Received: 23 May 2017 | Accepted: 28 July 2017

DOI: 10.1111/jpn.12803

ORIGINAL ARTICLE

WILEY Journal of Animal Physiology and Animal Nutrition

How does dietary particle size affect carnivore gastrointestinal transit: A dog model

A. De Cuyper[1] | M. Hesta[1] | S. Tibosch[2] | C. Wanke[2] | M. Clauss[3] | G. P. J. Janssens[1]

[1]Laboratory of Animal Nutrition, Department of Nutrition Genetics and Ethology, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium

[2]Medimetrics Personalized Drug Delivery Group, Eindhoven, The Netherlands

[3]Clinic for Zoo Animals Exotic Pets and Wildlife, University of Zurich, Zurich, Switzerland

Correspondence
A. De Cuyper, Laboratory of Animal Nutrition, Department of Nutrition, Genetics and Ethology, Faculty of Veterinary Medicine, Ghent University, Merelbeke, Belgium.
Email: annelies.decuyper@ugent.be

Funding information
Agentschap voor Innovatie door Wetenschap en Technologie, Grant/Award Number: 131448

Summary

The effect of dietary particle size on gastrointestinal transit in carnivores has not been studied and might offer more insight into their digestive physiology. This study evaluated the effect of two dietary particle sizes (fine = 7.8 mm vs. coarse = 13 mm) of chunked day-old chicks on transit parameters in dogs. Six beagle dogs were fed both dietary treatments in a crossover design of 7 days with transit testing on the fifth day. Transit parameters were assessed using two markers, that is a wireless motility capsule (IntelliCap®) and titanium oxide (TiO_2). Dietary particle size did not affect gastric emptying time (GRT), small bowel transit time (SBTT), colonic transit time (CTT) and total transit time (aTTT) of the capsule ($p > .05$). There was no effect of dietary particle size on TiO_2 mean retention time (MRT) ($p > .05$). The time of last TiO_2 excretion (MaxRT) differed ($p = .013$) between diets, being later for the coarse diet. Both MRT ($R = 0.617$, $p = .032$) and MaxRT ($R = 0.814$; $p = .001$) were positively correlated to aTTT. The ratio MRT/aTTT tended towards a difference between diets ($p = .059$) with the coarse diet exceeding fine diet values. Results show that the difference between capsule measurements and TiO_2 is larger for the fine than the coarse diet suggesting that the capsule becomes more accurate when dietary particle size approaches marker size. Dietary particle size might have affected transit parameters but differences are too small to claim major physiological consequences.

KEYWORDS
dietary particle size, dog, marker, motility capsule, transit

1 | INTRODUCTION

Texture is a complex dietary characteristic influenced by several factors such as hardness, viscosity and many others (Chen & Rosenthal, 2015). Particle size is one such factor that affects dietary texture and might influence digestive physiology. It is known that dietary particle size affects digesta passage in ruminants (Udén, 1988). In monogastric animals, particle size of plant-derived fibre in the diet can affect gastrointestinal transit times in humans, horses, rabbits, rats, pigs and poultry (Carré, 2000; Ferguson & Harris, 1997; Gidenne, Carré, Segura, Lapanouse, & Gomez, 1991; Heller et al., 1980; Stanogias & Pearce, 1985; Svihus, Hetland, Choct, & Sundby, 2002; Van Weyenberg, Sales, & Janssens, 2006; Vincent et al., 1995). However, little information exists on the effect of dietary particle size variation on transit times in carnivores. In dogs, the effect of varying texture—although not by particle size but by adding insoluble plant-derived fibre—on gastric emptying and/or total transit time has been studied to some extent. According to Burrows, Kronfeld, Banta, and Merritt (1982), the inclusion of cellulose in a canine diet decreases total transit time. By contrast, Pedreira et al. (2013) showed that the inclusion of 10% insoluble fibre (sugarcane fibre) in a dog's diet delayed the gastric emptying and colonic filling time.

