

1 **Study of ingredients and nutrient composition of commercially available treats for dogs**

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3 Giada Morelli,¹ Eleonora Fusi,² Sandro Tenti,¹ Lorenzo Serva,¹ Giorgio Marchesini,¹ Marianne
4 Diez,³ Rebecca Ricci¹

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6 ¹Department of Animal Medicine, E-mail for correspondence: Production and Health, University of
7 giada.morelli@phd.unipd.it Padua, Legnaro, Italy

8 ²Department of Health, Animal Science and Food Safety, University of Milan, Milan, Italy

9 ³Department of Animal Productions, Liege University, Liege, Belgium

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11

12 **Abstract**

13 Forty-one dog treats were selected from the market with the aim of providing more insight into
14 supplemental pet food composition. Thirty-two products (four biscuits, nine tender treats, two meat-
15 based strips, five rawhides, eight chewable sticks, four dental care sticks) were analysed for proximate
16 nutrient composition and quantification of minerals, hydroxyproline (Hyp), starch, glucose, fructose
17 and sucrose. Labelled ingredients were often expressed as non-specific categories. A treat supplied a
18 mean of 332.0±39.2kcal metabolisable energy (ME)/100g, and the most energy-dense product was a
19 tender treat (475.0 kcal ME/100g). Small dogs receive the highest percentage of maintenance energy
20 requirement when producers' feeding instructions are followed. Treat categories revealed variability
21 in dry matter, crude protein, ash, Hyp and starch. Rawhides showed the highest Hyp content. Simple
22 sugars were identified in most treats, and sucrose was the most prevalent. Results of the study suggest
23 treat labelling should include more information on the ingredients used, and the varying nutrient and
24 caloric density of treats should be considered. Specific attention should be given to the use of treats
25 in dogs with specific ingredient sensitivities or nutrient considerations.

26

27 **Introduction**

28

29 Dog treats represent the fastest growing segment of the pet food industry today, and nearly every pet
30 food brand produces many types of treats.¹ The latest EU regulation² states that dog treats should be
31 labelled as ‘complementary feed’, which is legally defined as ‘compound feed which has a high
32 content of certain substances but which, by reason of its composition, is sufficient for a daily ration
33 only if used in combination with other feed’. European feed law also establishes rules and
34 requirements for labelling to provide adequate information for consumers.²

35 Little is known about the nutritional value of treats and their impact on the dog’s diet, health and
36 wellness despite the popularity of such products. Previous studies performed on treats have assessed
37 their microbiological quality with specific regard to the potential risk of bacteria¹ contamination for
38 dogs and human beings rather than evaluating their chemical composition.³⁻⁶ Moreover, treats are not
39 intended to contribute significantly to the daily ration, but they may be given in quantities that impact
40 total energy intake. Therefore, feeding instructions should provide clear recommendations on how
41 not to overfeed dogs as suggested by FEDIAF Nutritional Guidelines for Complete and
42 Complementary Pet Food for Cats and Dogs.⁷ The literature^{8,9} clearly demonstrated treats to be a risk
43 factor for the development of excess body fat in dogs.

44 The aim of this study was to provide greater insight into the nutrient composition and ingredients
45 used in the production of dog treats given the scarce information presently available in the literature.
46 A secondary aim was to verify whether producer’s daily intake recommendations on the label were
47 in accordance with the common recommendation that treats should not exceed 10 per cent of
48 maintenance energy requirement (MER).¹⁰ Analyses were inclusive of quantification of minerals,
49 starch, simple sugars and the amino acid hydroxyproline.

50 These analytes were selected to provide information about protein quality and the presence of
51 potentially high amounts of minerals or simple sugars, which may be of importance in managing
52 certain nutritionally responsive disease conditions.

53 **Materials and methods**

54

55 *Sample recruitment and classification*

56 Forty-one dog treats of different international pet food brands were collected from different stores
57 (pet shops and supermarkets); the most popular and purchased ones, according to the shop assistants,
58 were chosen. The selected samples were divided into six categories as follows: five biscuits, ten
59 tender treats, three meatbased strips, five rawhides, twelve chewable sticks and six sticks for dental
60 care. Biscuits included dry treats mainly made of cereals, similar to those manufactured for human
61 consumption; tenders were small semimoist treats having meat and cereals as main ingredients; meat-
62 based strips were rectangular pieces of dry meat; rawhides were bone-shaped treats made of bovine
63 skin; chewable sticks were semimoist to dry sticks made of various ingredients; and dental care sticks
64 were identified by a label that reported an oral health benefit.

