# Prediction of lamb body composition using in vivo bioimpedance analysis

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17	Abstract
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19	The objective of this study was to evaluate the potential of <i>in vivo</i> bioimpedance analysis
20	(BIA) as a method to estimate body composition in lambs. Thirty-one Texel x lle de France
21	crossbreed ram lambs were slaughtered at pre-determined intervals of average weights of
22	20, 26, 32, and 38 kg. Before the slaughter of the animals, their body weight (BW) and
23	body length (BL) were measured. The values for resistance (Rs) and reactance (Xc) were
24	collected using a single-frequency BIA equipment (Model RJL Quantum II Bioelectrical
25	Body Composition Analyzer). The BIA main variables such as body bioelectrical volume
26	(V), phase angle (PA), resistive density (RsD), and reactive density (XcD) were then

27 calculated. The soft tissue mass of the right-half cold carcass was analyzed in order to 28 determine its chemical composition. Multiple regression analyses were performed using 29 the lamb body composition as dependent variables and the measurements related to 30 bioimpedance as independent variables. The best regression models were evaluated by 31 cross-validation. The predictive model of moisture mass, which was developed by using 32 XcD and V, accounted for 84% of its variation. Resulting models of percentage moisture  $(R^2 = 0.79)$ , percentage lean mass  $(R^2 = 0.79)$ , percentage fat  $(R^2 = 0.79)$ , and fat mass 33 34  $(R^2 = 0.87)$  were obtained using RsD and V. Furthermore, the values of RsD regarding V, 35 and PA in the prediction models accounted for 91% and 89% of variation in protein mass 36 and lean mass, respectively. Bioimpedance analysis proved to be an efficient method to 37 estimate the body composition of lambs slaughtered at different body mass stages.

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39 **Keywords:** sheep, lean mass, carcass composition, impedance, resistance

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## 41 **1. Introduction**

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43 Sheep farming is an essential economic activity in many countries especially the poorest 44 ones (Herrero et al., 2013). However, recent sheep meat consumption patterns are in line 45 with current market trends indicating that increased sheep meat consumption depends on 46 the final product quality to satisfy a higher purchasing power in emerging economies 47 (Sepúlveda, Maza, & Pardos, 2011; Shackelford, Leymaster, Wheeler, & Koohmaraie, 48 2012). Therefore, to develop the lamb production sector to reach its full potential farmers 49 need to be more efficient, achieve economies of scale as well as consider engaging in 50 producing certified lamb to fulfil market demand. As a result, more integration among all 51 links of the production chain is required in order to achieve the consolidation of sheep 52 farming aimed at quality meat production (Ricardo et al., 2015).

53 A solid understanding of tissue growth and tissue development rate that make up lamb 54 carcasses is extremely important, because from such information better strategic 55 management interventions could be considered in order to improve the desired market 56 body tissue deposition. Besides, carcasses tend to be paid according to their quality, 57 which is related to a greater lean mass percentage and minimal fat amount to satisfy 58 market demand (Álvarez et al., 2013; della Malva et al., 2016; Font-i-Furnols & Guerrero, 59 2014). The latter should be just enough to deliver the expected nutritional and organoleptic 60 characteristics from sheep meat.

The evaluation of carcass composition using *in vivo* methodology allows firstly, for a better use of the factors of production, secondly, to determine the ideal age of slaughter, and, thirdly, to hopefully provide a uniform product for the meat processing industries. Furthermore, the use of *in vivo* body composition analysis also contributes to the reduction of error in carcass since classification tends to be subjective (Zollinger, Farrow, Lawrence, & Latman, 2010).