Elucidating how dietary particle size influences gastrointestinal passage rate in carnivores might offer more insight into carnivore digestive physiology. It is known—although not well substantiated—that extending gastric emptying time in dogs may help to influence satiety.

Gastric fill and stomach extension followed by a subsequent slowing of gastric emptying (Weber et al., 2007) might be an important mechanism through which dogs get satiated. Pappas, Melendez, and Debas (1989) found that gradual gastric distention caused gradual inhibition of food intake in a non-cholinergic way and that satiety was not influenced by the nutrient content of the food. Given that free-ranging carnivores can be expected to swallow larger chunks of their prey compared to domestic animals fed processed feeds, there might be a general difference in the level of satiety experienced between free-ranging and domesticated carnivores.

We want to elucidate how particle size, as a texture-influencing factor, affects gastric emptying time and total transit time in carnivores. For this aim, the dog was used as a model for "a" carnivore. To remain true to the carnivore's natural diet, particle size was varied in a complete animal-based diet (Bosch, Hagen-Plantinga, & Hendriks, 2015; Plantinga, Bosch, & Hendriks, 2011) rich in animal fibre (i.e., poorly digestible animal tissues ((glyco)protein-rich matter such as raw bones, tendons, cartilage, skin, hair or feathers)) (Depauw et al., 2013). Currently, several methods are available for assessing transit in dogs (Wyse et al., 2003) but not all are equally accurate, non-invasive and practical. Recently, wireless motility capsules that are administered orally and measure pH and temperature throughout the gastrointestinal tract have been used successfully to assess passage rate in dogs (Boillat, Gaschen, Gaschen, Stout, & Hosgood, 2010). Therefore, transit was monitored using the IntelliCap® system (Medimetrics, Personalized drug delivery group, the Netherlands) together with the control marker titanium oxide (TiO_2).

2 | MATERIALS AND METHODS

2.1 | Animals and housing

Six healthy adult laboratory beagle dogs (*Canis lupus familiaris* L.) (four females and two males) aged between two and seven years with a body weight between 9 and 14.1 kg and body condition score between 3 and 5 of 9 were housed individually in neighbouring kennels at the Laboratory of Animal Nutrition of Ghent University (Merelbeke, Belgium).

2.2 | Experimental design and diets

Experimental procedures were approved by the Ethical Committee of the Faculty of Veterinary Medicine of Ghent University (EC2015/45) following the EU and Belgian Government-established norms and procedures. Dogs were fed two test diets in a crossover design of 7 days with passage rate testing on the fifth day of both periods. The test diets consisted exclusively of chunked day-old chicks (Kiezebrink Putten B.V., Hoge Eng Oost, the Netherlands) and differed only in particle size. The fine diet had a particle size of 7.8 mm whereas the coarse diet had a particle size of 13 mm (KOLBE AW 130 meat mincer; die size fine diet = 7.8 mm; die size coarse diet = 13 mm) (Figure 1). Because of the limited duration of the trial, it was chosen not to adjust the diet for eventual deviations from nutrient requirement guidelines (i.e., minerals and vitamins), in order to keep the intervention simple.

The nutrient composition of the diet is shown in Table 1. Dry matter (DM) and ash contents were determined by drying to a constant weight at 100°C and combustion at 550°C respectively. Crude protein (6·25 × N) was analysed using the Kjeldahl method (ISO 5983-1, ISO 2005a), and crude fat was analysed according to the Soxhlet method (ISO 1443, ISO 1973). Total fibrous matter and insoluble fibre were analysed according to the method of Cools, De Cuyper, Pauwels, and Janssens (2015). This method is based on the in vitro digestive simulation of Boisen and Fernández (1995) and Hervera, Baucells, Blanch, and Castrillo (2007) and resembles the TDF analysis according to Prosky et al. (1985) with this difference that the fibre fraction obtained includes not only the plant-derived carbohydrate fraction (TDF) but also animal fibre (protein-rich). Amino acids were analysed using the ISO 13903 method (ISO 2005b).