65

66 *Label reading*

67 Labels of the selected samples were examined and the following data were recorded using
68 commercially available software (Excel, Microsoft): product name, brand, number and type of
69 ingredients and their label order, analytical composition (moisture, when stated; crude protein (CP);
70 ether extract (EE); crude fibre (CF); ash), net weight and/or number of pieces per pack, recommended
71 quantities of treats/day, and place of production. Nitrogen-free extract (NFE) was calculated from
72 label information (100 per cent – moisture – crude protein – crude fat – crude fibre – ash).

73 For each sample, metabolisable energy (ME, expressed as kcal/100g) was calculated using the
74 predictive equation for energy content based on the ‘modified Atwater’ factors of 3.5, 8.5 and 3.5 on
75 as-fed basis for protein, fat and NFE, respectively.⁷ A value of 10 per cent moisture was assumed if
76 moisture was not stated in the label because regulations do not require a moisture declaration if the
77 content is less than 14 per cent (according to European Regulation (EC) No 767/2009).² If more than
78 one treat was contained in a package, two or three were weighed in order to obtain the mean weight

79 of each sample. The mean weight of each treat (expressed as g) was then multiplied by the calculated
80 ME/g value, and the mean caloric amount (kcal/piece) was obtained as the producers are not required
81 to report it in the label according to the EU regulation (EC 767/2009).² Feeding recommendations
82 (number of pieces/day), if provided on a label (as it is not mandatory in EU according to EC
83 767/2009),² were converted into percentages of MER for a small, a medium and a large dog in order
84 to investigate whether any treats exceeded 10 per cent. A 7-kg dog, a 15-kg dog and a 30-kg dog were
85 chosen as reference bodyweight dogs, and their daily energy requirements were calculated according
86 to the formula $95 \times \text{kg Body Weight}^{0.7511}$: 409 kcal, 724 kcal and 1218 kcal, respectively. The
87 production site, if not explicitly reported, was obtained from the corporate code printed on the label.

88

89 **Chemical analyses**

90 Thirty-two of the 41 products (four biscuits, nine tender treats, two meat-based strips, five rawhides,
91 eight chewable sticks and four dental sticks) were sent to the Chemical–NIRs–XRF Laboratory of
92 the University of Padua for proximate analysis (dry matter, DM; CP; EE; CF; ash), and quantification
93 of minerals (Calcium (Ca); Phosphorus (P); Sodium (Na); Potassium (K); Magnesium (Mg)),
94 hydroxyproline (Hyp), starch and simple sugars (glucose; fructose; sucrose).

95

96 *Proximate analysis and mineral quantification*

97 The DM content was determined by oven-drying previously ground samples at 105°C for 24 hours.
98 CP content was determined according to the Kjeldahl method (EC 152/2009—annex III method C)
99 and calculated using a nitrogen conversion factor of 6.25. EE analysis was performed according to
100 method H, procedure B reported in annex III of the EC 152/2009, by Soxhlet extraction using
101 petroleum ether. The CF was obtained by the Weende method according to the same regulation (EC
102 152/2009—annex III method I). Defatted samples were treated with boiling solutions of sulphuric
103 acid and potassium hydroxide, and the residue was separated by filtration on sintered glass, washed,
104 dried, weighed and heated in a furnace at 475°C–500°C. Weight loss after combustion was expressed

105 as CF. The ash content was measured gravimetrically after combustion at 550°C until white, light
106 grey or reddish ash was obtained, and subsequently cooled to environmental temperature (EC
107 152/2009—annex III method A). Mineral analyses (Ca; P; Na; K; Mg) were performed by the
108 Inductively Coupled Plasma – Optical Emission Spectrometry (ICPOES) method (Spectro EOP,
109 Ciros Vision) after microwave digestion (Association of Official Analytical Chemists 2000, 999.10).

