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ORIGINAL ARTICLE

Insoluble fibres, satiety and food intake in cats fed kibble diets*

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Summary

Fibre is generally considered to dilute food energy, alter intestinal transit time and promote satiety; however, in cats, conflicting results have been found. In this study, two insoluble fibres were evaluated in four feline diets: control (no added fibre); diet with 10% sugar cane fibre; diet with 20% sugar cane fibre; and diet with 10% cellulose. The experiment was conducted with 32 cats, eight animals per diet, over 42 days: 1–7 for diet adaptation; 8–14 for total collection of faeces for digestibility; 15–17 for fresh faeces collection for fermentation products measurements; 18–20 for gastrointestinal transit time determination; 21 and 37 to evaluate the pattern of food intake; and 22 and 42 to assess satiety. Means were compared by analysis of variance and orthogonal contrasts, and the pattern of food intake was compared by repeated-measures analysis of variance ($p < 0.05$). The cats exhibited increased food intake after fibre addition to the diets ($p < 0.05$), achieving similar energy consumption. Cellulose and the two levels of sugar cane fibre reduced nutrient availability and energy digestibility, but only sugar cane fibre reduced fat digestibility ($p < 0.05$). Faecal output and the number of defecations per day increased with fibre inclusion ($p < 0.05$). Gastrointestinal transit time did not change with sugar cane fibre inclusion, but it was reduced with cellulose addition ($p = 0.032$). The pattern of food intake did not change, but cats fed fibre-supplemented diets exhibited greater consumption of a challenge meal, increasing energy intake ($p < 0.01$) when exposed to a palatable, energy-dense food.

Keywords digestibility, gastrointestinal transit time, energy intake, feline, propionate, satiety

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Introduction

Cats, despite being carnivorous, require some amount of indigestible material in their food for proper regulation of intestinal peristalsis and gut physiology (Kienzle et al., 1991). Thus, many diet formulations including fibre sources have been developed by the pet food industry. Insoluble fibres are also used to reduce energy content by reducing nutrients digestibility (Earle et al., 1998; Fekete et al., 2004), promote faecal formation and quality (Wichert et al., 2002; Prola et al., 2010) and prevent hairball formation (Beynen et al., 2011; Loureiro et al., 2014). Of these, the use of fibre as a tool to dilute food energy and promote satiety to support feline weight maintenance or weight loss is of particular interest in recent years (Fekete et al., 2001; Vasconcellos et al., 2009; Cline et al., 2012). These actions are most likely performed by insoluble and non-fermentable fibres, such as sugar cane fibres. Sugar cane fibre is a source of insoluble fibre and has been used successfully in feline diets (Fischer et al., 2012), because their relatively low fermentability insoluble fibres allow for higher inclusion with less negative effects like gas formation and soft stools which are associated with soluble and fermentable fibres (Fischer et al., 2012).

Among the proposed mechanisms of action, insoluble fibre is believed to reduce gastric emptying while increasing intestinal filling, thereby possibly stimulating the release of hormones that reduce hunger

(Butterwick and Hawthorne, 1998; Weber et al., 2007). However, these effects are not yet completely established and the scientific information about the efficacy of insoluble fibres on appetite regulation and other aspects of feline gut physiology is insufficient and controversial (Chandler et al., 1999; Bissot et al., 2010; Cline et al., 2012). It is also important to perform a balanced evaluation of the potential benefits of fibre alongside its undesirable effects such as constipation, diarrhoea, excessive faeces production and reduced nutrient digestibility (Fekete et al., 2001; Fischer et al., 2012).

To this end, this study was designed to investigate the effects of two sources of insoluble fibre, purified cellulose and sugar cane fibre, added to an extruded feline diet. The measured outcomes included nutrient and energy digestibility, faecal fermentation products, stool characteristics, gastrointestinal transit time, pattern of food intake and animal satiety.

Materials and methods

Animals and experimental design

All experimental procedures were approved by the Ethics and Animal Welfare Committee of the College of Agrarian and Veterinarian Sciences, São Paulo State University, Jaboticabal, Brazil (Protocol 20.481/10).

Thirty-two mixed-breed cats, 15 males and 17 females, aged 6.0 ± 1.2 years on average were used for the study. The cats had an average body weight of 4.2 ± 0.1 kg and mean body condition score of 5.6 ± 0.7 (Laflamme, 1997). All the cats were neutered, and the health status of the animals was confirmed prior to beginning the study. The animals were kept under the same environmental conditions and belonged to the Laboratory of Research on Nutrition and Nutritional Diseases of Dogs and Cats, College of Agrarian and Veterinarian Sciences, São Paulo State University.

The study followed a completely randomized design with four diets and eight cats per diet. The experiment lasted 42 days, distributed as follows: adaptation to the diets occurred from days 1 to 7; total faeces were collected from days 8 to 14 to calculate apparent nutrients digestibility; fresh faeces collection (for the measurement of fermentation products) was performed from days 15 to 17; gastrointestinal transit time was determined from days 18 to 20; food intake patterns were measured on days 21 and 37; and satiety was assessed on days 22 and 42.

