Animal Journal- Supplementary material

Meat quality of farmed red deer fed a balanced diet: effects of supplementation with copper bolus on different muscles

M. P. Serrano 1,2,3, A. Maggiolino 4, J. M. Lorenzo 5, P. De Palo 4, A. García 1,2,3, T. Landete-Castillejos 1,2,3, P. Gambín 1,2,3, J. Cappelli 1,2,3, R. Domínguez 4, F. J. Pérez-Barbería 1,2,3 and L. Gallego 1,2,3

1*Animal Science Techniques Applied to Wildlife Management Research Group, Instituto de Investigación en Recursos Cinegéticos, Albacete Section of CSIC-UCLM-JCCM, Universidad de Castilla-La Mancha (Campus Universitario sn, 02071, Albacete), Spain*

2*Sección de Recursos Cinegéticos y Ganaderos, Instituto de Desarrollo Regional of Universidad de Castilla-La Mancha (Campus Universitario sn, 02071, Albacete), Spain*

3*Departamento de Ciencia y Tecnología Agroforestal y Genética, Escuela Técnica Superior de Ingenieros Agrónomos y de Montes of Universidad de Castilla-La Mancha (Campus Universitario sn, 02071, Albacete), Spain*

4 *Department of Veterinary Medicine, University of Bari A. Moro, Italy, S.P. per Casamassima, km 3, 70010, Valenzano, Bari, Italy*

5*Centro Tecnológico de la Carne de Galicia, Parque Tecnológico de Galicia (Rúa Galicia 4, 32900, San Cibrán das Viñas, Ourense), Spain*

Corresponding author: Martina Pérez Serrano. E-mail: Martina.Perez@uclm.es

Short title: Cu supplementation and meat quality of farmed deer

**Supplementary Table S1** *Statistical model codes for quality characteristics, chemical composition and mineral content of meat from red deer yearling males1*

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  |  | Intercept | *R*2 | Adjusted *R*2 |
| Dependent variable |  | RMSE | F-value | *P*-value |
| pH at 24 h *post mortem* |  |  6.6 |  211.9 | \*\*\* | 0.283 |  0.185 |
| *Meat colour* |  |  |  |  |  |  |
|  Lightness (*L\**) |  |  484.9 |  39.1 | \*\*\* | 0.143 |  0.026 |
|  Redness (*a\**) |  |  5.8 |  1.2 | Ns | 0.298 |  0.202 |
|  Yellowness (*b*\*) |  |  11.8 |  4.4 | \* | 0.233 |  0.129 |
|  Chroma (*c*\*) |  |  18.7 |  4.4 | \* | 0.368 |  0.282 |
|  Hue angle (*H*º) |  |  0.12 |  11.0 | \*\* | 0.014 | -0.120 |
| *Chemical composition (%)* |  |  |  |  |  |  |
|  Moisture |  | 1006 8 |  1766 4 | \*\*\* | 0.549 |  0.488 |
|  Protein |  |  90.1 |  270.6 | \*\*\* | 0.610 |  0.557 |
|  Intramuscular fat |  |  1.8 |  4.4 | 0.05 | 0.109 | -0.018 |
|  Ash |  |  0.69 |  33.2 | \*\*\* | 0.261 |  0.160 |
| Cholesterol (mg/100 g sample) |  |  603.4 |  20.6 | \*\*\* | 0.383 |  0.299 |
| Mineral content |  |  |  |  |  |  |
|  Calcium (mg/kg) |  |  5.30 |  43.9 | \*\*\* | 0.298 |  0.202 |
|  Potassium (mg/kg) |  | 7882 3 |  57.6 | \*\*\* | 0.195 |  0.085 |
|  Magnesium (mg/kg) |  |  87.2 |  14.4 | \*\*\* | 0.297 |  0.201 |
|  Sodium (mg/kg) |  | 1318 0 |  34.0 | \*\*\* | 0.050 | -0.079 |
|  Phosphorus (mg/kg) |  | 5880 6 |  157.0 | \*\*\* | 0.328 |  0.237 |
|  Copper (mg/kg) |  |  0.009 |  6.2 | \* | 0.094 | -0.030 |
|  Iron (mg/kg) |  |  0.49 |  19.9 | \*\*\* | 0.144 |  0.027 |
|  Manganese (µg/100 g) |  |  140.3 |  3.2 | 0.09 | 0.163 |  0.049 |
|  Zinc (mg/kg) |  |  0.77 |  14.9 | \*\*\* | 0.599 |  0.545 |

1 The experimental unit was the sample (*n* = 10 and 16 for Cu bolus supplemented and control groups, respectively and *n* = 13 for *Sternocephalicus* and *Rectus Abdominis* muscles).