110

111 *Hyp quantification*

112 Hyp determination in previously hydrolysed samples was performed by high-performance liquid
113 chromatography (HPLC) technique following the previously published recommendations.^{12 13} A
114 prior preparation was required to ensure the block of primary amine groups with o-phthalaldehyde
115 and the derivatisation of the molecules with secondary amine groups, among which Hyp, with 9-
116 fluorenylmethyloxycarbonyl chloride which developed highly fluorescent, stable compounds.
117 Chromatographic analysis was performed using Shimadzu Series 10 Avp equipment with fluorimetric
118 revelator based on the following operating conditions: SAX column 5µm, 150mm, inner diameter
119 4.6mm (Eurisco Diagnostica); mobile phase: mixture water/acetonitrile/buffer in the ratio of 45/50/5;
120 injection volume: 10µL; room temperature; max work pressure 1800psi; mobile phase flow:
121 1.2mL/minute; fluorimeter RF10AXL, 260nm λ of emission and 330nm λ of excitement; and
122 analytical time: 10minutes, Hyp reading at minute 5.4, and internal standard at minute 6.4. The
123 volume (expressed as µL) of Hyp per litre of injected solution was multiplied by its own molecular
124 weight (131.11g/mol) and by appropriate coefficients derived from the dilution ratio and the DM
125 contained in the sample. The total content of Hyp was therefore expressed as milligram per 100g of
126 sample. The Hyp content obtained by analysis permitted an indirect estimate of the treats' collagen
127 content using the formula $\text{Hyp (\%)} \times 7.14$.

128

129 *Starch and sugar quantification*

130 The starch content of each sample was quantified by HPLC technique using Shimadzu SLC10 Avp

131 equipment with refractometric detector Refractive Index Detector 10A and software class VP 7 based
132 on the following operating conditions: refractometer equipped with thermostatted cell at 40°C; H₂SO₄
133 0.0025 n; 0.6 mL/minute flow; analytical time: 10 minutes; injection volume: 20 μL; and Bio-Rad
134 column HPX-87H-300×7.8 mm thermostatted at 90°C. Each sample (100 mg per glass tube) was
135 finely milled with a 0.5 mm grid and had the following added: 3.9 mL of acetate buffer 0.1 M, pH
136 4.2 and α-amylase. The glass tubes were then placed in a water bath at 80°C for 15 minutes. After
137 allowing cooling at room temperature, 1 mL of enzymatic solution (amyloglucosidase 5 g/L, Sigma
138 A-7255 from Rhizopus mould) was added. The samples were then incubated in a stove at 40°C for
139 24 hours, centrifuged at 3500 g/ minute for 10 minutes and filtered with a 0.45-μm disposable filter.
140 The refractometer determined the amount of glucose resulting from starch hydrolysis and from the
141 glucose already contained in the sample, corrected according to the following formula:

$$142 \quad \% \text{ Starch as is} = X \sim 0.9 \sim 10 / \text{weighted value} \sim 100$$

143 where X=glucose determined by HPLC or possibly corrected by the glucose contained in the sample
144 before analysis; 0.9=glucose molecular weight (MW)/ glucose MW+H₂O ratio; and 10=sample
145 dilution. The quantification of simple sugars (glucose, fructose and sucrose) was carried out
146 according to Charles¹⁴ by HPLC technique using a Shimadzu SCL10 Avp system with refractometric
147 detector RID 10A and software class VP 7 based on the following operating conditions: refractometer
148 equipped with thermostatted cell at 40°C; H₂O; 0.6 mL/minute flow; analytical time: 20 minutes;
149 injection volume: 20 μL; and Bio-Rad column HPX-87C-300×7.8 mm thermostatted at 90°C. Every
150 sample was extracted with aqueous solution, centrifuged and filtered with a 0.45-μm disposable filter.
151 The refractometer determined the total amount of glucose, fructose and sucrose in the sample,
152 expressed as mg/mL, according to the following formula:

$$153 \quad \text{Simple sugar(g/100 g of the sample)} = (\text{extracted sample(mg/mL)}) \text{ extraction volume/sample} \\ 154 \quad \text{weight(g)} \sim 100/1000$$

155 The amount of glucose obtained with this analysis was subtracted from the amount of starch to reduce
156 the over-rated error of the starch due to the presence of free sugar in the sample. Sucrose was not
157 hydrolysed; therefore, the amounts of free glucose and fructose were not overestimated.