67 *In vivo* bioimpedance analysis (BIA) is a simple, rapid, relatively cheap, non-destructive, 68 and minimally invasive method to estimate the body composition of sheep (Altmann, 69 Pliquett, Suess, & Von Borell, 2004; Avril, Lallo, Mlambo, & Bourne, 2013; Berg & 70 Marchello, 1994). Bioimpedance measures both modulus and phase of an equivalent 71 impedance at a certain frequency, which is then calculated both resistance and reactance. 72 Bioimpedance uses an alternating current (eq., 800 µA at 50 kHz) which is injected into the 73 biological material (Swantek, Crenshaw, Marchello, & Lukaski, 1992). Furthermore, BIA 74 may act as an estimator of body composition since it senses the difference in the 75 conductivity between fat and fat-free mass (Lukaski et al., 1985). However, BIA is not a 76 direct method to assess body composition and its accuracy, among other non-linearities, 77 depends also on the precision of the chosen regression equations (Norman, Stobäus, 78 Pirlich, & Bosy-Westphal, 2012). The use of resistive and reactive densities to predict beef

carcass composition has already been investigated (Zollinger et al., 2010). As far as our knowledge is concerned, the use of such predictors have not been used in lambs. The objective of this study was to evaluate the potentiality of *in vivo* bioimpedance analysis to estimate body composition in lambs at different ages of slaughter.

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#### 84 2. Material and methods

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The research was conducted at the Laboratory of Sheep of the Department of Animal Science of the Federal University of Santa Maria (UFSM), Santa Maria, RS, Brazil, after being approved by the Ethics Committee on Animal Trials of this University, under the protocol #8259211015. All handling from the lamb rearing until their slaughter and laboratory analyzes were carried out within the Sheep Laboratory confines.

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### 93 2.1 Animals

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Thirty-one Texel and IIe de France crossbred three-month old weaned ram lambs were used for the purpose of this research. The animals were marked, weighed and distributed into eight pens covered from the elements. They were distributed in groups according to similar weight but on average they had live weight of 19.7 kg. The pens had slatted-floor and were equipped with feeders and water troughs. The animals were fed *ad libitum* and the amount of feed was adjusted in order to keep the leftover food at around 10% of the total amount given.

A balanced diet calculated according to the NRC (2007) guidelines was administered to the animals in order for them to gain 0.200Kg/day. The formulation based on dry matter accounted for the 30:70 ratio alfafa hay:concentrate (37.9% of maize grain, 14.4% of soy

bran, 16.4% of wheat bran, and 1.27% of calcitic lime). The mineral mixture was supplied *ad libitum* in separated feeders. Weight measurements took place at weekly intervals until they reached pre-established slaughter weights of 20, 26, 32, or 38 kg. Before the slaughter of the animals, body weight (BW) (kg) was recorded (after 14 hours of fasting of solids), body length (BL) (cm) was measured (from the last cervical vertebrae to the first sacral vertebrae), and the BIA readings were taken.

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## 112 2.2 Bioimpedance measurements

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114 A single-frequency BIA equipment (Model RJL Quantum II Bioelectrical Body Composition 115 Analyzer) was used to make the measurements. This apparatus injects an alternating 116 electrical current of 800  $\mu$ A at 50 kHz into the different body tissues by using two 117 electrodes and then measures the voltage resultant across other two electrodes. The 118 equipment is connected by four cables that are attached to the electrodes by color-coded 119 electric clips. The black and red electrodes were configured as current transmitters and 120 current detectors, respectively. Stainless steel acupuncture needles with spiral cable (0.40 121 x 15 mm) were used as electrodes. They were inserted into the animal muscle at a 122 standard depth just to make a good electrode contact in all body tissues (Berg & 123 Marchello, 1994).

Data were collected following the methodology proposed by Jenkins, Leymaster, and Turlington (1988) and adapted for live lambs. During data collection, the lambs were initially contained with an insulating rope tied to their feet, in order to keep them immobile without the need of use of anesthetics. Then, they were laid on plastic tarpaulin for insulating them from any contact with the ground, preventing leakage current. The animals were positioned on their right side (*lateral decubitus*) and a small area of the animal wool was shaven off in a straight line on the central region of the lateral face of the leg region

(rear limb) and forearm region (front limb). The distal transmitter electrodes were attached onto the extensor muscle complex of the front and rear limbs, approximately 3.0 cm proximal to the carpal and tarsal articulations, respectively. The proximal detector electrodes were inserted 10 cm from the transmitter ones, caudal to the knee region (rear limb) and cranial to the elbow region (front limb) (Fig. 1).

