.82.5.1542 (1997).
- 3 Brantlov, S., Ward, L. C., Jodal, L., Rittig, S. & Lange, A. Critical factors and their impact on bioelectrical impedance analysis in children: a review. *J Med Eng Technol* **41**, 22-35, doi:10.1080/03091902.2016.1209590 (2017).
- 4 Lyons-Reid, J., Ward, L. C., Kenealy, T. & Cutfield, W. Bioelectrical impedance analysis—an easy tool for quantifying body composition in infancy? *Nutrients* **12**, 920, doi:10.3390/nu12040920 (2020).
- 5 Brantlov, S., Jodal, L., Lange, A., Rittig, S. & Ward, L. C. Standardisation of bioelectrical impedance analysis for the estimation of body composition in healthy paediatric populations: a systematic review. *J Med Eng Technol* **41**, 460-479, doi:10.1080/03091902.2017.1333165 (2017).
- 6 Gibson, A., Beam, J., Alencar, M., Zuhl, M. & Mermier, C. Time course of supine and standing shifts in total body, intracellular and extracellular water for a sample of healthy adults. *Eur J Clin Nutr* **69**, 14-19, doi:10.1038/ejcn.2013.269 (2014).
- 7 Segal, K. R., Van Loan, M., Fitzgerald, P. I., Hodgdon, J. A. & Van Itallie, T. B. Lean body mass estimation by bioelectrical impedance analysis: a four-site cross-validation study. *Am J Clin Nutr* **47**, 7-14, doi:10.1093/ajcn/47.1.7 (1988).
- 8 Kushner, R. F., Gudivaka, R. & Schoeller, D. A. Clinical characteristics influencing bioelectrical impedance analysis measurements. *Am J Clin Nutr* **64**, 423S-427S, doi:10.1093/ajcn/64.3.423S (1996).
- 9 Kyle, U. G. *et al.* Bioelectrical impedance analysis-part II: utilization in clinical practice. *Clin Nutr* **23**, 1430-1453, doi:10.1016/j.clnu.2004.09.012 (2004).
- 10 National Institutes of Health. Bioelectrical impedance analysis in body composition measurement: National Institutes of Health Technology Assessment Conference Statement. *Am J Clin Nutr* **64**, 524S-532S, doi:10.1093/ajcn/64.3.524S (1996).
- 11 Sesmero, M. A., Mazariegos, M., Pedrón, C., Jones, J. & Solomons, N. W. Bioimpedance electrical spectroscopy in the first six months of life: some methodologic considerations. *Nutrition* **21**, 567-573, doi:10.1016/j.nut.2004.10.010 (2005).
- 12 Raghavan, C. V. *et al.* Estimation of total body water in very-low-birth-weight infants by using anthropometry with and without bioelectrical impedance and H<sub>2</sub>[(18)O]. *Am J Clin Nutr* **68**, 668-674, doi:10.1093/ajcn/68.3.668 (1998).
- 13 Sidhu, J. S., Triggs, E. J., Charles, B. G. & Tudehope, D. I. Electrode placement in neonatal bioelectrical impedance analysis. *Med Biol Eng Comput* **32**, 456-459, doi:10.1007/BF02524702 (1994).
- 14 Godfrey, K. M. *et al.* Nutritional Intervention Preconception and During Pregnancy to Maintain Healthy Glucose Metabolism and Offspring Health ("NiPPeR"): study protocol for a randomised controlled trial. *Trials* **18**, 131, doi:10.1186/s13063-017-1875-x (2017).
- 15 World Medical Association. World Medical Association Declaration of Helsinki: Ethical Principles for Medical Research Involving Human Subjects. *JAMA* **310**, 2191-2194, doi:10.1001/jama.2013.281053 (2013).
- 16 ImpediMed. SFB7, <<https://www.impedimed.com/products/sfb7/>> (2020).
- 17 Lingwood, B. E., Coghlan, J. P., Ward, L. C., Charles, B. G. & Colditz, P. B. Measurement of extracellular fluid volume in the neonate using multiple frequency bio-impedance analysis. *Physiol Meas* **21**, 251-262 (2000).