158

159 **Results**

160

161 *Information collected from labels*

162 Ten treats (24 per cent) were composed of one to three ingredients; 20 (49 per cent) were composed
163 of four to six ingredients, and 11 (27 per cent) were composed of seven to nine ingredients. The
164 composition of the treats was indicated on the label by a list of specific ingredients and in most cases
165 by category of ingredients (eg, ‘meat and animal derivatives’, ‘cereals’). The most widely represented
166 ingredient categories were ‘meat and animal derivatives’ (n=35), ‘vegetable by-products’ or
167 ‘vegetable protein extracts’ (n=32), ‘cereals’ (n=29), ‘minerals’ (n=27), ‘oils and fats’ (n=22) and
168 ‘sugars’ (n=17). Minor ingredients included ‘glycerol’/‘glycerin’ or ‘sorbitol’ (n=12), which are used
169 as sweeteners and emulsifiers, ‘yeasts’ (n=5), ‘milk and milk derivatives’ (n=5), ‘fish and fish by-
170 products’ (n=4), ‘seeds’ (n=4), ‘aromatic plants’ (n=4), ‘eggs and egg derivatives’ (n=2) and
171 ‘additives’ (n=1). ‘Meat and animal derivatives’ was the first ingredient in 21 products and ‘cereals’
172 in 18. The second leading ingredient categories listed were ‘vegetable by-products’ or ‘vegetable
173 proteins extracts’ (n=14), ‘meat and animal derivatives’ (n=9) and ‘cereals’ (n=8). The third
174 ingredient was most commonly ‘vegetable by-products’ (n=8), ‘sugars’ (n=7), ‘minerals’ (n=6) and
175 ‘vegetable protein extracts’ (n=3). Biscuits and dental sticks had ‘cereals’ mentioned as the first
176 ingredient of the list, while tenders, meat strips, rawhides and chewable sticks had ‘meat and animal
177 derivatives’ as the first.

178 The caloric density of each category of treat (kcal ME/100g) calculated from the label is reported in
179 Table 1. The most calorically dense treats were biscuits, whereas the least calorically dense were
180 dental sticks. A treat supplied 303.5 ± 46.8 kcal ME/100g on average.

181 When caloric density was expressed as kcal/treat, rawhides were the heaviest and the most energy-
182 dense products, followed by chewable sticks and dental sticks (Table 1). The feeding instructions
183 were reported in the label of 21 products, none of which was a rawhide (so one piece per day was
184 assumed). If the feeding instructions were followed, biscuits and rawhides provide the highest
185 percentage of MER for small-sized and medium-sized dogs, while biscuits and chewable sticks
186 provide the highest percentage for big-sized dogs (Table 2). On average, biscuits accounted for 16
187 per cent of MER for dogs of any size; rawhides exceeded 25 per cent MER for small-sized dogs and
188 18 per cent MER for medium-sized dogs; chewable sticks surpassed 10 per cent MER for all size
189 dogs, reaching 16.9 per cent MER in small-sized dogs; the feeding instructions of dental sticks
190 remained below 10 per cent MER for every dog size.

191 The most common origin country of the selected treats was Germany (35 per cent of the samples),
192 followed by China (18 per cent), Hungary (13 per cent), UK (10 per cent), Italy (8 per cent), Holland
193 (5 per cent), Thailand (5 per cent), France (3 per cent) and Spain (3 per cent).

194

195 *Chemical analysis of 32 treats*

196 The results of the chemical analyses divided by treat category are shown in Table 3. In agreement
197 with the labels reading in the previous section, the proximate analysis revealed that biscuits are the
198 most caloric treat type with 351.3 ± 16.5 kcal ME/100g. All categories showed DM of greater than 80
199 per cent, with biscuits showing the highest and meat strips the lowest mean value. Tenders also
200 showed the most variable DM content, with the lowest and highest values recorded among all treats.
201 Rawhides had the highest CP value, while tenders had the highest fat content. Selected treats were
202 generally low in fibre, ranging from 0.2 to 13.9g/1000 kcal. Rawhides showed the lowest ash level,
203 whereas the remaining categories ranged from 17 to 25g/1000 kcal on average, with a tender product
204 reaching up to 42.5g/1000 kcal. Although high variability among treat categories was detected also
205 in Ca, P, K, Mg and Na, no significant differences were apparent.

206 The treat category with the highest Hyp content was rawhide; chewable sticks showed a high mean
207 level (6.9 ± 9.9 g/1000 kcal), while the other categories recorded an average Hyp content lower than
208 2.5g/1000 kcal.