136 Both resistance (Rs) ( $\Omega$ ) and reactance (Xc) ( $\Omega$ ) were measured once for each animal, 137 then values of conductance (C) and impedance (Z) were calculated by using the formulas 138 described by Lukaski, Johnson, Bolonchuk, and Lykken (1985), where C = 1/Rs ( $\Omega$ ) and Z 139 =  $(Rs^2+Xc^2)^{0.5}$  ( $\Omega$ ). The relationship between resistance and reactance values results in an 140 angle, which is defined as phase angle (PA) (°). The PA was directly calculated as the arc 141 tangent of the ratio of reactance and resistance, where PA = tan<sup>-1</sup> (Xc/Rs), expressed in 142 radians. To convert the results into degrees, the PA values were then multiplied by  $180^{\circ}/\pi$ 143 as described by Lukaski (2013).

The body bioelectrical volume (V) was obtained by the relationship between animal body length and resistance, where V=BL<sup>2</sup>/Rs (cm<sup>2</sup>/ $\Omega$ ), adapted from Jenkins et al. (1988). Both resistive (RsD) and reactive density (XcD) were also calculating according to Zollinger et al. (2010). They proposed to replace the half carcass weight by body weight as well as replacing the distance between the electrodes by the body length of the animals. Therefore, the final formula used in this work is as it follows: RsD = BW<sup>2</sup>/(BL<sup>2</sup>/Rs) (kg<sup>2</sup>/cm<sup>2</sup>  $\Omega$ ) and XcD = BW<sup>2</sup>/(BL<sup>2</sup>/Xc) (kg<sup>2</sup>/cm<sup>2</sup>  $\Omega$ ).

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152 2.3 Laboratory analyses

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After the slaughter, the carcasses were weighed and stored in a chilling chamber at a temperature of 2°C. After 24 hours, they were weighed for a second time in order to obtain the cold carcass weight (CCW) (kg). The carcasses were longitudinally split into two half

157 carcasses and the right half carcasses were weighed and divided into four regional cuts; 158 neck, shoulder, ribs, and leg. These cuts were boned in order to obtain the soft tissue 159 mass of the half carcasses, which are composed basically by muscles, fat, blood vessels, 160 nerves, and connective tissue. The soft tissue mass obtained from each cut was ground, 161 homogenized, and approximately 200 g of representative sample were extracted for 162 laboratory analyses. Four samples from each carcass were analyzed in duplicates. 163 Moisture (930.15), protein (992.15), and ash (942.05) were determined in these samples 164 according to the methodology described on AOAC International (1995). Fat was 165 determined according to the method proposed by Bligh and Dyer (1959). The chemical 166 composition determined in each cut was used to calculate the weight and the proportion of 167 each chemical component of the half carcass and adjusted to the cold carcass weight. The 168 weight and percentage of the lean mass was obtained by the sum of weight and 169 percentage, respectively, of protein and moisture of the carcass soft tissue, then Lean 170 mass (kg) = protein (kg) + moisture (kg) and Lean mass (%) = protein (%) + moisture (%), 171 according to Jenkins et al. (1988).