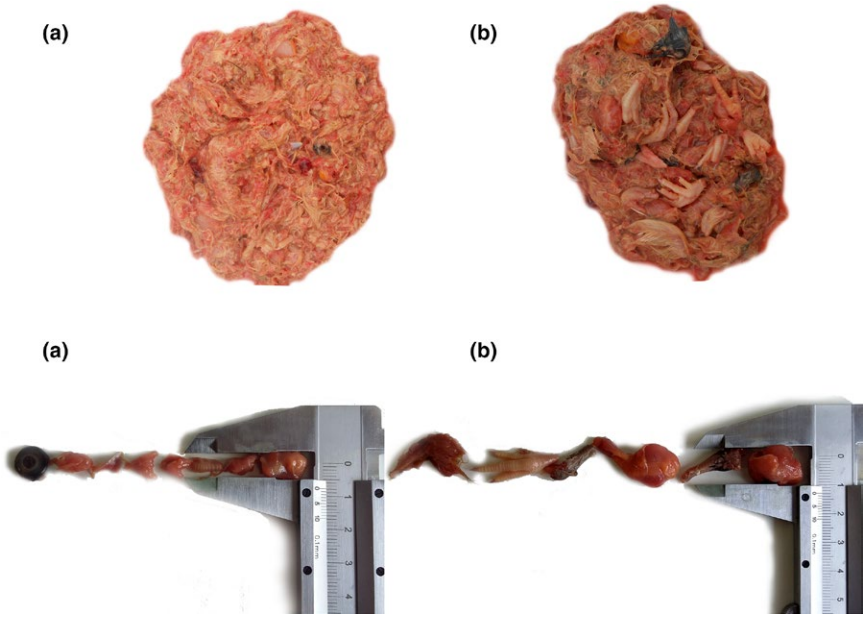

FIGURE 1 Chunked day-old chicks Particle size 7.8 mm (a) and 13 mm (b); particles can be lower or exceed the average particle size within a diet which is visible in the depicted caliper measurements

TABLE 1 Nutrient composition of chunked day-old chicks

	Chunked day-old chicks
Nutrient composition, % of DM	
Dry matter	24.9
Crude protein	57.3
Crude fat	22.7-26.4[a]
Total fibrous matter	38.0
Insoluble fibre	26.2
Crude ash	7.1
Crude fibre	2.5
Amino acid composition, g/kg DM	
Cysteine	14.5
Taurine	5.6
Methionine	14.7
Aspartic acid	57.6
Threonine	28.5
Serine	37.9
Glutamic acid	83.7
Glycine	47.7
Alanine	38.9
Valine	34.7
Isoleucine	28.3
Leucine	50.5
Tyrosine	20.8
Phenylalanine	32.2
Histidine	16.3
Lysine	39.7
Arginine	39.0
Metabolisable energy, kJ/100 g DM	418.4[b]

DM, dry matter.
[a]smallest value without hydrolysis, largest value with hydrolysis.
[b]The ME is the average of the values calculated by Atwater factors (16.7 * Crude protein + 37.7 * crude fat + 16.7 * NfE and the alternative predictive equation of the NRC (2006) (with NfE (Nitrogen free extract; 100 - moisture% - Crude protein% - Crude fat% - Crude fibre% - Crude ash%))).

Before the onset of the trial, all dogs were fed with chunked day-old chicks (13 mm) for three weeks. During the first week, the chunked chicks were gradually added to the usual kibble diet (0% to 100% chunked chicks of MER). The following two weeks, dogs were meal fed exclusively with chunked day-old chicks (13 mm) according to their maintenance energy requirements (based on NRC requirements (NRC, 2006) for adult laboratory dogs) to maintain constant bodyweight, which was assessed weekly. Five of the six dogs were willing to consume the diet from the beginning and consumed it within 5 min. One dog was more reluctant whereupon its meal was spread throughout the day. After the adaptation period, the crossover trial was executed with dogs being meal fed every day according to their individual maintenance energy requirements. Each dog always received the same amount of food throughout the crossover experiment so there was no difference in food intake between dietary treatments. The mean food intake (as fed) was 907.1 g day^{-1} dog^{-1} (± 348.4 g). All dogs had ad libitum water access.

