

1 *Brief report*

2 **Bioimpedance vector references need to be period-** 3 **specific for assessing body composition and cellular** 4 **health in elite soccer players: a brief report**

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16 **Abstract:** Purpose: Bioimpedance data through bioimpedance vector analysis (BIVA) is used to
17 evaluate cellular function and body fluid content. This study aimed to i) identify whether BIVA
18 patterns differ according to the competitive period and ii) provide specific references for assessing
19 bioelectric properties at the start of the season in male elite soccer players. Methods: The study
20 included 131 male soccer players (age: 25.1 ± 4.7 yr, height: 183.4 ± 6.1 cm, weight: 79.3 ± 6.6)
21 registered in the first Italian soccer division (Serie A). Bioimpedance analysis was performed just
22 before the start of the competitive season and BIVA was applied. In order to verify the need for
23 period-specific references, bioelectrical values measured at the start of the season were compared to
24 the reference values for the male elite soccer player population. Results: The results of the two-
25 sample Hotelling T^2 tests showed that in the bivariate interpretation of the raw bioimpedance
26 parameters [resistance (R) and reactance (Xc)] the bioelectric properties significantly ($T^2 = 15.3$, $F =$
27 7.6 , $p = <0.001$, Mahalanobis $D = 0.45$) differ between the two phases of the competition analyzed. In
28 particular, the mean impedance vector is more displaced to the left into the R-Xc graph at the
29 beginning of the season than in the first half of the championship. Conclusions: For an accurate
30 evaluation of body composition and cellular health, the tolerance ellipses displayed by BIVA
31 approach into the R-Xc graph must be period-specific. This study provides new specific tolerance
32 ellipses (R/H: 246 ± 32.1 , Xc/H: 34.3 ± 5.1 , $r = 0.7$) for performing BIVA at the beginning of the
33 competitive season in male elite soccer players.

34 **Keywords:** BIVA; phase angle; R-Xc graph; tolerance ellipses

35

36 **1. Introduction**

37

38 **Body composition analysis is currently one of the most studied evaluations in sport, mainly for**
39 **the relationship between physical characteristics and sports performance [1]. In sports, excess fat**
40 **mass reduces endurance performance, while an increase in lean mass, especially muscle mass, is**
41 **associated with an increase in power and strength [2]. Furthermore, the assessment of localized body**
42 **composition allows the identification of differences in muscle mass and strength between areas of the**
43 **body and may allow a reduction in the risk of injury (evaluation of contralateral limbs, agonist-**
44 **antagonists) [3].**

45 Body composition assessment should also be considered in sports involving weight categories,
46 where athletes benefit from being placed in a lower weight category, in these cases any weight loss
47 must therefore be monitored closely. Excessive training coupled with calorie restrictions can lead to
48 excessive, unnecessary and dangerous weight loss. This weight loss in both women and men
49 decreases performance, bone mineral density, muscle mass and is detrimental to health [4, 5].

50 Bioelectrical impedance vector analysis (BIVA) is a method widely used to evaluate body
51 composition and cellular health in athletes, as well as in the general population [6-9]. This method
52 considers the raw bioelectrical parameters (resistance and reactance) standardized for the height of
53 the subjects as a vector within a graph. Resistance (R) is the opposition to the flow of an injected
54 alternating current, at any current frequency, through intra- and extracellular ionic solutions, while
55 reactance (Xc) represents the dielectric or capacitive component of cell membranes and organelles,
56 and tissue interfaces [10].

57 BIVA allows for the monitoring of vector changes over time or the comparison of the vector
58 position within the R-Xc graph on specific population tolerance ellipses [11-13]. Given the ease and
59 repeatability of this method, several references for athletes have recently been proposed, including
60 those for soccer players [14], volleyball players [15], and cyclists [16], while also considering the
61 competitive level of the athlete.

62 In soccer, Levi Micheli et al. [14] were the first to demonstrate how athletes need to be assessed
63 on specific tolerance ellipses, showing bioelectric values that were far different than those of the
64 normal healthy population. Subsequently, Mascherini et al. [17] suggested how bioimpedance
65 vectors show displacements over the season, reflecting the changes that occur in the body
66 composition and physical condition of the players. This was later confirmed by Campa et al. [18] who
67 analyzed the bioelectrical changes comparing BIVA to results obtained by DXA and dilution
68 techniques over a season in athletes, also showing that these vector changes occur in many other
69 sports.