The *P*-values are indicated by \*, \*\* and \*\*\* for *P* < 0.05, *P* < 0.01 and *P* < 0.001, respectively.

**Supplementary Table S2** *Statistical model codes for* *fatty acid profile (mg/g of fat) of red deer yearling males1*

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  |  | Intercept | *R*2 | Adjusted *R*2 |
| Dependent variable |  | RMSE | F-value | *P*-value |
| C10:0 |  |  0.000 |  1.6 | Ns | 0.104 | -0.018 |
| C12:0 |  |  0.001 |  0.41 | Ns | 0.299 |  0.203 |
| C13:0 |  |  0.000 |  2.5 | Ns | 0.202 |  0.093 |
| C14:0 |  |  2.8 |  5.7 | \* | 0.174 |  0.062 |
| C14:1n‒5 |  |  0.27 |  0.67 | Ns | 0.306 |  0.211 |
| C15:0 |  |  0.14 | 25.9 | \*\*\* | 0.222 |  0.116 |
| C15:1n‒5 |  |  0.003 |  7.3 | \* | 0.324 |  0.231 |
| C16:0 |  | 180.7 | 76.1 | \*\*\* | 0.311 |  0.217 |
| C16:1n‒7 |  |  24.0 |  4.2 | 0.05 | 0.288 |  0.191 |
| C17:0 |  |  0.11 | 43.9 | \*\*\* | 0.197 |  0.088 |
| C17:1n‒7 |  |  0.07 | 22.0 | \*\*\* | 0.153 |  0.038 |
| C18:0 |  |  17.3 | 13.6 | \*\*\* | 0.224 |  0.118 |
| C18:1n‒7 |  |  2.4 |  4.8 | \* | 0.165 |  0.051 |
| C18:1n‒9 |  |  40.2 | 34.6 | \*\*\* | 0.106 | -0.015 |
| *9t-*C18:1 |  |  0.009 |  6.1 | \* | 0.175 |  0.062 |
| *11t-*C18:1 |  |  2.1 |  4.2 | 0.05 | 0.038 | -0.093 |
| C18:2n‒6 |  |  39.2 |  7.5 | \* | 0.319 |  0.226 |
| *9c,11t*-C18:2 |  |  2.1-5 |  0.002 | Ns | 0.082 | -0.043 |
| C18:3n‒3 |  |  0.06 | 24.4 | \*\*\* | 0.413 |  0.333 |
| C18:3n‒6 |  |  0.001 |  4.5 | \* | 0.231 |  0.127 |
| C20:0 |  |  9.1-5 |  4.2 | 0.05 | 0.562 |  0.502 |
| C20:1n‒9 |  |  0.002 |  8.8 | \*\* | 0.045 | -0.085 |
| C20:2n‒6 |  |  0.001 |  3.0 | Ns | 0.441 |  0.364 |
| C20:3n‒3 |  |  1.5-5 |  0.14 | Ns | 0.038 | -0.093 |
| C20:3n‒6 |  |  0.03 |  7.2 | \* | 0.376 |  0.291 |
| C20:4n‒6 |  |  3.5 |  4.6 | \* | 0.493 |  0.423 |
| C20:5n‒3 |  |  0.006 |  3.2 | 0.09 | 0.165 |  0.051 |
| C21:0 |  |  0.000 |  1.4 | Ns | 0.201 |  0.092 |
| C22:0 |  |  0.002 |  3.0 | Ns | 0.435 |  0.358 |
| C22:1n‒9 |  |  2.7-5 |  0.45 | Ns | 0.196 |  0.086 |
| C22:5n‒3 |  |  0.04 |  2.9 | Ns | 0.518 |  0.452 |
| C22:6n‒3 |  |  0.001 |  0.76 | Ns | 0.399 |  0.317 |
| C23:0 |  |  0.002 |  2.1 | Ns | 0.379 |  0.294 |
| C24:0 |  |  5.7-6 |  0.11 | Ns | 0.232 |  0.127 |
| SFA2 |  | 403.1 | 73.3 | \*\*\* | 0.083 | -0.043 |
| MUFA3 |  | 232.2 | 14.9 | \*\*\* | 0.202 |  0.093 |
| PUFA4 |  |  79.7 |  7.0 | \* | 0.386 |  0.303 |
| PUFA/SFA |  |  0.04 |  5.2 | \*\* | 0.396 |  0.313 |
| ∑ n‒6 |  |  70.3 |  6.9 | \* | 0.375 |  0.290 |
| ∑ n‒3 |  |  0.30 |  5.8 | \* | 0.421 |  0.342 |
| n‒6/n‒3 |  |  43.8 | 37.8 | \*\*\* | 0.048 | -0.082 |
| NV5 |  |  0.23 | 10.8 | \*\* | 0.536 |  0.472 |
| h/H6 |  |  0.18 | 13.3 | \*\*\* | 0.489 |  0.419 |
| IA7 |  |  0.14 | 12.8 | \*\* | 0.196 |  0.086 |
| IT8 |  |  0.39 | 23.2 | \*\*\* | 0.170 |  0.057 |