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173 2.4 Statistical analysis

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175 Multiple regression analyses were performed using the lamb carcass composition as 176 dependent variables (weight and percentage of moisture, protein, fat, and lean mass) and 177 independent variables (resistance, bioimpedance measurements as reactance. 178 conductance, phase angle, body bioelectrical volume, resistive density, and reactive 179 density). The body weight and body length of the animals were only used to calculate V, 180 RsD, and XcD. The normality of the data was presumable based on the central limit 181 theorem, which considers an acceptable normal distribution with 30 or more observations. 182 Correlation coefficients between the BIA measurements and the carcass composition were

determined. Stepwise regression was used to eliminate the variables that did not increase the prediction power of the model. Those variables that did not significantly contribute to the model (P > 0.05) were eliminated. The biostatistical models were selected which presented the highest coefficient of determination ( $\mathbb{R}^2$ ), lowest root mean square error (RMSE), and the Mallows Cp statistic (Cp) closest to the number of parameters included in the model.

189 The best regression models were evaluated by cross-validation according to the following 190 procedure, which was the same for each model. Briefly, from the complete dataset, one 191 animal was selected and regression parameters were estimated with data of the remaining 192 n - 1 animals. Values of weight and percentage of moisture, protein, fat, and lean mass 193 were predicted for the selected animal by this regression function. Then, the squared 194 difference between the predicted and measured chemical compounds of the selected 195 animal was calculated. This procedure was repeated for every animal. The mean of the n-196 squared differences between the predicted and measured carcass compounds of all n 197 animals was calculated to obtain the mean squared error of the predicted weight and 198 percentage of moisture, protein, fat, and lean mass. Precision and accuracy of the 199 equations were measured by evaluating the highest coefficient of determination  $(R^2)$ , the 200 lowest root mean squared error (RMSE), the mean prediction error (MPE), and relative 201 mean prediction error (RMPE; %) of the cross-validation. The statistical software SAS 202 (SAS University Edition, 2017) was used for all statistical analyses.

- 203
- 204 **3. Results**

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206 3.1 Descriptive statistics

The descriptive statistics of linear measurements and bioimpedance assessments in lambs are shown in Table 1. The body weight presented a larger variation range than body length. The minimum and maximum resistance readings ranged from 60 to 110  $\Omega$ , while the reactance variation was from 6 to 15  $\Omega$ . Body bioelectrical volume, resistive density, and reactive density, which used weight and/or length values in their formulas, had larger variation than resistance and reactance alone.

The mean values, range, and variability of carcass linear measurements and lamb body composition are shown in Table 2. The weight of cold carcass and soft tissue had great variation between the minimum and maximum values. The same can be observed with the mass of the lamb body components. When the body composition was expressed in percentage values, fat content showed to present the largest variation, ranging from 8.39 to 26.9%.

220

3.2 Correlations

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223 The results of the correlations between BIA measurements and chemical analyses of the 224 lamb carcasses are presented in Table 3. The mass of body chemical constituents 225 increased with the increase in BL, BW, PA, V, RsD, and XcD. Although fat percentage also 226 increased, in contrary, protein, moisture, and lean mass percentages decreased. The 227 relationship between BL and/or BW with Rs or Xc may have affected the stronger 228 correlations between V, RsD, and XcD and the lamb body composition. Protein 229 percentage was an exception because it was not significantly correlated to any BIA 230 variables (P > 0.05).

231

232 3.3 Prediction Models

234 The final predictive models of the lamb body composition are shown in Table 4. The 235 amount of moisture was mostly explained by the XcD ( $R^2 = 0.56$ ; Cp = 93.3). When V was 236 included in this model, they accounted for 84% of its variation on lamb carcass, and the Cp 237 value decreased to 17.6. Regarding protein and fat mass predictive models, the use of 238 RsD alone explained most of the variation of these components on carcass, 62% and 70%, respectively. When V was added to the models, their prediction power improved (R<sup>2</sup> 239 240 = 0.89 and  $R^2$  = 0.87) and the Cp values decreased from 87.3 and 41.2 to 9.04 and 3.56, 241 respectively. Furthermore, to estimate the protein content of the samples the PA value was 242 then added to the protein equation. The PA value together with RsD and V explained 91% 243 of protein variation on carcass. From these results, it is possible to note that predictive 244 models for protein mass and fat mass are more precise than the one for moisture mass.