- 18 Lingwood, B. E. *et al.* Prediction of fat-free mass and percentage of body fat in neonates using bioelectrical impedance analysis and anthropometric measures: validation against the PEA POD. *Br J Nutr* **107**, 1545-1552, doi:10.1017/S0007114511004624 (2012).
- 19 Cole, K. S. Permeability and impermeability of cell membranes for ions. *Cold Spring Harbor Symp Quant Biol* **8**, 110-122, doi:10.1101/SQB.1940.008.01.013 (1940).
- 20 Matthie, J. R. Bioimpedance measurements of human body composition: critical analysis and outlook. *Expert Rev Med Devices* **5**, 239-261, doi:10.1586/17434440.5.2.239 (2008).
- 21 Rush, E. C., Bristow, S., Plank, L. D. & Rowan, J. Bioimpedance prediction of fat-free mass from dual-energy X-ray absorptiometry in a multi-ethnic group of 2-year-old children. *Eur J Clin Nutr* **67**, 214-217, doi:10.1038/ejcn.2012.182 (2013).
- 22 Heymsfield, S. B., Wang, Z., Baumgartner, R. N. & Ross, R. Human body composition: advances in models and methods. *Annu Rev Nutr* **17**, 527-558, doi:10.1146/annurev.nutr.17.1.527 (1997).
- 23 Bland, J. M. & Altman, D. G. Statistical methods for assessing agreement between two methods of clinical measurement. *Lancet* **1**, 307-310 (1986).
- 24 Jensen, B. *et al.* Configuration of bioelectrical impedance measurements affects results for phase angle. *Med Eng Phys* **84**, 10-15, doi:10.1016/j.medengphy.2020.07.021 (2020).
- 25 Kagawa, M., Wishart, C. & Hills, A. P. Influence of posture and frequency modes in total body water estimation using bioelectrical impedance spectroscopy in boys and adult males. *Nutrients* **6**, 1886-1898, doi:10.3390/nu6051886 (2014).
- 26 Kushner, R. F., Gudivaka, R. & Schoeller, D. A. Clinical characteristics influencing bioelectrical impedance analysis measurements. *Am J Clin Nutr* **64**, 423S-427S, doi:10.1093/ajcn/64.3.423S (1996).
- 27 Slinde, F., Bark, A., Jansson, J. & Rossander-HulthÉN, L. Bioelectrical impedance variation in healthy subjects during 12 h in the supine position. *Clin Nutr* **22**, 153-157, doi:10.1054/clnu.2002.0616 (2003).
- 28 Allison, G. T., Singer, K. P. & Marshall, R. N. The effect of body position on bioelectrical resistance in individuals with spinal cord injury. *Disabil Rehabil* **17**, 424-429, doi:10.3109/09638289509166656 (1995).
- 29 Rush, E. C., Crowley, J., Freitas, I. F. & Luke, A. Validity of hand-to-foot measurement of bioimpedance: standing compared with lying position. *Obesity (Silver Spring)* **14**, 252-257, doi:10.1038/oby.2006.32 (2006).
- 30 Esco, M. R. *et al.* Agreement between supine and standing bioimpedance spectroscopy devices and dual-energy X-ray absorptiometry for body composition determination. *Clin Physiol Funct Imaging* **39**, 355-361, doi:10.1111/cpf.12585 (2019).
- 31 Silva, A. M. *et al.* Lack of agreement of in vivo raw bioimpedance measurements obtained from two single and multi-frequency bioelectrical impedance devices. *Eur J Clin Nutr* **73**, 1077-1083, doi:10.1038/s41430-018-0355-z (2019).
- 32 Tinsley, G. M., Moore, M. L., Silva, A. M. & Sardinha, L. B. Cross-sectional and longitudinal agreement between two multifrequency bioimpedance devices for resistance, reactance, and phase angle values. *Eur J Clin Nutr*, doi:10.1038/s41430-019-0496-8 (2019).
- 33 Ward, L. C. Inter-instrument comparison of bioimpedance spectroscopic analysers. *The Open Medical Devices Journal* **1**, 3-10, doi:10.2174/1875181400901010003 (2009).
- 34 Gridneva, Z. *et al.* Bioimpedance spectroscopy in the infant: effect of milk intake and extracellular fluid reservoirs on resistance measurements in term breastfed infants. *Eur J Clin Nutr* **70**, 843-851, doi:10.1038/ejcn.2016.26 (2016).
- 35 Gonzalez, C., Evans, J. A., Smye, S. W. & Holland, P. Variables affecting BIA measurement of body water. *Med Biol Eng Comput* **37**, 106-107 (1999).

- 36 Demura, S., Yamaji, S., Goshi, F. & Nagasawa, Y. The Influence of Posture Change on Measurements of Relative Body Fat in the Bioimpedance Analysis Method. *J Physiol Anthropol Appl Human Sci* **20**, 29, doi:10.2114/jpa.20.29 (2001).