70 During the different phases of competition, the one which precedes the start of the season is
71 among the most important periods in which to evaluate the athlete's physical condition and the body
72 composition adjustments that are sought during the pre-season. Considering the vector changes that
73 occur over the season, the bioelectrical references used in the BIVA assessment must be specific for
74 the competitive period in which the athlete is tested. Therefore, the purpose of this study was to show
75 how BIVA references provided in different phases of the season differ in male elite soccer players,
76 also providing new references for assessing body composition in the start-of-the season period.

77 2. Materials and Methods

78 2.1. Design and participants

79 A total of 131 male professional soccer players (age: 25.1 ± 4.7 yr; height: 183.4 ± 6.1 cm; weight:
80 79.3 ± 6.6 Kg) was recruited and participated in this observational study.

81 The inclusion criteria were: 1) players registered and participating in the first (Serie A) Italian
82 National division; 2) non-injured at the time of the assessment. After having been informed about the
83 aims and the procedures of the research, all athletes gave their written informed consent. The project
84 was approved by the Bioethics Committee of the University of Milan (approval code: 1052019) and
85 was conducted in accordance with the guidelines of the declaration of Helsinki.

86 2.2. Procedures

87 All measurements were performed in resting and fasting conditions at the facilities of the teams
88 the last week of August at 8.30 AM: Generally, this period corresponds to the end of the preparation
89 for the competitive season; therefore, it coincides with the start of the season. Body height was
90 recorded to the nearest 0.1 cm with a stadiometer (SECA® 240, Hamburg, Germany) and weight was
91 measured to the nearest 0.1 Kg with a calibrated weight scales (SECA® 877, Hamburg, Germany).

92 Whole-body impedance was obtained using a bioimpedance analyser (BIA 101 Anniversary
93 Edition, Akern, Florence, Italy). The device emits an alternating sinusoidal electric current of 400 mA

94 at an operating single frequency of 50 kHz ($\pm 0.1\%$). Subjects were positioned with a leg opening of
 95 45° with respect to the midline of the body, and with the upper limbs positioned 30° away from the
 96 trunk. The bioelectric phase angle (PhA) was calculated as the arctangent of $X_c/R \times 180/\pi$. BIVA was
 97 carried out using the classic methods, e.g., normalizing R (ohm) and X_c (ohm) for height in meters
 98 [6,8].

99 2.3. Statistical analyses

100 The two-sample Hotelling T^2 test was used to compare the differences in the mean impedance
 101 vectors between the bioimpedance data measured on the athletes of this study and the reference
 102 bioelectric values proposed by Levi Micheli et al. [14]. The 50%, 75%, and 95% tolerance ellipses were
 103 generated using the BIVA software [19]. Statistical significance was predetermined as $p < 0.05$. Data
 104 were analyzed with IBM SPSS Statistics, version 24.0 (IBM Corp., Armonk, NY, USA).

105 3. Results

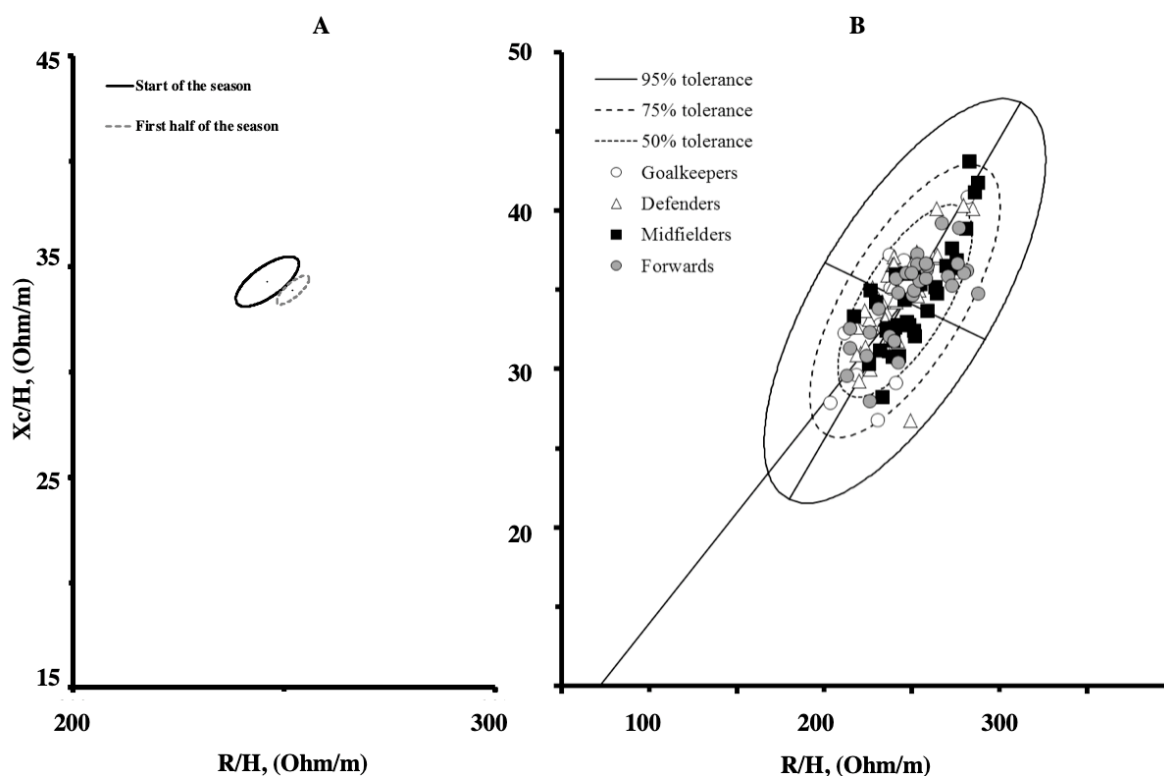
106 Table 1 shows anthropometric and bioelectrical characteristics of the soccer player.