From 1600 h to 0800 h (16 consecutive hours), the cats were restricted to individual stainless steel metabolic cages (0.80 m × 0.80 m × 1.0 m), when their experimental foods and water were available. From 0800 h to 1600 h (eight consecutive hours), the cats were kept in a collective cattery (50 m^2) for exercise and socialization, where they had access to water but not to food. During the digestibility phase, fresh faeces collection and food retention time periods, the cats were fully restricted to their metabolism cages, but the food supply schedule was not changed. Throughout the study, a 12-h dark: 12-h light cycle was maintained.

The daily amount of food was initially defined according to the energy maintenance requirements of cats, calculated as 418 kJ of metabolizable energy (ME) per $kg^{0.67}$ where the ME of the diets was estimated from their chemical composition according to NRC (2006). The amount of food presented at feeding as well as the amount of food remaining after feeding was recorded daily to calculate food intake. The cats were weighted weekly and if a gain in body weight was observed the amount of food provided would be reduced to control obesity development. However, if food was available and a cat lost body weight, there was no attempt to increase food intake during the experiment, so cats were not allowed to gain weight, but they could lose weight, as a result of the diet characteristics. In a third situation, if a cat ate everything and lost weight, more food would be offered (but this never happened during the study). Water was always available *ad libitum*.

Diets

Four diets were formulated according to the nutritional recommendations of the European Pet Food Industry (FEDIAF, 2008) for adult cats: a control diet (CO) with no additional fibre; two diets including 10% or 20% by weight sugar cane fibre (SF10 and SF20); and a diet including 10% by weight cellulose (CEL10). The fibres in the experimental diets were used in substitution of maize in the CO diet, with small adjustments to poultry fat and maize gluten meal in order to normalize the diets for fat and crude protein contents (Table 1). Sugar cane fibre is produced after the washing and centrifugation of the sugar cane bagasse, which is further dried and micro grinded. Its composition includes 1.7% crude protein, 1.3% fat, and 90.8% insoluble dietary fibre. The cellulose sample used contains 91.4% of insoluble dietary fibre (Calabro et al., 2013).

All diets were mixed and ground in a hammer mill (Model 4, D'Andrea, Limeira, Brazil) fitted with a 0.8-mm screen before being extruded and kibbled under identical processing conditions in a single-screw

Table 1 Ingredient and chemical compositions, processing parameters and kibble quality of the experimental diets for cats with different insoluble fiber sources (as-fed basis)

	Experimental diets*			
Item	CO	SF10	SF20	CEL10
Ingredient composition (%)				
Sugar cane fiber†	0.0	10.0	20.0	0.0
Cellulose‡	0.0	0.0	0.0	10.0
Broken rice	15.0	15.0	15.0	15.0
Maize	33.7	22.3	11.1	21.7
Poultry by product meal	28.0	28.0	28.0	28.0
Corn gluten meal	11.7	12.7	13.6	13.3
Poultry fat	6.6	6.9	7.2	7.0
Liquid Palatant§	2.5	2.5	2.5	2.5
Minor ingredients¶	2.5	2.5	2.5	2.5
Total	100.0	100.0	100.0	100.0
Chemical composition** (%)				
Dry matter	94.5	94.6	95.3	94.8
Crude protein	32.2	31.9	30.6	32.8
Acid-hydrolyzed fat	11.8	11.8	11.9	12.2
Starch	37.7	29.6	23.5	30.3
Total dietary fiber	10.4	20.8	27.1	19.9
Ash	8.5	8.3	8.3	7.2
Calcium	1.4	1.3	1.3	1.3
Phosphorus	1.0	1.1	1.0	0.9
Gross energy (kJ/g)	20.1	20.2	20.3	20.2
Processing parameters and kibble quality				
Preconditioner temperature (°C)	94	96	97	92
Extruder temperature (°C)	131	131	126	131
Food apparent density (g/l)	350	353	373	363
Starch gelatinization degree (%)	76.7	73.5	91.7	78.1
Cutting force (kgf)††	1.74	2.57	3.06	3.09

*C, control without fiber supplementation; SF10, with 10% sugar cane fiber addition; SF20, with 20% sugar cane fiber addition; CEL10, with 10% cellulose addition.

†Vit2be Fiber, Dilumix, Leme, SP, Brasil. Mean geometric diameter = 188 ± 2 μm.

‡Cellulose, Minérios Ouro Branco Ltda, São Paulo, SP, Brasil. Mean geometric diameter = 112 ± 2 μm.

§Liquid palatant, SPF do Brazil, Descalvado, Brazil.

¶Supplied per kilogram of diet: vitamin A, 18 000 IU; vitamin D, 1200 IU; vitamin E, 200 IU; thiamin, 6 mg; riboflavin, 10 mg; pantothenic acid, 40 mg; niacin, 60 mg; pyridoxine, 6 mg; folic acid, 0.30 mg; vitamin B12, 0.1 mg; iron, 100 mg; copper, 10 mg; magnesium, 10 mg; zinc, 150 mg; iodine, 2 mg; selenium, 0.3 mg; taurine, 1.2 g; choline chloride, 2.2 g; lysine, 1.1 g; potassium chloride, 5 g; sodium chloride, 5 g; calcium carbonate, 1.1 g; mold inhibitor (MoldZap: ammonium dipropionate, acetic acid, sorbic acid and benzoic acid – Alltech do Brasil Agroindustrial Ltda, Curitiba, Brazil), 1 g; antioxidant (Banox: butylated hydroxyanisole, butylated hydroxytoluene, propyl gallate and calcium carbonate – Alltech do Brasil Agroindustrial Ltda, Curitiba, Brazil), 0.4 g.