209 Hyp/CP ratio calculation showed that Hyp accounted for 19.1 per cent of the protein content of
210 rawhides on average, 4.6 per cent of chewable sticks, 3.2 per cent of dental sticks, 2.1 per cent of
211 meat strips, 1.7 per cent of tenders and 1.0 per cent of biscuits. Estimated collagen, expressed as
212 average per treat category, was distributed as follows: 83.4 per centDM in rawhides, 20.0 per centDM
213 in chewable sticks, 6.1 per centDM in meat strips, 4.1 per centDM in tenders, 2.0per cent DM in
214 dental sticks and 1.3 per centDM in biscuits. Starch was one of the nutrients with the highest
215 variability: dental sticks and biscuits showed the highest content, while rawhides had no starch. A
216 wide variability in sucrose content was observed among treat categories, and the maximum
217 concentrations detected were 35.9g/1000 kcal in a biscuit and 51.7g/1000 kcal in a tender treat.

218 Low glucose and fructose concentrations were found in most treats, and no significant differences
219 among treat types emerged. Rawhides contained no measurable sugars.

220

221 **Discussion**

222

223 Literature addressing the nutrient composition of common treats is lacking. Most studies conducted
224 on these products assessed whether any bacterial contamination poses risks to consumers and possibly
225 to pet owners.³⁻⁶ For example, Freeman and others⁵ collected 26 bully stick treats for dogs, but beyond
226 microbiological testing, only five were submitted to proximate analysis. Therefore, our study is the
227 first that reports and compares the nutrient composition of different categories of treats. The findings
228 demonstrated that dog treats varied widely in chemical composition among the categories considered
229 and even among products in the same category.

230 Information on the label is an important benchmark in feeding practices for both owners and
231 veterinarians.

232 This study revealed that 76 per cent of the selected treats contained from four to nine ingredients, and
233 that ingredients were not precisely described on the label. It is significant that the ingredient
234 categories listed most often in treats were ‘meat and meat derivatives’, ‘vegetable by-products’ and
235 ‘cereals’, none of which permitted the identification of the precise animal or plant species in question.
236 Categories of ingredients are allowed to be listed on complementary pet food rather than single and
237 specific ingredients, according to the European Regulation (EC) No 767/2009 – Article 17 (2) I,² and
238 this lack of clarity confirms that the use of these products should be strictly avoided with dogs
239 undergoing an elimination diet whenever adverse food reactions may be suspected.¹⁵

240 Interestingly, many treats are composed of ingredients not usually seen in maintenance pet food, such
241 as ‘milk and milk derivatives’, and ‘sugars’ and sweeteners such as glycerol, glycerin and sorbitol,
242 which were listed in the labels of many products (almost half of products mentioned ‘sugars’ on the
243 label’s ingredient list). It is well-known that dogs appreciate sweet taste,¹⁶ so sugars may have been
244 added by producers to increase treat palatability (although glycerol, glycerin and sorbitol are also
245 used as emulsifiers or humectants). ‘Milk and milk derivatives’ was listed in the labels of five of the
246 selected treats, and this should be taken into consideration when feeding treats to dogs with known
247 intolerance to these foods and their derivatives. The ingredients on the label must be listed in
248 descending order by weight, inclusive of water weight, as per Regulation (EC) No 767/2009 Article
249 17²; in the treats selected in this study, it is clearly evident that the ingredients at the top of the list
250 are ‘meat and meat derivatives’ and ‘cereals’.

251 The energy value of treats was previously investigated in two studies.^{5 17} The former study included
252 only three products; however, among those, a rawhide with a reported digestible energy of 334.4
253 kcal/100g is unfortunately incomparable with the mean ME value we calculated using ‘modified
254 Atwater’ factors for our five rawhides (309.5 kcal/100g). Freeman’s study calculated the ME content
255 to a mean value of 301 kcal/100g using ‘modified Atwater’ factors in five bully sticks, a type of
256 product which was not included in our study because it is unavailable in the Italian market. According
257 to the World Small Animal Veterinary Association (WSAVA) nutritional assessment guidelines,¹⁰

258 daily treat intake should not exceed 10 per cent of the dog's energy requirement in order to avoid
259 dietary imbalances.