Resistive density, alone, accounted for 63% of variation of moisture, fat, and lean mass percentages on lamb carcass. When V was added to these models, there was an increase of 16% in their coefficients of determination. Furthermore, the Cp values decreased from 20.5, 25.3, and 25.1 to 2.32, 4.70, and 4.56, respectively. However, none of the BIA variables contributed significantly (P > 0.05) to the prediction of protein percentage. This fact may be explained due to the little variation in protein percentage on lamb carcass (18.9 ± 0.12%) (Table 2).

252 The lean mass model was initially mostly explained by XcD ( $R^2 = 0.56$ ). Afterwards, V was 253 added to the model, which increased the prediction power in 28%. Thereafter, the addition 254 of RsD enhanced R<sup>2</sup> by 2%. Using the Stepwise procedure, when RsD was added to the 255 model, it was observed that XcD stopped contributing to the prediction power of the model 256 and, therefore, it was removed from the equation. Besides, when PA was added to that 257 model, it accounted for 89% of the variation of the lean mass on lamb carcass. The 258 prediction power of these equations can be observed by the statistic results of the cross-259 validation. Regarding the R<sup>2</sup> of the cross-validation, the models for predicting the mass of moisture, protein, fat, and lean mass presented good precision with  $R^2$  of 0.93, 0.95, 0.91 and 0.93, respectively. Although the prediction models of the percentage of moisture, fat, and lean mass resulted in a lower  $R^2$  compared to the absolute values, they also had an acceptable prediction power with  $R^2$  of 0.86, 0.88, and 0.88, respectively.

264

#### 265 **4. Discussion**

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The lamb body development is typically understood as an increase in mass. Growth rates of different tissues which compound the animal carcass are influenced by many factors such as genetics, age, and nutrition (Owens, Dubeski, and Hansont, 1993). Nonetheless, the lamb body shape change over time, especially before they reach maturity, indicating they tend to take longer to grow (in centimeters) than to gain weight (in kilograms). That could be confirmed in the sample because body weight varied more than body length (Table 1).

The carcass composition can be expressed in either mass or percentage. As the increase of body weight at slaughter increases the weight of cold carcass, consequently the weight of each body compound also increases with a heavier carcass. Body composition when expressed in percentages shows that as the carcass fat content increases, the lean mass content decreases. The protein percentage had the smallest variation in the carcasses (Table 2). The small variation in protein percentage is a limitation in the prediction using the bioimpedance method.

The results of this study indicate that the electrical properties of lamb body tissues were affected by their body composition, whereas longer and heavier animals, with more body fat deposits presented higher BIA values than smaller and lighter ones (Table 3). Due to the fact that it was performed a four electrode impedance measurement, as known by transfer impedance, this agrees with the bioimpedance principle as, it depends on the

electrode geometry, volume and length of the biological conductor material, and on the
frequency of the applied current (Berg & Marchello, 1994; Lukaski et al., 1985; Swantek,
Crenshaw, Marchello, & Lukaski, 1992). Electrode material, distance between electrodes,
and the place where they are insert also influence BIA measurements. Therefore, BIA
reproducibility depends on the electrode placement.

291 The body volume of lambs is composed of intra- and extracellular fluids that behave as 292 heterogeneous electrical conductors with cell membranes working as electrical capacitors, 293 and body fat acting as an insulator material and generating electrical resistance (Altmann 294 et al., 2004; Swantek et al., 1992). For humans, this volume is obtained by the relationship 295 between the squared height and resistance (Lukaski et al., 1985). However, in lambs, 296 Jenkins et al. (1988) replaced the height by carcass length. Then Berg and Marchello 297 (1994) replaced it by the conductor length (distance between current detector electrodes). 298 Regardless of how the volume was obtained, it is needed to estimate the body 299 composition of lambs. This fact was evidenced in this study since this variable was 300 selected in the prediction models for both mass and percentage of the lamb body 301 components (Table 4).