**Analyzed in duplicate.

††n = 20 kibbles.

extruder (Mab 400S, Extrucenter, Monte Alto, Brazil). The manufacturing process was controlled by adjusting kibble density between 350 and 370 g/l (as-is basis) every 20 min to ensure consistent cooking and kibble quality (size and expansion). Extruder preconditioning temperature was kept close to 95 °C. Water, steam, screw speed and ration flux were adjusted according to diet formulation, and the temperature of the dough under extrusion varied between 126 °C and 131 °C. After extrusion, the kibbles were dried in a forced-air dryer at 105 °C and coated with fats and palatability enhancers.

The kibbles were submitted to a cutting test performed with a texturometer (TA-XT2 SMS, Stable Micro Systems, Godalming, UK) set to operation mode strength/compression, return to start option enabled, pretest speed of 2 mm/s, speed during the test of 0.5 mm/s and speed before test of 10 mm/s. The test was conducted on 20 units for each sample, using a probe with a blade set with a Warner–Bratzler knife (heavy-duty platform/blade set) with a cutting distance of 10 mm. The data were analysed with the software Texture Expert (Stable Micro Systems, Godalming, UK). Kibble apparent density was determined by weight a recipient with 1 l volume capacity filled with the kibbles in study. The processing parameters and kibble quality of the diets are presented in Table 1.

Total tract apparent digestibility and metabolizable energy content of experimental diets

Food consumption was recorded daily. On the first day of faeces collection (day 8), all faeces were removed from the cages and discarded before 0730 h and total faecal output for each cat was collected from this point on for the next 7 days. Faeces were collected twice per day and pooled per cat. Faecal samples were weighed and frozen (−15 °C). After, faeces were dried in a forced-air oven (Fanem, São Paulo, Brasil) at 55 °C for 72 h. Dried faecal samples and diets were ground in a cutting mill (Mod MA-350, Marconi, Piracicaba, Brazil) fitted with a 1 mm screen and analysed for DM by oven-drying the sample (method 934.01) and conversion to ash by muffle furnace incineration (method 942.05). Crude protein (CP) was analysed by the Kjeldahl method (method 954.01), and acid hydrolysed fat was assessed using a Soxhlet apparatus (method 954.02) according to the Association of Official Analytical Chemists (AOAC, 1995). Organic matter (OM) was calculated as DM minus ash. Minerals were analysed after nitro perchloric digestion; phosphorus was measured by visible spectrophotometery (Labquest Bio 2000, Labtest Diagnóstica S.A., Lagoa Santa, Brazil) and calcium by flame atomic absorption spectrophotometry (GBC-932 AA, Scientific Equipment PTY, Melbourne,

Australia). Dietary fibre was measured using a combination of enzymatic and gravimetric procedures (AOAC, method 985.29, 1995). Total amount of starch was determined according to the method described by Hendrix (1995). Gross energy (GE) was determined in a bomb calorimeter (model 1261 Parr Instrument Company, Moline, IL, USA). The degree of gelatinization of the starch of the experimental diets was determined by the amyloglucosidase method (Sá et al., 2013). All analyses were carried out in duplicate and repeated when the coefficient of variation was higher than 5%.

The coefficients of total tract apparent digestibility (CTTAD) of nutrients were calculated according to the quantitative collection of faeces protocol. ME was calculated by subtracting 3.77 kJ/g digestible crude protein from the digestible energy content (NRC, 2006).

Stool characteristics and fermentation products

Faecal samples were scored according to the following system: 1 = watery – liquid that can be poured; 2 = soft, unformed – stool assumes shape of container; 3 = soft, formed, moist – softer stool that retains shape; 4 = hard, formed, dry stool – remains firm and soft; 5 = hard, dry pellets – small, hard mass.

Fresh faecal samples were collected immediately after elimination on days 15 to 17 to measure short-chain fatty acids (SCFA) and ammonia. Approximately 10 g of faeces was mixed with 30 ml of a 16% (v/v) formic acid solution, precipitated at 4 °C for 72 h, and the supernatant was centrifuged (5804R, Eppendorf, Hamburgo, Brazil) three times at 4500 G at 15 °C for 15 min. The SCFA were analysed in the supernatant by gas chromatography (model 9001, Finnigan, San Jose, USA) according to Erwin et al. (1961) using a glass column 2 m in length and 3.17 mm in width, covered with 80/120 Carbopack B-DA/4% Carbowax 20M. Nitrogen was the carrier gas with a flow rate of 25 ml/min. Working temperatures were 220 °C at injection, 210 °C in the column and 250 °C in the flame ionization detector. The ammonia concentration was determined according to Vieira (1980) in the same extracts used for the determination of SCFA. Faecal pH was determined by mixing 10 ml of distilled water with 5 g of fresh faeces (v/w) and measuring the result with a pH meter (DM20, Digicrom Analítica Ltda, São Paulo, Brazil).