107 **Table 1.** Descriptive statistics for the soccer players according to playing position.

Variable	Goalkeepers n = (15)	Defenders n = (38)	Midfielders n = (38)	Forwards n = (40)	All n = (131)
Age (years)	24.2 \pm 5.9	26.6 \pm 4.8	25.0 \pm 4.8	24.5 \pm 3.9	25.1 \pm 4.7
Weight (kg)	86.7 \pm 5.4	80.6 \pm 5.6	76.8 \pm 5.6	77.6 \pm 6.5	79.3 \pm 6.6
Height (cm)	188.3 \pm 3.5	185.1 \pm 5.0	181.4 \pm 5.2	181.8 \pm 7.2	181.8 \pm 7.2
BMI (kg/m ²)	24.5 \pm 1.0	23.5 \pm 0.8	23.3 \pm 1.0	23.5 \pm 1.0	23.5 \pm 1.0
R/H (ohm/m)	234.0 \pm 18.1	242.9 \pm 17.0	251.9 \pm 18.9	254.6 \pm 21.9	248.1 \pm 20.3
X_c/H (ohm/m)	33.0 \pm 3.9	34.4 \pm 3.1	34.7 \pm 3.2	35.4 \pm 3.0	34.6 \pm 3.3
PhA (degree)	8.0 \pm 0.7	8.1 \pm 0.5	7.8 \pm 0.4	7.9 \pm 0.4	8.0 \pm 0.5

108 Abbreviations: BMI, body mass index; R/H, resistance standardized for height; X_c/H , reactance
 109 standardized for height; PhA, phase angle.

110 The results of the two-sample Hotelling's T^2 test showed separate 95% confidence ellipses
 111 indicating a significant difference ($T^2=15.3$, $F=7.6$, $p < 0.001$, Mahalanobis $D=0.45$) between the BIVA
 112 patterns measured in this study and those proposed by Levi Micheli et al. [14] as a reference for the
 113 male elite soccer players population (Figure 1a).



114

115 **Figure 1.** Mean impedance vectors with the 95% confidence ellipses for the soccer players measured
 116 at the start and at the first half of the competitive season [10] (Panel A). Scattergrams of the individual
 117 impedance vectors plotted on the new tolerance ellipses (Panel B).

118 The new reference ellipses and the single bioimpedance vectors measured in the soccer players
 119 at the start of the season are shown in Figure 1b.

120 **4. Discussion**

121 The aim of this study was to show the importance of evaluating bioelectric properties using
 122 BIVA references that are suitable for the competitive period in which the assessment is carried out.
 123 The results of this study, which provide bioelectrical impedance data for 131 elite players, showed
 124 how the tolerance ellipses created on the basis of measurements during the different phases of the
 125 competition differ significantly for elite soccer players.