260 The MER estimation calculated in the present study for a small-sized, medium-sized and large-sized
261 dog revealed that producers should reconsider the feeding instructions (number of treats/day) they
262 provide on labels, especially for small dogs. This result agrees with the finding of Freeman and
263 others,⁵ who demonstrated that an average-sized 6' bully stick provides 30 per cent of the daily calorie
264 requirements for a 4.5-kg dog. Unlike in EU countries, American pet food manufacturers are required
265 to report the caloric density of all types of treats in the label,¹⁸ except for rawhides which are not
266 included in this category. Because the use of treats has been considered a risk factor for obesity in
267 dogs,^{8,9} the energy content per piece of treat should be added to packaging labels in order to help
268 veterinarians recommend the proper daily intake for each individual.

269 Treats must be carefully considered in feeding regimens of dogs afflicted with chronic heart failure
270 (CHF) and chronic kidney disease (CKD) due to their potential high mineral content.¹⁶ However,
271 conflicting literature is available on the importance of mineral modifications for some nutrients like
272 sodium and no studies have addressed the impact of treats. The minerals to be kept under closest
273 control in the disorders above are P and Na. Given that the recommended P and Na concentrations
274 for foods used in managing CKD and CHF in dogs range from 0.2 to 0.5 per cent DM and 0.3 per
275 cent DM (estimated 500–1250mg/1000 kcal and 750mg/1000 kcal for a diet containing 4000 kcal/kg
276 DM) or less at late stage of the diseases, respectively,¹⁶ only rawhides respected the P
277 recommendations for P intake, whereas meat strips, chewable sticks and dental sticks failed to fulfil
278 those for Na. An online questionnaire used in a study by Freeman and others¹⁹ to determine the dietary
279 patterns and intake of nutrients of concern in dogs with cardiac disease showed that even when owners
280 fed dogs low Na dog food, they may be adding large amounts of Na to the diet (25 per cent of total
281 daily intake, on average) via treats and foods used to administer pills. Therefore, veterinarians should
282 consider treats in the feeding regimens of CKD and CHF patients and evaluate whether the products
283 owners feed are suitable for their dogs. Rawhides, whose only ingredient is dried bovine skin, were

284 the treats richest in Hyp, and recent findings suggest that the consumption of Hyp-containing protein
285 sources should be discouraged for dogs prone to calcium-oxalate urolith formation.²⁰ A study by
286 Dijcker and others²¹ showed Hyp to be related to the synthesis of endogenous oxalate, a potential
287 substrate in the formation of calcium-oxalate uroliths in dogs and cats. A recent study²⁰ involving
288 cats demonstrated that collagen tissue-rich diets (collagen is the protein source in which Hyp is most
289 concentrated) significantly increased urinary oxalate excretion proportionally to Hyp intake. No
290 recommended levels of dietary Hyp have been proposed thus far, but this study suggests that the use
291 of products such as rawhides should be minimised in calcium-oxalate preventive regimen diets.

292 Starch was predictably the main nutrient in dental sticks and biscuits because both categories had
293 ‘cereals’ as the first ingredient in the list. Interestingly, some products showed very high levels of
294 sucrose (maximum 51.7g/1000 kcal in one product), which was more than 5 per centDM in half of
295 the selected treats. As reported above, dogs like sweet taste,¹⁶ so producers may add sugars as
296 palatability enhancers or as humectants in semimoist products. In dogs fed diets containing high
297 concentrations of lactose and sucrose (>10 per cent and 30 per centDM, respectively), higher water
298 content was observed in small and large intestine chyme and in faeces as well.¹¹ However, as treats
299 generally comprise a small portion of a pet’s diet, even those treats with sugars higher on the
300 ingredient list would be unlikely to cause clinically significant changes in faecal quality.

301 The small number of treats that were analysed per each category is one limitation of this study. This
302 is the first investigation that aimed to categorise dog treats and to determine their nutrient profile, and
303 future studies should sample a greater number of products to provide more precise data. Also, these
304 results may not be representative of all products worldwide given the wide number of dog treats
305 available on the market of many countries to state that. More research is needed to augment the
306 availability of data about complementary pet food.

307 In conclusion, the treat categories considered in this study showed wide variability in chemical
308 composition. Dog treats are usually made of numerous ingredients, which are often undefined
309 because their specific name is replaced by the name of the category to which the feed materials

310 belong; in many cases sugars are mentioned among the ingredients listed. Treat labelling should be
311 more explicit and provide more detailed information on ingredients. Given that treats have been
312 identified as a risk factor in making dogs overweight, product energy values should also be specified
313 in order to help veterinarians prescribe proper amounts for dogs, especially those of small size. Treats
314 contain varying amounts of minerals, and because more specific information is not available on the
315 label, caution should be adopted when feeding treats to individuals requiring mineral restrictions in
316 their diet. Rawhides contain a high concentration of Hyp and for this reason should be avoided in
317 dogs predisposed to calcium-oxalate urolith formation. Further studies are deemed necessary in order
318 to compare treats of similar categories selected from different markets with the results provided here.