## **Acknowledgements**

J.L-R. is currently receiving University of Auckland Doctoral Scholarship. The authors would like to acknowledge Auckland NiPPeR research staff Christine Creagh, Marysia Depczynski, and Sarah Wilkins for their contribution to this study. Public good funding for the investigator-led NiPPeR study is through the UK Medical Research Council (as part of an MRC award to the MRC Lifecourse Epidemiology Unit (MC\_UU\_12011/4)); the Singapore National Research Foundation, National Medical Research Council (NMRC, NMRC/TCR/012-NUHS/2014); the National University of Singapore (NUS) and the Agency of Science, Technology and Research (as part of the Growth, Development and Metabolism Programme of the Singapore Institute for Clinical Sciences (SICS) (H17/01/a0/005); and as part of Gravida, a New Zealand Government Centre of Research Excellence. Funding for aspects of the NiPPeR study has been provided by Société Des Produits Nestlé S.A under a Research Agreement with the University of Southampton, Auckland UniServices Ltd, SICS, National University Hospital Singapore PTE Ltd and NUS. K.M.G. is supported by the National Institute for Health Research (NIHR Senior Investigator (NF-SI-0515-10042), NIHR Southampton 1000DaysPlus Global Nutrition Research Group (17/63/154) and NIHR Southampton Biomedical Research Center (IS-BRC-1215-20004)), British Heart Foundation (RG/15/17/3174) and the European Union (Erasmus+ Programme ImpENSA 598488-EPP-1-2018-1-DE-EPPKA2-CBHE-JP). S.Y.C. is supported by a Singapore NMRC Clinician Scientist Award (NMRC/CSA-INV/0010/2016). The funders had no role in the data collection and analysis, and the decision to submit for publication.

## **Author contributions statement**

W.S.C. and T.K. supervised all aspects of the research study. K.M.G., S-Y.C., and W.S.C. led the NiPPeR trial conception and design. J.L-R., W.S.C., and T.K. conceived and designed the body position sub-study. J.L-R. compiled the data and carried out the statistical analyses. J.L-R. wrote the manuscript with critical input from all other authors. All authors have approved

the final version of this manuscript and have agreed to be accountable for all aspects of this work.

### **Competing interests**

L.C.W. provides consultancy services to ImpediMed Ltd (a manufacturer of devices for bioelectrical impedance analysis). ImpediMed Ltd was not involved in the inception and conduct of this research, or in the writing of this manuscript. K.M.G. has received reimbursement for speaking at conferences sponsored by companies selling nutritional products, and is part of an academic consortium that has received research funding from Abbott Nutrition, Nestec, BenevolentAI Bio Ltd. and Danone. The other authors have no financial or non-financial conflicts of interest to declare.

**Table 1 Study population characteristics.**

	Mean (SD) population characteristics		
	Boys (n = 20)	Girls (n = 30)	All (n = 50)
Age (years)	3.38 (0.14)	3.38 (0.15)	3.38 (0.14)
Height (cm)	99.26 (3.63)	98.96 (3.87)	99.08 (3.74)
Weight (kg)	16.08 (1.72)	15.74 (2.04)	15.88 (1.91)
BMI <sub>SDS</sub>	0.60 (0.19)	0.45 (0.15)	0.51 (0.83)

**Table 2 Mean bioimpedance parameters when the participants were measured supine and standing (<1 minute and  $\geq 4$  minutes).**

	Mean (SD) impedance parameters		
	Supine	Standing (<1 min)	Standing ( $\geq 4$ min)
$R_0$ ( $\Omega$ )	813.5 (76.6)	789.3 (76.7)	786.1 (77.1)
$R_\infty$ ( $\Omega$ )	598.3 (63.8)	578.9 (65.0)	576.2 (67.1)
$Z_c$ ( $\Omega$ )	709.0 (69.7)	687.1 (70.3)	684.2 (71.5)
$Z_{50}$ ( $\Omega$ )	746.3 (72.5)	724.6 (72.5)	720.9 (73.1)
$R_{50}$ ( $\Omega$ )	743.8 (72.5)	722.2 (72.5)	718.5 (73.1)
$X_{c50}$ ( $\Omega$ )	60.1 (7.2)	57.9 (6.8)	58.1 (6.6)
Abbreviations: $R_0$ , resistance at 0 kHz; $R_\infty$ , resistance at infinite kHz; $Z_c$ , impedance at the characteristic frequency; $Z_{50}$ , impedance at 50 kHz; $R_{50}$ , resistance at 50 kHz; $X_{c50}$ , reactance at 50 kHz.			