126 The bioimpedance data reported in the present study are comparable to previous values
 127 reported during start-of-the season period in elite soccer players [20, 22]. In comparison with the elite
 128 Italian male soccer population investigated by Levi Micheli et al. [14], the elite soccer players
 129 measured in this study showed a significant vector shift to the left on the minor axis of the tolerance
 130 ellipses. This could indicate a greater cell mass, which is a consequence of the effects sought in the
 131 preparation phase (training and controlled diet) typically, designed to increase endurance level and
 132 increase strength [23]. In fact, in a previous research, Mascherini et al. [17] suggested that the
 133 shortening of the vector was associated with changes in hydration status and increases in body cell
 134 mass. In this study, the preparation phase could have increased the intracellular/extracellular water
 135 (ICW/ECW) ratio as can be seen from a higher PhA than that measured by Levi Micheli et al. [14] (8.0
 136 $\pm 0.5^\circ$ vs. $7.7 \pm 0.6^\circ$). Indeed, PhA is positively associated with the ICW/ECW ratio in athletes [18, 24]
 137 Bioelectric data reflects the content of body fluids and the cellular health of the athlete and during
 138 the season, which change in response to training load and physical condition over the season [25].
 139 In fact, the new tolerance ellipses proposed in this study differ significantly from those generated in the
 140 study by Levi Micheli et al. [14], in which bioimpedance measurements were collected in the first half
 141 of the competitive period. Furthermore, Micheli Levi et al. [14] reported that BIA data was collected
 142 over 5 months, from October to January 2009-10, a period of time that may have generated vector

143 changes in the athletes themselves. Our hypothesis is that the increase in workload (training) and
144 official matches from August to October (about 6-8 matches played) or from August to January (16-
145 17 matches played) could lead to fatigue and increased muscle turnover, reduced muscle function
146 which could result in a shift to the right of the biimpedance vector. In fact, during the season, the
147 reduction of the PhA could indicate a decreased muscle function as shown by Norman et al. [26]
148 However, since we have not performed any muscle function tests, this hypothesis will have to be
149 further investigated in future studies.

150 The reference ellipses proposed in the literature for athletes are population-specific. In addition
151 to those for soccer players proposed by Levi Micheli [14], Campa and Toselli [15] measured male
152 volleyball players in the second half of the in-season and showed specific BIA vector distribution in
153 elite players in comparison to lower levels athletes. Subsequently, Giorgi et al. [16] provided
154 bioelectrical impedance data of male road cyclists of varying performance levels, measured at the
155 time of their optimal performance level and identified the 50, 75 and 95% tolerance ellipses for the
156 road cyclists population, as well as for the high-performance road cyclists. In addition to these, there
157 are also ellipses for healthy athletes built on more than 1,000 male and 440 female athletes during the
158 off-season period, therefore suitable for evaluating BIVA in the first phase of the competitive season
159 [12].

160 The authors are also aware of the limitations of the study. Firstly, the subjects come from the
161 same territory; therefore, the results obtained are not generalizable to all the soccer players around
162 the world: a larger sample size is required even in different countries. The second is that no division
163 by ethnicity of the players has been made in order to obtain a sample as large as possible: currently
164 an international data collection is active that will allow us to investigate both these two limitations.

165 A strength of this study is in the specific time period in which the measurements were collected,
166 not only in regard to the competitive level of the athletes, but above all for the time span in which
167 BIA assessments were performed. In fact, BIA measurements were collected within a week, just
168 before the start of the season, a period of time too short to generate vector changes between the
169 players.

170 For the reasons mentioned above, future studies conducted with the aim of providing BIVA
171 references for athletes should carry out the measurements according to the competitive phase for
172 which they want to provide the new references. This is very significant given that vector changes
173 occur during the different phases of the season in athletes, and bioelectrical values must be as
174 informative and specific as possible, in order to obtain accurate monitoring of the body composition
175 and physical condition of the athlete. This study demonstrates the importance of evaluating athletes
176 on period-specific BIVA references, providing new tolerance ellipses for assessing body composition
177 and cellular health before the start of the competitive season in elite soccer players.

178 5. Conclusions

179 Through BIVA, it is possible to evaluate body composition and the state of physical condition in
180 the different phases of the competition in elite soccer players. This study provides specific BIVA
181 references for the start of the season period, through which the physical condition achieved after the
182 preparation micro cycle in soccer can be assessed.

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184 G.P.; formal analysis, F.C.; investigation, T.B.; data curation, F.G.; writing—original draft preparation, F.C. and
185 G.M.; supervision, A.T. and M.V.; project administration, G.A. All authors have read and agreed to the published
186 version of the manuscript.

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