1 The experimental unit was the sample (*n* = 10 and 16 for Cu bolus supplemented and control groups, respectively and *n* = 13 for *Sternocephalicus* and *Rectus Abdominis* muscles).

2 Saturated fatty acids.

3 Monounsaturated fatty acids.

4 Polyunsaturated fatty acids.

5 Nutritional value = Σ(C12:0+C14:0+C16:0)/Σ(C18:1n‒9+C18:2n‒6).

6 Hypocholesterolemic/hypercholesterolemic ratio = [Σ(C18:1n‒9+C18:1n‒7+C18:2n‒6+C18:3n‒3+C20:3n‒6+C20:4n‒6)/Σ(C14:0+C16:0)].

7 Index of atherogenicity = [C12:0+(4\*C14:0)+C16:0]/[(∑MUFA)+(∑PUFA)].

8 Index of thrombogenicity = [C14:0+C16:0+C18:0]/[(0.5\*∑MUFA)+(0.5\*n‒6)+(3\*n‒3)+(n‒3/n‒6)].

The *P*-values are indicated by \*, \*\* and \*\*\* for *P* < 0.05, *P* < 0.01 and *P* < 0.001, respectively.

**Supplementary Table S3** *Statistical model codes for* *amino acid (AA) content of meat (mg/100 g of sample) of red deer yearling males1*

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  |  | Intercept | *R*2 | Adjusted *R*2 |
| Dependent variable |  | RMSE | F-value | *P*-value |
| *Essential* |  |  |  |  |  |  |
|  Histidine |  |  101 726 |  19.2 | \*\*\* | 0.162 |  0.048 |
|  Isoleucine |  |  210 525 |  18.9 | \*\*\* | 0.027 | -0.106 |
|  Leucine |  |  663 427 |  21.0 | \*\*\* | 0.017 | -0.117 |
|  Lysine |  |  748 073 |  18.8 | \*\*\* | 0.027 | -0.106 |
|  Methionine |  |  1 007 |  0.25 | Ns | 0.122 |  0.003 |
|  Phenylalanine |  |  117 783 |  13.1 | \*\* | 0.062 | -0.066 |
|  Threonine |  |  241 138 |  17.5 | \*\*\* | 0.034 | -0.098 |
|  Valine |  |  257 162 |  22.0 | \*\*\* | 0.030 | -0.102 |
|  Tyrosine |  |  51 504 |  8.5 | \*\* | 0.269 |  0.169 |
|  Cysteine |  |  7 093 |  7.9 | \*\* | 0.034 | -0.098 |
|  Total essentials |  |  22 924905 |  21.2 | \*\*\* | 0.024 | -0.109 |
| *Non-essential* |  |  |  |  |  |  |
|  Arginine |  |  917 177 |  15.7 | \*\*\* | 0.068 | -0.059 |
|  Alanine |  |  498 809 |  26.1 | \*\*\* | 0.087 | -0.038 |
|  Aspartic acid |  |  792 684 |  23.0 | \*\*\* | 0.009 | -0.126 |
|  Glutamic acid |  |  2118 793 |  20.0 | \*\*\* | 0.019 | -0.115 |
|  Glycine |  |  334 069 |  18.7 | \*\*\* | 0.235 |  0.131 |
|  Proline |  |  280 567 |  32.5 | \*\*\* | 0.240 |  0.137 |
|  Serine |  |  128 879 |  19.7 | \*\*\* | 0.039 | -0.093 |
|  Total non-essentials |  | 2464 2528 |  25.9 | \*\*\* | 0.046 | -0.084 |
| Total AA |  | 9423 5779 |  24.0 | \*\*\* | 0.023 | -0.110 |
| Essential/non-essential |  |  0.18 | 223.9 | \*\*\* | 0.391 |  0.308 |