2.3 | Gastric emptying and gastrointestinal passage

On the fifth day of every test period, gastric emptying and gastrointestinal transit were monitored by the IntelliCap® system (Medimetrics, Personalized drug delivery group, the Netherlands). The IntelliCap system consists of an electronic capsule that can be administered orally and measures pH and temperature throughout the gastrointestinal tract. This renders information on capsule location in the gastrointestinal tract. Capsule sizes were 11 mm diameter by 26.7 mm long (Zou, Shimizu, Wanke, & Iordanov, 2013).

All dogs were offered their daily meal in the morning. A maximum of 30 min was allowed to ingest the meal. All dogs finished their meal within the first 5 min except for one, which finished in 30 min. Before administration, IntelliCap® capsules were assembled according to Medimetrics standard operating procedures. After every dog had finished its meal, the IntelliCap capsule was administered by deep throat deposition followed by a rinse (approximately 20 mL) of drinking water to assist swallowing. A single IntelliCap® portable unit (data receiver) was mounted at the front of each kennel. From administration until excretion of the capsule, pH and temperature were measured and reported every 60 s until deactivation of the capsule.

Administration and excretion of the capsule were determined by the temperature profile and render a good estimate for the total transit time (aTTT). Gastric emptying is characterised by a quick rise in pH. Gastric residence time (aGRT) was therefore defined as the time interval between capsule administration and the abrupt increase in pH profile, that is passage of the pylorus. After entering the small intestine, there is a steady rise in pH followed by a pH plateau phase. Afterwards the pH suddenly drops by approximately 1.0 pH unit or more. This decrease indicates transit through the ileocolic valve. The time between the entry into the small bowel and the entry into the colon was defined as the small bowel transit time (aSBTT). Finally, the colonic transit time (aCTT) was defined as the time between the colon entry and the excretion of the capsule from the body (Figure 2) (Zou et al., 2013). Additionally, relative rGRT, rSBTT and rCTT were calculated by dividing the absolute value by the aTTT.

Additionally, on the same day as the capsule administration, both diets were enriched with 2 g/kg TiO_2 (VWR, International BVBA, Leuven, Belgium) as a control marker for total transit time. Faecal samples were collected constantly from one day before TiO_2 addition to the diet until two days after the TiO_2 addition. Faecal samples were always collected within 15 min after defecation. Faecal samples were weighed, dried at 60°C and analysed for Ti following Myers, Ludden, Nayigihugu, and Hess (2004) and the mean retention time was calculated. Mean retention time (MRT) of TiO_2, the best single measure of rate of passage through the gastrointestinal tract, was calculated using the Thielemans method (Thielemans, Francois, Bodart, & Thewis, 1978):