Gastrointestinal transit time

Digesta retention time in the gastrointestinal tract was evaluated according Burrows et al. (1982). On days 18, 19 and 20, the cats were fed at 1600 h, and immediately following the first act of food intake, the cats were orally dosed with gelatin capsules containing 12 radiopaque markers (Sitzmarks, Konsyl Pharmaceuticals Inc., Fort Worth, Texas USA). Each marker was 4.5 mm in diameter and densities of 1.25 g/ml. On each day, a different marker format was utilized, allowing three consecutive observations of the retention time. The time of the marker administration was registered, and the cages were observed at 2-h intervals until the last marker was recovered in the faeces. All faeces were collected, weighed and the time of sampling recorded. When the exact faecal elimination time was not observed, the mean time between samplings was considered for diet comparison. All stools were radiographed, and the time excretion of the last marker was recorded.

The gastrointestinal transit time was computed as the time interval (in hours) between the food intake and capsule administration and the time of the stools containing the last marker recovered. The mean food retention time was the average for the 3 days of observation. In addition, the marker recovery rate was computed as: follows

$$\text{Marker recovery rate (\%)} = \frac{\text{Number of markers recovered on stools}}{\text{Total of markers orally dosed}} \times 100$$

To validate the observation, a minimum recovery rate of 90% of the markers was established. The number of defecations per day and the weight of each defecation were also recorded.

Pattern of food intake and satiety challenge

The kinetics of food intake was evaluated on days 21 and 37, by recording food intake at each 2-h interval. The food not consumed after the 2-h feeding window was subtracted from the total weight of food offered to calculate the consumption.

The satiety of the cats was indirectly evaluated by measuring the consumption of a challenge meal. On days 22 and 42, the cats were fed their experimental diets at 1600 h which they had access to until 0800 h. Additionally, they were exposed during two periods of 1 h each, from 2100 h to 2200 h and from 0700 h to 0800 h, to an *ad libitum* amount of a palatable commercial diet for cat maintenance (Guabi Natural Gatos Adultos, Mogiana Alimentos, Campinas, Brazil. Composition: Crude protein, 34%; Fat, 16%; Moisture, 10%; Crude fibre, 3%; Ash, 8%; Metabolizable energy, 17.6 kJ/g). The weights of the experimental

and the challenge foods were recorded before they were given to the cats, and any remaining food was weighed and subtracted from the initial weight to derive the intake amount. The objective was to observe how the consumption of the fibre-supplemented diets would affect the intake of a second food, in particular whether they would limit or stimulate ingestion behaviour. The intakes of the experimental and challenge foods were calculated on both a DM basis and on an energy basis taking into account the diets' ME.

Statistical analysis

Data were analysed using a completely randomized design and submitted to analysis of variance followed by F-tests utilizing the general linear model (GLM) procedure of the Statistical Analysis System (SAS) software (Version 9.1, SAS Institute Inc., Cary, NC, USA). Model sums of squares were separated into treatment (food) effects. When treatment differences for digestibility, stool characteristics and gastrointestinal transit time were detected, sum of squares was partitioned in a set of three orthogonal contrast: two polynomial contrast, to evaluate whether the effect of inclusion levels of sugar cane fibre was constant (lineal) or no (quadratic), and CEL10 vs. CO + SF10 + SF20 to contrast the diet with 10% cellulose with the mean of the diets without cellulose. Repeated-measures analysis of variance was used to evaluate the effects of diet and time on the pattern of food intake and animal satiety. The mixed models procedure of the SAS software was used, following the model:

$$y_{\mathrm{ijkl}} = \mu + \alpha_i + v_j(\alpha_i) + \beta_k + (\alpha\beta)_{\mathrm{ik}} + v_j(\alpha\beta_{\mathrm{ik}}) + \delta_l + (\alpha\delta)_{\mathrm{il}} + (\beta\delta)_{\mathrm{kl}} + (\alpha\beta\delta)_{\mathrm{ikl}} + \varepsilon_{\mathrm{ijkl}}$$

where y_{ijkl} = value of the observation of treatment i, repetition j, day k and hour l; μ = general mean effect; α_i = effect of treatment i; $v_j(\alpha_i)$ = effect of repetition j of the treatment i; β_k = effect of day k; $(\alpha\beta_{\mathrm{ik}})$ = effect of the interaction of the treatment i and day k; $v_j(\alpha\beta_{\mathrm{ik}})$ = effect of the repetition j inside the interaction of the treatment i and day k; δ_l = effect of hour l; $(\alpha\delta)_{il}$ = effect of the interaction of treatment i and hour l; $(\beta\delta)_{kl}$ = effect of the interaction of the day k and hour l; $(\alpha\beta\delta)_{kl}$ = effect of the interaction of the treatment i, day k and hour l; $\varepsilon_{\mathrm{ijkl}}$ = random error of the treatment i, repetition j, day k and hour l. The α, β, $(\alpha\beta)$, δ, $(\alpha\delta)$, $(\beta\delta)$ and $(\alpha\beta\delta)$ were factors of fixed effects. Pair-wise means comparisons were made using Tukey's test when the F-test was significant. To compare the body weight of animals before and after the experimental period, a paired T analysis was conducted. All data were found to comply with the assumptions of the analysis of variance procedures. The values of $p < 0.05$ were considered significant.