302 Reactance is the property of storing alternating electrical energy under the form of an 303 electric field. It is related to the dynamic performance of cell membranes, which work as an 304 electrical capacitor (Swantek et al., 1992). The cell membrane capacitance can be used as 305 an indicator of lean mass and intracellular body mass, and it is also related to extra- and 306 intracellular hydric balance (Altmann et al., 2004). This fact may explain the model 307 selection of volume and reactive density to estimate the water amount on lamb carcass 308 (Table 4). The reactive density explained 56% of the variation of lean mass on lamb 309 carcasses. This might be due to the large amount of moisture (65.4% in average) in the 310 soft tissue mass of the lamb carcasses (Table 2). The negative correlations obtained

between XcD and moisture, protein, and lean mass percentages reinforce this fact (Table312 3).

313 The resistive part of the impedance can be attributed to the opposition of a current flow 314 through intra- and extracellular ions inside the tissue (Lukaski, 2013), which can be directly 315 correlated to the difference in conductivity between fat and lean mass (Jenkins et al., 316 1988). Hence, Rs is directly related to the body hydration level and body fat. Animals with 317 a larger percentage of fat in the carcass present smaller body water percentage and 318 higher resistance to the applied current. As the mass of carcass components increases, 319 there is an increment in body weight and in some BIA variables like PA, V, RsD, and XcD 320 (Table 3). These increases were also related to an increase of fat percentage, but to 321 decreased moisture, protein, and lean mass percentages on lamb carcasses (Table 3). 322 According to Owens, Dubeski, and Hansont (1993), changes in body composition are 323 expected since they naturally occur throughout the physiological maturity of the lambs. 324 Based on these results it is possible to note a significant efficiency of BIA in detecting 325 these changes in body composition of lambs slaughtered at different body masses. 326 According to Zollinger et al. (2010), who assessed BIA in beef cattle carcasses, RsD and 327 XcD decreased with the increase of lean mass, and increased with the increase of fat 328 percentage on carcass. This indicates a strong relationship of RsD and XcD with lean 329 mass and fat body contents.

The phase angle was another important variable used in this work, which was obtained by the relationship between resistance and reactance values. The PA may vary from zero (eg., medium without cell membranes) to 90 degrees (eg., a medium full of cell membranes but no fluids) (Lukaski, 2013). This angle depends on the capacitance due to the cell membranes and, in humans, it is a marker of amount and quality of soft tissue mass, as well as body hydration status (Norman et al., 2012). Therefore, its variation indicates changes in body composition, especially for protein and lean mass.

Predictive models for body composition need to be accurate in order to become suitable to farmers. Therefore, Mallows Cp statistic is considered a robust tool for predictive model selection. The Cp value that is closest to the number of parameters included in the model indicates less biased estimates and, therefore, a more precise model. Similar to what Berg and Marchello (1994) found, this study identified some possible outliers which were not removed from the database given the difficulty of identifying the origin of the error. Removal of these outliers could also improve the results from the statistical analysis.

On the one hand, the predictive models for mass of body constituents presented higher R<sup>2</sup> and lower RMSE values when compared to their respective percentages, and therefore, greater precision. On the other hand, if the Cp value is considered, the moisture percentage and lean mass percentage presented a higher precision to estimate the real regression coefficients and predict future answers than their absolute masses. Likewise, considering these statistical parameters, it is possible to note that predictive models for protein mass and fat mass are more precise than the one for moisture mass (Table 4).

351 Regarding the cross-validation, the RMPE values for predicting fat, both in absolute and 352 relative values, were higher than the other carcass components. The RMPE values for 353 predicting the absolute carcass compounds were also higher comparing to the relative 354 ones. This may be due to the higher variation range of the values expressed in grams 355 related to those presented as percentage. Nevertheless, concerning the  $R^2$  of the cross-356 validation, the absolute values of the lamb carcass components had the highest accuracy. 357 These findings indicate that the models obtained for predicting lamb carcass composition 358 are quite robust.