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394 TABLE 1: Mean value, sd, and minimum and maximum of metabolisable energy (ME, kcal/100g),
 395 treat weight (g) and treat caloric density (kcal/treat) per category of treats
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	ME* (kcal/100g) from label			Weight/treat (g)			Caloric density (kcal/treat)		
	Mean±sd	Min	Max	Mean±sd	Min	Max	Mean±sd	Min	Max
Biscuits	329.2±13.0	307.4	339.9	6.5±3.5	2.9	11.8	21.7±11.6	10.2	37.6
Tenders	312.2±70.6	252.5	484.3	3.5±3.9	0.7	12.0	10.9±12.6	1.9	37.3
Meat strips	284.4±19.2	262.8	299.1	10.0±0.6	9.2	11.0	28.6±1.5	27.4	30.3
Rawhides	309.5±20.6	296.3	340.1	45.0±16.0	27.4	68.5	132.5±44.4	83.7	161.8
Chewable sticks	294.0±25.9	251.0	337.1	33.3±63.8	5.8	214.2	97.7±191.3	20.9	640.8
Dental sticks	267.7±17.6	246.5	293.9	20.4±9.7	7.5	30.8	55.4±27.1	20.0	82.5

397 *Calculated using the FEDIAF formula: kcal ME/100g =% crude protein x 3.5+% crude fat x 8.5+%
 398 NFE x 3.5. NFE, nitrogen-free extract.

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401 TABLE 2: Mean value and sd of the percentage of the daily metabolisable energy requirement
402 (MER)* of a small-sized/medium-sized/big-sized dog supplied by the different categories of treats,
403 according to the feeding indications reported on the label.

404

	% MER 7-kg dog (409 kcal/day)	% MER 15-kg dog (724 kcal/day)	% MER 30-kg dog (1218 kcal/day)
	Mean±sd	Mean±sd	Mean±sd
Biscuits	16.4±12.6	16.9±12.1	16.8±11.7
Tenders	10.1±3.8	10.0±3.5	9.7±3.2
Meat strips	10.5±4.1	10.0±2.5	9.6±0.6
Rawhides	25.5±7.2	18.3±5.1	10.9±3.0
Chewable sticks	12.2±1.7	12.4±2.9	16.9±14.6
Dental sticks	7.8±3.4	7.9±4.1	4.7±2.5

405 *MERs were calculated according to the following formula: $95 \times \text{kg Body Weight}^{0.75}$.¹¹

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407 **TABLE 3:** ME, DM and nutrient composition (mean±sd, min, max) obtained from the proximate
 408 analysis of treats divided into categories

	Biscuits	Tenders	Meat strips	Rawhides	Chewable sticks	Dental sticks
	Mean±sd (min–max)	Mean±sd (min–max)	Mean±sd (min–max)	Mean±sd (min–max)	Mean±sd (min–max)	Mean±sd (min–max)
ME* (Kcal/100g)	351.3±16.5 (331.6– 372.0)	337.6±65.2 (281.9– 475.0)	302.8±5.8 (298.6– 306.9)	349.0±13.0 (328.4– 363.9)	327.4±22.3 (299.0– 353.5)	302.9±6.6 (297.1– 311.1)
DM (%)	89.9±1.3 (88.6–91.6)	82.5±7.1 (70.6–95.8)	80.2±3.3 (77.9– 82.6)	87.9±1.1 (86.2–89.0)	82.9±3.7 (79.1– 86.8)	84.7±2.3 (81.2–86.0)
CP (g/1000 kcal)	47.0±12.0 (36.7–62.4)	78.0±31.3 (19.2– 118.7)	99.8±14.2 (89.8– 109.9)	171.9±67.5 (78.0– 244.6)	108.9±54.1 (22.0– 204.2)	29.4±15.6 (10.8–48.9)
EE (g/1000 kcal)	27.0±9.5 (15.8–35.9)	29.2±16.0 (5.3–54.9)	24.8±9.8 (17.9– 31.8)	10.7±9.9 (1.2–23.7)	19.7±15.4 (4.8–53.9)	11.5±3.7 (6.5–14.6)
CF (g/1000 kcal)	6.7±3.1 (3.4–10.1)	3.7±2.1 (0.2–6.8)	8.6±3.5 (6.1–11.1)	5.8±5.7 (0.7–13.9)	3.6±1.6 (1.8–6.3)	3.1±2.9 (0.6–7.3)
Ash (g/1000 kcal)	17.1±3.5 (13.7–21.2)	20.7±11.9 (6.2–42.5)	25.0±10.8 (17.4– 32.6)	5.7±4.8 (1.9–13.4)	17.9±10.4 (5.5–34.1)	25.5±4.3 (21.2–31.2)