Results

Diets were similar in crude protein and fat contents, but their total dietary fibre content increased according to each formulation. Starch gelatinization was similar among the CO, SF10 and CEL10 diets, but higher in the SF20 diet. For both fibre types, fibre inclusion increased the hardness of the kibbles as measured by higher cutting force (Table 1).

All foods were well accepted and consumed by the cats, and there were no episodes of diarrhoea, vomiting or soft stools as presented in Table 2. The body weight of the animals remained constant during the 42 days of the experiment ($p = 0.622$; Table 2). Food leftovers were found in all cats, with a mean of 6.3 ± 0.5 g/cat/day (results similar between foods; data not shown), and due this food intake should be considered *ad libitum*. A linear increase in food DM intake was observed after sugar cane fibre inclusion ($p = 0.015$). For the cellulose-supplemented diet, the orthogonal contrast did not detect difference in comparison with the average of the other diets. The intake of ME, on the other hand, was similar among the diets with a mean intake of 339.9 kJ/kg$^{0.67}$ regardless of fibre inclusion (Table 2).

During the digestibility study DM, fat and total dietary fibre intake increased with sugar cane fibre and cellulose addition in comparison with the control diet ($p < 0.05$; Table 3). This was expected for fibre and is explained for the other nutrients due the higher food intake to achieve similar available energy consumption. Nutrient digestibility (except starch), gross energy digestibility and ME content decreased linearly with sugar cane fibre additions ($p < 0.001$). Cellulose supplementation resulted in reductions in nutrient digestibility comparable with the promoted by sugar cane fibre inclusion (the orthogonal contrast did not detect difference in comparison with the average of control, SF10 and SF20 diets). Fat digestibility was an exception, as it was not reduced by cellulose but was by sugar cane fibre ($p = 0.029$; greater value for cellulose than the average of control, SF10 and SF20 diets). The total dietary fibre digestibility also differed between the cellulose and sugar cane fibre diets ($p < 0.001$); it was lower for the cellulose diet.

Table 2 Initial and final body weight, and dry matter and metabolizable energy intake of cats fed with experimental kibble diets with different insoluble fiber sources

	Experimental diets*					Contrasts‡		
Item	CO	SF10	SF20	CEL10	SEM†	Linear	Quadratic	CO + SF10 + SF20 vs. CEL10
Body weight (kg)								
Initial	4.4	4.0	4.4	4.1	0.12	ns§	ns	ns
Final	4.4	4.0	4.3	4.1	0.11	ns	ns	ns
Food intake (Mean of 42 days of study)								
Dry matter (g/kg $BW^{0.67}$/day)	20.2	21.2	26.0	24.5	1.4	0.015	ns	ns
Metabolizable energy (kJ/kg $BW^{0.67}$/day)¶	335.1	316.7	335.6	372.3	21.7	ns	ns	ns

*CO, control without fiber supplementation; SF10, with 10% sugar cane fiber addition; SF20, with 20% sugar cane fiber addition; CEL10, with 10% cellulose addition.
†SEM = standard error of the mean (n = 8 cats per treatment).
‡Linear and quadratic effect of sugar cane fiber addition.
§Not significant ($p \geq 0.05$).
¶Calculated with the food metabolizable energy values determined *in vivo*.

Table 3 Nutrient intake (g/cat/day), coefficient of total tract apparent digestibility (CTTAD) and metabolizable energy content (ME, kJ/g as-fed basis) of experimental diets for cats with different insoluble fiber sources

	Experimental diets*					Contrasts‡		
Item	CO	SF10	SF20	CEL10	SEM†	Linear	Quadratic	CO + SF10 + SF20 vs. CEL10
Nutrient intake during the digestibility study								
Dry matter	56.2	58.7	72.6	65.1	2.3	0.013	ns§	ns
Crude protein	18.1	18.8	22.2	21.0	0.72	ns	ns	ns
Acid-hydrolyzed fat	6.7	7.0	8.7	8.0	0.28	0.009	ns	ns
Total Dietary Fiber	5.9	13.0	21.2	13.7	0.98	<0.001	ns	ns
Starch	21.2	17.4	17.0	19.7	0.71	ns	ns	s
CTTAD (%)								
Dry matter	82.8	73.1	62.2	73.6	1.41	<0.001	ns	ns
Organic matter	86.3	76.2	64.9	76.2	1.44	<0.001	ns	ns
Crude protein	84.0	81.9	79.0	83.1	0.64	0.005	ns	ns
Acid-hydrolyzed fat	86.8	83.4	78.0	85.9	0.83	<0.001	ns	0.029
Total Dietary Fiber	52.9	44.4	28.3	33.7	2.22	<0.001	ns	<0.001
Starch	99.5	99.2	99.6	99.1	0.06	ns	ns	ns
Gross energy	84.9	75.8	65.7	77.4	1.30	<0.001	ns	ns
Food ME	16.66	14.95	12.94	15.24	0.06	<0.001	ns	ns

*CO, control without fiber supplementation; SF10, with 10% sugar cane fiber addition; SF20, with 20% sugar cane fiber addition; CEL10, with 10% cellulose addition.
†SEM = standard error of the mean (n = 8 cats per treatment).
‡Linear and quadratic effect of sugar cane fiber addition.
§Not significant ($p \geq 0.05$).