359

**5. Conclusion** 

361

362	Bioimpedance is an efficient method to estimate the carcass composition of lambs
363	slaughtered at different body masses. The use of resistive and reactive densities data
364	improved the results of in vivo bioimpedance analysis used to assess the lamb body
365	composition. This might be the case of measuring lamb body composition in order to
366	reduce errors related to subjectivity.
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368	Conflict of interests
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370	There is no conflict of interests involved in this work.
371	
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378	
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	n¹	Min <sup>2</sup>	Max <sup>3</sup>	Mean	SD <sup>4</sup>	SEM <sup>5</sup>
Body weight, kg	31	18.1	38.6	27.9	5.48	0.98
Body length, cm	31	47.0	64.0	55.8	4.76	0.85
Resistance, Ω	31	60.0	110.0	87.7	11.9	2.14
Reactance, Ω	31	6.00	15.0	10.4	2.33	0.42
Conductance, Ω	31	0.01	0.02	0.01	0.00	0.00
Impedance, Ω	31	60.3	111.0	88.3	12.0	2.16
Phase angle, °	31	4.81	8.60	6.70	1.03	0.18
Body bioelectrical volume, $cm^2/\Omega$	31	24.5	58.2	36.5	8.45	1.52
Resistive density, $kg^2/cm^2 \Omega$	31	9.05	41.6	22.1	7.06	1.27
Reactive density, $kg^2/cm^2 \Omega$	31	0.82	5.61	2.64	1.06	0.19

454 Descriptive statistics of body characteristics and bioimpedance assessments on lambs.

455 <sup>1</sup>n = number of lambs, <sup>2</sup>Min = Minimum, <sup>3</sup>Max = Maximum, <sup>4</sup>SD= Standard deviation, <sup>5</sup>SEM=

456 Standard error of mean.

459	Descriptive statistics of	carcass characteristics	and body composition	of lambs.
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	n <sup>1</sup>	Min <sup>2</sup>	Max <sup>3</sup>	Mean	SD <sup>4</sup>	⁵SEM
Cold carcass weight, kg	31	6.36	17.4	11.9	2.94	0.53
Soft tissue mass weight, kg	31	5.07	14.6	9.71	2.58	0.46
Moisture, kg	31	3.55	8.59	6.26	1.35	0.24
Moisture, %	31	54.0	72.2	65.4	4.03	0.72
Ash, kg	31	0.05	0.13	0.09	0.02	0.00
Ash, %	31	0.59	1.14	0.91	0.12	0.02
Protein, kg	31	0.98	2.65	1.83	0.47	0.08
Protein, %	31	18.0	20.8	18.9	0.69	0.12
Fat, kg	31	0.49	3.61	1.53	0.83	0.15
Fat, %	31	8.39	26.9	14.8	4.40	0.79
Lean mass, kg	31	4.53	11.1	8.09	1.81	0.32
Lean mass, %	31	72.3	90.6	84.3	4.32	0.78

<sup>1</sup>n = number of lambs, <sup>2</sup>Min = Minimum, <sup>3</sup>Max = Maximum, <sup>4</sup>SD= Standard deviation, <sup>5</sup>SEM=
Standard error of mean.

464	Simple Pearson	correlations between body	v characteristics	and bioimpedance	assessments to la	mb body composition.
			/			2 1