**Table 3 Mean body composition values when the participants were measured supine and standing (<1 minute and ≥4 minutes).**

	Mean (SD) body composition values			% difference <sup>1</sup>
	Supine	Standing (<1 min)	Standing (≥4 min)	
TBW <sub>SFB7</sub> (L)	9.14 (1.28)	9.36 (1.31)	9.40 (1.35)	-2.73 (0.285)
ECW <sub>SFB7</sub> (L)	4.08 (0.56)	4.17 (0.58)	4.18 (0.58)	-2.31 (1.49)
ICW <sub>SFB7</sub> (L)	5.06 (0.79)	5.19 (0.81)	5.22 (0.82)	-3.14 (4.71)
FFM <sub>SFB7</sub> (kg)	12.49 (1.75)	12.79 (1.79)	12.84 (1.84)	-2.73 (2.85)
FM <sub>SFB7</sub> (kg)	3.38 (0.92)	3.09 (0.94)	3.03 (1.04)	13.75 (22.01)
FFM <sub>Rush</sub> (kg)	15.63 (1.29)	15.78 (1.30)	14.81 (1.32)	-1.12 (0.76)
FM <sub>Rush</sub> (kg)	0.25 (0.94)	0.10 (0.92)	0.07 (0.93)	9.12 (140.60)

<sup>1</sup>Percentage difference between mean supine and standing (≥4 min) body composition values.

Abbreviations: TBW<sub>SFB7</sub>, total body water from ImpediMed SFB7 built-in equation; ECW<sub>SFB7</sub>, extracellular water from ImpediMed SFB7 built-in equation; ICW<sub>SFB7</sub>, intracellular water from ImpediMed SFB7 built-in equation; FFM<sub>SFB7</sub>, fat-free mass from ImpediMed SFB7 built-in equation; FM<sub>SFB7</sub>, fat mass from ImpediMed SFB7 built-in equation; FFM<sub>Rush</sub>, fat-free mass from Rush et al. 2013 equation; FM<sub>Rush</sub>, fat mass from Rush et al. 2013 equation.

**Table 4 Regression equations developed in development sub-group (n=35) to allow measurements obtained when standing ( $\geq 4$  minutes) to be comparable to those obtained when supine.**

	Equation	R	R <sup>2</sup>
$R_{0\text{supine}}$	$31.138 + 0.996 R_{0\text{standing}}$	0.977	0.954
$R_{\infty\text{supine}}$	$39.498 + 0.970 R_{\infty\text{standing}}$	0.972	0.945
$Z_{c\text{supine}}$	$30.659 + 0.992 Z_{c\text{standing}}$	0.980	0.960
$Z_{50\text{supine}}$	$21.978 + 1.005 Z_{50\text{standing}}$	0.979	0.959
$R_{50\text{supine}}$	$21.720 + 1.005 R_{50\text{standing}}$	0.980	0.960
$X_{C50\text{supine}}$	$3.986 + 0.967 X_{C50\text{standing}}$	0.925	0.856
Abbreviations: $R_0$ , resistance at 0 kHz; $R_{\infty}$ , resistance at infinite kHz; $Z_c$ , impedance at the characteristic frequency; $Z_{50}$ , impedance at 50 kHz; $R_{50}$ , resistance at 50 kHz; $X_{C50}$ , reactance at 50 kHz.			

**Table 5 Bioimpedance body position adjustment equations applied to standing ( $\geq 4$  minutes) measurements in validation sub-group (n=15).**

	Validation cohort (n = 15)		T test		Bland-Altman		
	Mean	SD	t	p	Bias	Limits of agreement	
						Lower	Upper
<b>R<sub>0</sub></b>							
Supine	798.59	73.21	-0.369	0.718	-1.82	-39.33	35.69
Standing (adjusted)	800.41	78.70			-0.23%	-4.92%	4.47%
<b>R<sub>∞</sub></b>							
Supine	581.27	62.84	-0.170	0.867	-0.93	-42.50	40.64
Standing (adjusted)	582.20	71.66			-0.16%	-7.31%	6.99%
<b>Z<sub>c</sub></b>							
Supine	693.22	67.64	-0.260	0.799	-1.22	-36.89	34.45
Standing (adjusted)	694.44	75.52			-0.18%	-5.32%	4.97%
<b>Z<sub>50</sub></b>							
Supine	730.40	71.46	-0.017	0.987	-0.08	-36.57	36.41
Standing (adjusted)	730.49	78.16			-0.01%	-5.01%	4.98%
<b>R<sub>50</sub></b>							
Supine	727.82	71.56	0.026	0.980	0.13	-36.46	36.71
Standing (adjusted)	727.70	78.30			0.02%	-5.01%	5.04%
<b>Xc<sub>50</sub></b>							
Supine	61.01	5.57	-0.264	0.795	-0.26	-7.82	7.29
Standing (adjusted)	61.27	3.38			-0.43%	-12.82%	11.95%
Abbreviations: R <sub>0</sub> , resistance at 0 kHz; R <sub>∞</sub> , resistance at infinite kHz; Z <sub>c</sub> , impedance at the characteristic frequency; Z <sub>50</sub> , impedance at 50 kHz; R <sub>50</sub> , resistance at 50 kHz; Xc <sub>50</sub> , reactance at 50 kHz.							

## Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- [BIAbodypositionSupplementaryinformation.pdf](#)