Data describing fermentation products in cat faeces are presented in Table 4. A linear increase in propionate and a linear decrease in butyrate were observed for sugar cane fibre diets ($p < 0.01$). Propionate concentration was lower in the faeces of cats fed the cellulose diet in comparison with the other diets ($p = 0.005$). No differences in ammonia or faecal pH were found. All fibre additions induced higher faecal production ($p < 0.001$); however, they did not alter faecal DM or score (Table 5). Cats consuming the fibre diets displayed more defecations per day, and the mass per defecation was also higher ($p < 0.03$; Table 5). Sugar cane fibre addition did not change the gastrointestinal transit time, but cellulose reduced it by approximately 6 h ($p = 0.032$).

The pattern of food intake was not altered by fibre inclusion. After repeated-measures analysis, no effects of the day of observation or diet x day interactions

Table 4 Schort-chain fatty acids (mMol/g of fecal DM) and ammonia concentrations, and fecal pH of cats fed experimental diets with different insoluble fiber sources

	Experimental diets*					Contrasts‡		
Item	CO	SF10	SF20	CEL10	SEM†	Linear	Quadratic	CO + SF10 + SF20 vs. CEL10
Acetic	495	448	421	406	16.1	ns§	ns	ns
Propionic	103	124	146	89	5.9	0.005	ns	0.005
Butyric	114	95	60	76	6.5	0.002	ns	ns
Isobutyric	5.5	3.8	4.4	4.1	0.36	ns	ns	ns
Isovaleric	13.0	9.0	10.1	9.7	0.73	ns	ns	ns
Valeric	37.6	34.6	27.3	29.2	1.66	ns	ns	ns
Ammonia (mmol/kg of fecal DM)	124	123	123	145	8.9	ns	ns	ns
Fecal pH	6.11	5.96	6.03	6.04	0.21	ns	ns	ns

*CO, control without fiber supplementation; SF10, with 10% sugar cane fiber addition; SF20, with 20% sugar cane fiber addition; CEL10, with 10% cellulose addition.
†SEM = standard error of the mean (n = 8 cats per treatment).
‡Linear and quadratic effect of sugar cane fiber addition.
§Not significant ($p \geq 0.05$).

Table 5 Fecal production and traits, and gastrointestinal transit time of cats fed experimental diets with different insoluble fiber sources

	Experimental diets*					Contrasts‡		
Item	CO	FC10	FC20	CEL10	SEM†	Linear	Quadratic	CO + SF10 + SF20 vs. CEL10
Fecal traits								
Dry matter (%)	31.9	37.4	35.6	39.1	1.03	ns§	ns	ns
g feces/cat/day (As fed)	33.0	40.8	76.3	45.3	30.7	<0.001	ns	ns
g feces/cat/day (DM basis)	9.4	14.9	26.2	17.3	9.2	<0.001	ns	ns
Fecal score¶	3.3	3.7	3.8	3.8	0.08	ns	ns	ns
Weight of each defecation (g/defecation)	27.5	30.0	44.8	40.4	2.23	0.005	ns	ns
Number of defecations per day	1.04	1.67	1.92	1.38	0.32	0.029	ns	ns
Gastrointestinal transit time (h)	20.6	22.6	22.1	15.2	1.21	ns	ns	0.032

*CO, control without fiber supplementation; SF10, with 10% sugar cane fiber addition; SF20, with 20% sugar cane fiber addition; CEL10, with 10% cellulose addition.
†SEM = standard error of the mean (n = 8 cats per treatment).
‡Linear and quadratic effect of sugar cane fiber addition.
§Not significant ($p \geq 0.05$).
¶Data transformed ($\sqrt{x}$) for statistical analysis.

were found; therefore, the average of days 21 and 37 are presented. For all diets, cats showed greater intake during the first two hours of exposure to their food (from 1600 to 1800 h; $p < 0.001$), and during the following 2-h intervals, they consumed approximately 2 g of DM/kg$^{0.67}$, without differences among the diets (Fig. 1). During the satiety evaluation, the cats displayed equivalent intake of the challenge food during both intervals. No effects of day or day × time interactions were found, so the data from days 22 and 42 were combined in Table 6. The consumption of the experimental diets was similar for all treatments, and the intake was 10–20% lower than the mean intake throughout the 42 days of experiment (Table 2). Cats fed the sugar cane fibre diets showed a linear increase in the challenge food consumption ($p = 0.014$), an effect not seen with the cats fed the cellulose diet ($p = 0.007$). As a result, the total DM and ME intake was greater by cats fed the fibre diets than by cats fed the control diet; the effect was verified for both sugar cane and cellulose diets ($p < 0.004$).