	BW <sup>1</sup> , kg	BL <sup>2</sup> , cm	Rs³, Ω	Xc <sup>4</sup> , Ω	C <sup>5</sup> , Ω	Ζ <sup>6</sup> , Ω	PA <sup>7</sup> , °	V <sup>8</sup> , cm²/Ω	RsD <sup>9</sup> , kg²/cm² Ω	XcD <sup>10</sup> , kg²/cm² Ω
Moisture, kg	0.93***	0.70***	-0.07	0.33	0.05	-0.06	0.51**	0.54**	0.75***	0.75***
Moisture, %	-0.85***	-0.59***	-0.05	-0.31	0.03	-0.05	-0.37*	-0.40*	-0.79***	-0.77***
Protein, kg	0.95***	0.70***	-0.04	0.34	0.02	-0.03	0.50**	0.52**	0.79***	0.78***
Protein, %	-0.33	-0.25	0.02	-0.02	-0.04	0.02	-0.01	-0.21	-0.32	-0.28
Fat, kg	0.90***	0.62***	0.05	0.32	-0.04	0.05	0.39*	0.42*	0.84***	0.81***
Fat, %	0.85***	0.59***	0.04	0.30	-0.02	0.05	0.36*	0.40*	0.79***	0.77***
Lean mass, kg	0.94***	0.70***	-0.06	0.33	0.04	-0.05	0.51**	0.54**	0.76***	0.76***
Lean mass, %	-0.85***	-0.59***	-0.04	-0.29	0.02	-0.05	-0.35	-0.41*	-0.79***	-0.76***

 $^{1}$ BW = Body weight,  $^{2}$ BL = Body length,  $^{3}$ Rs = Resistance,  $^{4}$ Xc = Reactance,  $^{5}$ C = Conductance,  $^{6}$ Z = Impedance,  $^{7}$ PA = Phase angle,  $^{8}$ V = Body bioelectric volume,  $^{9}$ RsD = Resistive density,  $^{10}$ XcD = Reactive density.

467 \* *P* < 0.05.

468 \*\* *P* < 0.01.

469 \*\*\* *P* < 0.001.

472 Predicting models of lamb body composition by *in vivo* bioimpedance analysis.

	Madala		Calibration					Cross-validation			
	Models	R <sup>2a</sup>	RMSE⁵	Ср <sup>с</sup>	<i>P</i> -value	$R^{2a}$	RMSE <sup>b</sup>	$MPE^d$	RMPE <sup>e</sup> ,%	<i>P</i> -value	
Moisture, kg	Y = 0.66 + 0.94XcD +0.09V	0.84	0.55	17.6	< 0.0001	0.93	0.50	0.43	7.47	< 0.0001	
Moisture, %	Y = 82.2 – 0.45RsD – 0.19V	0.79	1.92	2.32	< 0.0001	0.86	2.59	1.82	2.78	< 0.0001	
Protein, kg	Y = - 0.70 + 0.05RsD + 0.03V + 0.07PA	0.91	0.15	5.32	< 0.0001	0.95	0.15	0.12	6.97	< 0.0001	
Fat, kg	Y = - 2.11 + 0.10RsD + 0.04V	0.87	0.30	3.56	< 0.0001	0.91	0.39	0.29	20.9	< 0.0001	
Fat, %	Y = - 3.69 + 0.49RsD + 0.21V	0.79	2.09	4.70	< 0.0001	0.88	2.66	1.76	12.5	< 0.0001	
Lean mass, kg	Y = - 1.90 + 0.11V + 0.18RsD + 0.31PA	0.89	0.64	10.3	< 0.0001	0.93	0.65	0.55	7.36	< 0.0001	
Lean mass, %	Y = 102.5 – 0.48RsD – 0.21V	0.79	2.06	4.56	< 0.0001	0.88	2.65	1.75	2.05	< 0.0001	

473 <sup>a</sup>R<sup>2</sup> = Coefficient of determination; <sup>b</sup>RMSE = Root mean squared error; <sup>c</sup>Cp = Mallows Cp statistic; <sup>d</sup>MPE = Mean prediction error; <sup>e</sup>RMPE = Relative

474 mean prediction error.

475 XcD = reactive density, V = body bioelectric volume, RsD = resistive density, PA = phase angle.

476



Fig. 1. Placement of transmitter and detector electrodes in live lambs for resistance and reactance measurements.