1 The experimental unit was the sample (*n* = 10 and 16 for Cu bolus supplemented and control groups, respectively and *n* = 13 for *Sternocephalicus* and *Rectus Abdominis* muscles).

The *P*-values are indicated by \*\* and \*\*\* for *P* < 0.01 and *P* < 0.001, respectively.

**Supplementary Table S4** *Statistical model codes for mineral content of liver from red deer*

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
|  |  | Intercept | *R*2 | Adjusted *R*2 |
| Dependent variable |  | RMSE | F-value | *P*-value |
| *Macromineral content (g/100 g)* |  |  |  |  |  |  |
|  Calcium |  | 4.9-8 |  0.44 | Ns | 0.528 |  0.434 |
|  Potassium |  | 0.006 |  20.1 | \*\*\* | 0.030 | -0.163 |
|  Magnesium |  | 1.7-5 | 124.4 | \*\*\* | 0.441 |  0.301 |
|  Sodium |  | 0.000 |  15.7 | \*\* | 0.430 |  0.316 |
|  Phosphorus |  | 0.010 |  7.8 | \* | 0.174 |  0.009 |
|  Sulphur |  | 0.004 |  22.3 | \*\*\* | 0.176 |  0.011 |
| *Trace mineral content (mg/kg)* |  |  |  |  |  |  |
|  Aluminium |  | 0.064 |  0.98 | Ns | 0.344 |  0.212 |
|  Bismuth |  | 0.002 |  14.5 | \*\* | 0.593 |  0.503 |
|  Boron |  | 0.002 |  9.3 | \* | 0.474 |  0.369 |
|  Cadmium |  | 9.1-6 |  1.0 | Ns | 0.215 |  0.058 |
|  Cobalt |  | 0.001 |  6.8 | \* | 0.192 |  0.031 |
|  Chrome |  | 0.000 |  1.8 | Ns | 0.056 | -0.133 |
|  Copper |  | 604.0 |  3.1 | Ns | 0.027 | -0.168 |
|  Iron |  | 341.6 |  1.1 | Ns | 0.063 | -0.124 |
|  Lithium |  | 0.009 |  5.0 | 0.05 | 0.341 |  0.209 |
|  Manganese |  | 3.1 |  26.8 | \*\*\* | 0.164 | -0.003 |
|  Molybdenum |  | 0.23 |  12.3 | \*\* | 0.013 | -0.184 |
|  Nickel |  | 9.6-5 |  4.2 | 0.07 | 0.472 |  0.366 |
|  Lead |  | 1.3-5 |  0.045 | Ns | 0.615 |  0.538 |
|  Rubidium |  | 2.7 |  2.0 | Ns | 0.006 | -0.193 |
|  Selenium |  | 0.012 |  6.0 | \* | 0.118 | -0.058 |
|  Strontium |  | 0.000 |  1.8 | \* | 0.492 |  0.390 |
|  Titanium |  | 6.5-5 |  1.3 | \* | 0.423 |  0.307 |
|  Thallium |  | 0.13 |  45.2 | \*\*\* | 0.498 |  0.373 |
|  Zinc |  | 254.8 |  10.5 | \*\* | 0.050 | -0.140 |

The *P*-values are indicated by \*, \*\* and \*\*\* for *P* < 0.05, *P* < 0.01 and *P* < 0.001, respectively.