Discussion

Food intake

In the present study, cats fed dry kibble diets differing in ME and fibre contents were able to adjust DM intake to achieve similar energy consumption and

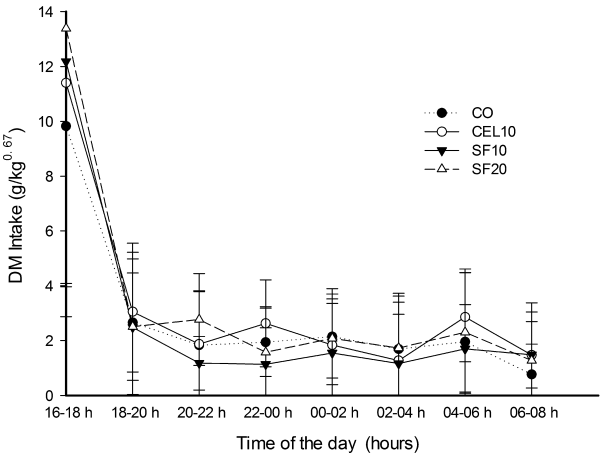

Fig. 1 Pattern of food intake of experimental diets for cats with different insoluble fiber sources. Mean intake of each 2-h interval recorded. CO, control without fiber supplementation; SF10, with 10% sugar cane fiber addition; SF20, with 20% sugar cane fiber addition; CEL10, with 10% cellulose addition. Mean of the observations of days 21 and 37 (n = 8 cats per diet).

maintain constant body weight. This was verified even considering the high levels of fibre included on the present experiment. No food adjustment was necessary to prevent weight loss or gain, showing that within the evaluated range of ME cats are able to balance energy intake over a period of 42 days. The mean ME consumed by the cats was approximately 80% of the recommended for cat maintenance by the NRC (2006). During the study, cats were restrained to their individual metabolic cages during 2/3 of the day, and this difference in energy consumption could be consequent to the low physical activity of the animals.

Some studies, also using diets with high-fibre contents for cats, have shown dietary energy dilution results in reduced energy intake and weight loss. Hirsch et al. (1978), using a mash diet diluted with kaolin, found reductions in energy intake and body weight; however, the diet was 40% kaolin by weight and diet acceptance by the cats is a potential concern, possibly interfering with the results. A more recent study (Prola et al., 2006) showed a reduction in energy intake by cats fed a diet with 24% cellulose. However, the cellulose was diluted in water and then added to a canned diet, resulting in a very watery final food. In this case, we estimate that the cats would have needed to eat high volumes of the food to consume enough energy (approximately 380 ml), in a relatively short time (6 h). Considering the cats in that study had a mean body weight of 3.6 kg, and that cats have a mean stomach volume of 80 ml/kg (Seim III and Bartges, 2003), the water dilution of the diet may have contributed to the limited energy intake. In addition, the period of diet adaptation was short, only 4 days, and may not have been long enough to allow anatomical adaptation, interfering with data interpretation. In another study, Kanarek (1975) also found that cats limited energy intake after 20% cellulose addition to diets, but the author did not provide complete information about the diet's composition and used only two cats per diet.

In the present study, the cats needed to eat only 176 ml of the SF20 food (food intake in g divided by food density in g per 1000 ml) to meet their daily energy requirement; therefore, food volume was unlikely to limit energy intake. In addition, the cats were exposed to the diets for 16 h each day and the study period was longer (42 days) allowing for a more complete anatomical adaptation to the intake of higher volumes of food. Our findings are also consistent with those of Thorne (1982) and Castonguay (1981), who

Table 6 Intake of the experimental diets with different insoluble fiber sources and the challenge food during the satiety evaluation periods of cats. Results are the mean of the challenges at days 22 and 42 of the experiment

	Experimental diets*					Contrasts‡		
Item	CO	FC10	FC20	CEL10	SEM†	Linear	Quadratic	CO+SF10 + SF20 vs. CEL10
Food Intake (g DM/kg$^{0.67}$)								
Experimental diet	18.2	17.0	21.7	20.8	1.60	ns§	ns	ns
Challenge food¶	9.7	18.1	18.4	9.4	1.42	0.014	ns	0.007
Experimental diet + Challenge food	27.9	35.1	40.6	30.3	1.93	0.016	ns	ns
ME consumed (experimental diet + challenge food, kJ ME/kg$^{0.67}$)	420.4	560.2	599.5	475.6	6.95	0.004	ns	ns

*CO, control without fiber supplementation; SF10, with 10% sugar cane fiber addition; SF20, with 20% sugar cane fiber addition; CEL10, with 10% cellulose addition.

†SEM = standard error of the mean (n = 8 cats per treatment).

‡Linear and quadratic effect of sugar cane fiber addition.

§Not significant ($p \geq 0.05$).

¶Sum of the challenges at 2100 to 2200 h and 0700 to 0800 h. Guabi Natural Gatos Adultos, Mogiana Alimentos, Campinas, Brazil.

also found that cats are able to adjust food and energy intake to a range of food processing types (dry, semi-moist and moist foods) and to varying energy densities from 3.3 to 14.6 kJ/g. Even though the present study differs due to the addition of fibre, the anatomical adaptation to higher volumes of intake is consistent among results.