Calcium (g/1000 kcal)	2.8±1.2 (1.1–3.5)	3.6±3.5 (0.2–11.2)	3.0±0.6 (2.6–3.4)	1.3±1.5 (0.1–3.9)	3.6±3.4 (0.2–9.9)	4.5±1.5 (3.0–6.6)
Phosphorus (g/1000 kcal)	2.3±0.9 (1.1–3–1)	2.9±2.4 (0.3–8.4)	2.9±0.6 (2.5–3.3)	0.2±0.3 (0.0–0.8)	1.9±1.6 (0.1–4.4)	2.2±1.5 (0.6–3.9)
Sodium (g/1000 kcal)	0.7±0.6 (0.3–1.5)	0.5±0.5 (0.0–1.4)	1.9±2.3 (0.3 – 3–6)	0.6±0.4 (0.3–1.2)	1.1±1.4 (0.3–4.5)	0.8±0.8 (0.2–2.0)
Potassium (g/1000 kcal)	2.6±1.8 (0.5–5.0)	2.0±1.3 (0.0–3.6)	2.5±0.8 (2.0–3.0)	3.4±1.6 (1.8–5.7)	4.1±2.6 (1.2–8.2)	3.3±1.3 (1.6–4.9)
Magnesium (g/1000 kcal)	0.4±0.1 (0.2–0.5)	0.3±0.3 (0.1–0.9)	0.4±0.0 (0.4–0.4)	0.2±0.2 (0.0–0.5)	0.2±0.0 (0.1–0.2)	0.3±0.1 (0.1–0.4)
Hyp (g/1000 kcal)	0.5±0.2 (0.3–0.8)	1.5±1.2 (0.0–3.3)	2.3±2.5 (0.5–4.0)	29.5±5.2 (20.4–33.0)	6.9±9.9 (0.1–26.3)	0.8±0.4 (0.4–1.1)
Starch (g/1000 kcal)	107.9±32.5 (63.6–135.6)	56.1±39.8 (0.0–137.4)	38.8±24.0 (21.8– 55.8)	0.0±0.0 (0.0–0.0)	55.0±61.7 (0.0– 186.3)	155.6±33.9 (112.1– 194.7)
Glucose (g/1000 kcal)	0.8±1.0 (0.2–2.3)	2.3±2.8 (0.0–8.1)	0.7±0.5 (0.3–1.0)	0.0±0.0 (0.0–0.0)	0.3±0.4 (0.0–1.0)	0.1±0.2 (0.0–0.4)

Fructose (g/1000 kcal)	0.6±0.4 (0.2–1.0)	1.2±2.1 (0.1–6.6)	0.6±0.2 (0.4–0.7)	0.0±0.0 (0.0–0.0)	0.5±0.6 (0.0–1.7)	0.2±0.2 (0.0–0.5)
Sucrose (g/1000 kcal)	13.3±15.3 (2.9–35.9)	11.8±17.4 (0.4–51.7)	13.9±16.1 (2.5–25.3)	0.0±0.0 (0.0–0.0)	3.0±4.9 (0.0–14.5)	0.9±1.1 (0.0–2.0)

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410 Percentage energy digestibility=91.2–(1.43xpercentage crude fibre in dry matter).

411 *Calculated using the National Research Council formula:

412 GE, (5.7xg protein)+(9.4xg fat)+[4.1x(g NFE+g fibre)].

413 DE, (GExpercentage energy digestibility/100).

414 ME, DE –(1.04xg protein).

415 CF, crude fibre; CP, crude protein; DE, digestible energy; DM, dry matter; EE, ether extract; GE,

416 gross energy; Hyp, hydroxyproline; ME, metabolisable energy; NFE, nitrogen-free extract.

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