Fibre addition did not alter food intake pattern in the present study. Cats did not have access to food from 0800 h to 1600 h, while they were group housed for socialization. This probably induced anticipatory food behaviour, explaining why the cats ate approximately 47% of the total daily consumption in the first two hours after they were exposed to the meal. During the remaining period of food exposure, the cats displayed relatively constant food consumption, amounting to approximately 7.5% of total intake every 2 h.

Fibre addition also did not result in reduced intake of the challenge meal, not supporting the hypothesis that insoluble fibre induces satiety in cats. There is a lack of scientific studies evaluating satiety in cats and is possible that some limitations could be attributed to the method used here. One finding of note, however, is the higher total DM and energy intake (experimental plus challenge foods) by the cats fed the fibre-supplemented diets, contrary to what was expected. On the days of the food challenge, intake of experimental diets was approximately 90% of that recorded when offered as the only food and then also ate the challenge food, resulting in up to 43% higher energy intake. It is possible the cats had adapted to eating energy-diluted foods by increasing their stomach volume or reducing the feedback of gastric filling on food intake. This may have resulted in a reduced stimulus to interrupt food intake, with higher energy and DM consumption when the animals were exposed to a palatable, energy-dense food. This same behaviour was previously observed by Prola et al. (2006) after switching cats from a fibre-supplemented diet to a more energy-dense food.

These findings, if confirmed by further studies under different conditions and dietary situations, might have important practical implications for the dietary management of obese cats. After being fed energy-diluted, high-fibre diets, cats may be more prone to overconsumption of palatable, energy-dense foods, favouring the 'rebound effect' and weight gain. More studies about the physiological implications of this mechanism of overconsumption are also required, including its magnitude, duration, implications to health and body weight maintenance, and effects on physiological and hormonal parameters. In the present study, the cats were challenged two times (days 22 and 42), but for only 1 day at a time, so it is unknown for how long the cats would have continued their pattern of overconsumption. Future studies are warranted to investigate the long-term implications to feline health.

Gastrointestinal transit time, nutrient digestibility and stool characteristics

The effect of fibre on gastrointestinal transit time was different depending on the fibre source; sugar cane fibre did not alter retention time, but cellulose addition significantly reduced it. We did not find published studies in cats, but several studies in dogs also failed to find an influence of fibre intake on retention time (Fahey et al., 1992; Hill et al., 2000). One study with cellulose (Burrows et al., 1982) found reduced gastrointestinal transit time in dogs. It is difficult to explain why the sugar cane and cellulose fibres would have different effects on this parameter, as they have very similar physical and chemical properties. One explanation may be the differences in water-holding capability and the particle size of the fibre source, larger for sugar cane fibre (188 μm) than for cellulose (112 μm). The results of increased faecal production (both volume and rate) were expected after the inclusion of non-digestible organic matter in the food. As non-fermentable fibre sources, however, the experimental diets did not reduce the faecal score of the cats, as has been previously shown (Meyer and Tungland, 2001).

The reduced digestibility of all nutrients except starch also was expected and has been observed previously (Fekete et al., 2004; Prola et al., 2010; Fischer et al., 2012). It was interesting that only sugar cane fibre inclusion reduced fat digestibility. This fibre source has around 6.4% lignin (Veloso et al., 2014), a compound with high affinity for binding bile acids (Kay, 1982) which reduces bile acid reabsorption and fat absorption (Kritchevsky and Story, 1974; Eastwood et al., 1986). For farm animals, lignin is usually considered undesirable, but considering the obesity problem of housecats, this might be considered a potential health-promoting compound. The high starch digestibility suggests the diets were adequately processed (de-Oliveira et al., 2008). Although the higher starch gelatinization for the SF20 diet, no difference in starch digestibility was verified among diets, suggesting adequate cooking degree. The reduction in total dietary fibre digestibility and butyric acid concentration in faeces by both fibre types suggests that these fibres have low fermentability by cats. Butyrate reduction after insoluble fibre addition may be a result of alteration on gut microbiota composition, SCFA intestinal absorption rate or a diluting effect promoted

by the increased stool production. However, it has been shown that sugar cane fibre has a slightly higher fermentability than cellulose (demonstrated by the higher dietary fibre apparent digestibility and higher faecal propionate concentration). This may be explained by its higher hemicellulose content (Veloso et al., 2014), which is zero in purified cellulose, and also by the higher gastrointestinal transit time which may allow more time for microbial degradation in the colon. This small fermentation of sugar cane fibre, however, did not reduce faecal DM or faecal score, and could be beneficial for gut health.

Conclusion

Adding insoluble fibre to the diet in the form of sugar cane fibre and cellulose does not induce satiety or change the pattern of food intake by cats. Furthermore, at the fibre inclusions used in the present study, cats can compensate for the lower food energy content by increasing DM intake, thereby maintaining ME consumption and body weight. An anatomical adaptation to higher DM consumption is postulated, and this predisposes, at least on a short-term basis, the cats to engage in higher food and energy intake when exposed to a palatable, energy-dense food.

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