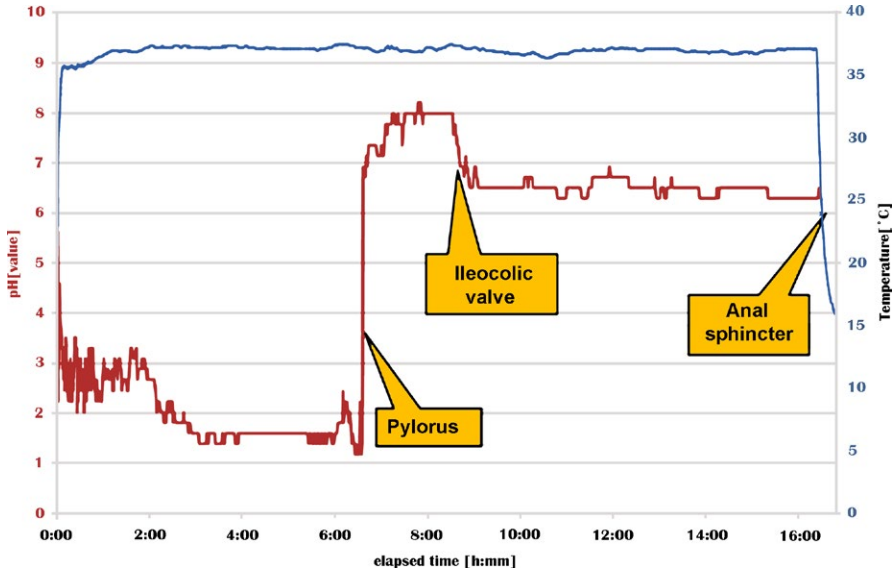

FIGURE 2 pH and temperature profile of a beagle dog gastrointestinal tract

$$\mathrm{MRT(h)} = \sum t_i C_i \Delta t_i / \sum C_i \Delta t_i$$

where C_i is the marker concentration in the interval indicated by time t_i (hours after marker administration) and Δt_i = the interval of the concerning sample:

$$\Delta t_i = ((t_{i+1} - t_i) + (t_i - t_{i-1}))/2$$

Furthermore, the time of last marker excretion (MaxRT) (<5% of the max. concentration) was registered for every dog and treatment. Additionally, the ratio MRT/aTTT and MaxRT/aTTT was calculated for both dietary treatments.

2.4 | Statistical analysis

Statistical analyses were performed using Superior Performing Software Systems version 23 (SPSS, Chicago, IL, USA). A paired *t* test was applied to compare the parameters aGRT, aSBTT, aCTT, aTTT, rGRT, rSBTT, rCTT, MRT, MRT/aTTT, MaxRT and MaxRT/aTTT for both diets. Additionally, a paired *t* test was applied to total transit measurements within diets for both types of marker (MRT vs. aTTT; MaxRT vs. aTTT).

Pearson correlations were performed between all capsule transit measures to test for example if a longer aGRT is linked to a longer aCTT (aGRT vs. aSBTT; aGRT vs. aCTT; aGRT vs. aTTT; aSBTT vs. aCTT; aSBTT vs. aTTT; aCTT vs. aTTT; rGRT vs. rSBTT; rGRT vs. rCTT; rSBTT vs. rCTT). Additionally, MRT and MaxRT were correlated with aTTT to compare the IntelliCap® method to the TiO_2 marker method.

3 | RESULTS

All dogs remained healthy throughout the study and consumed all provided food every day. The treatment with the IntelliCap system was safe and well tolerated by every dog. All capsules were excreted and recovered intact. On the fifth day of the first testing period when capsules were administered, one dog refused to eat the whole amount of food provided within the limited amount of time (356 g vs. 808 g). Subsequently, on the fifth day of the second test period, this dog was offered the same diminished amount of food to be able to compare test periods.

TABLE 2 Average transit parameters for two test diets (7.8 mm vs. 13 mm)

	Fine diet (7.8 mm)		Coarse diet (13 mm)		
	Mean	SD	Mean	SD	*p*
Absolute capsule times, hr					
aGRT	15.4	6.6	13.7	3.8	.36
aSBTT	2.6	0.75	2.4	0.34	.60
aCTT	14.8	8.3	12.2	5.1	.46
aTTT	32.8	13.5	28.2	7.5	.35
Relative capsule times, % of aTTT					
rGRT	48.3	10.7	48.8	10.2	.93
rSBTT	9.1	4.4	8.7	1.9	.80
rCTT	42.5	12.3	42.5	10.2	.99
Mean retention time, hr					
MRT	19.5	5.1	22.0	3.8	.16
Maximum retention time, hr					
MaxRT	30.8	10.6	33.3	9.6	.013
Ratio MRT vs. aTTT					
MRT/aTTT	0.67	0.24	0.81	0.17	.059
Ratio MaxRT vs. aTTT					
MaxRT/aTTT	0.97	0.12	1.19	0.21	.17

aGRT, absolute gastric residence time; aSBTT, absolute small bowel transit time; aCTT, absolute colonic transit time; aTTT, absolute total transit time; rGRT, relative gastric residence time; rSBTT, relative small bowel transit time; rCTT, relative colonic transit time; MRT, mean retention time; MaxRT, maximum retention time; n = 6 dogs.

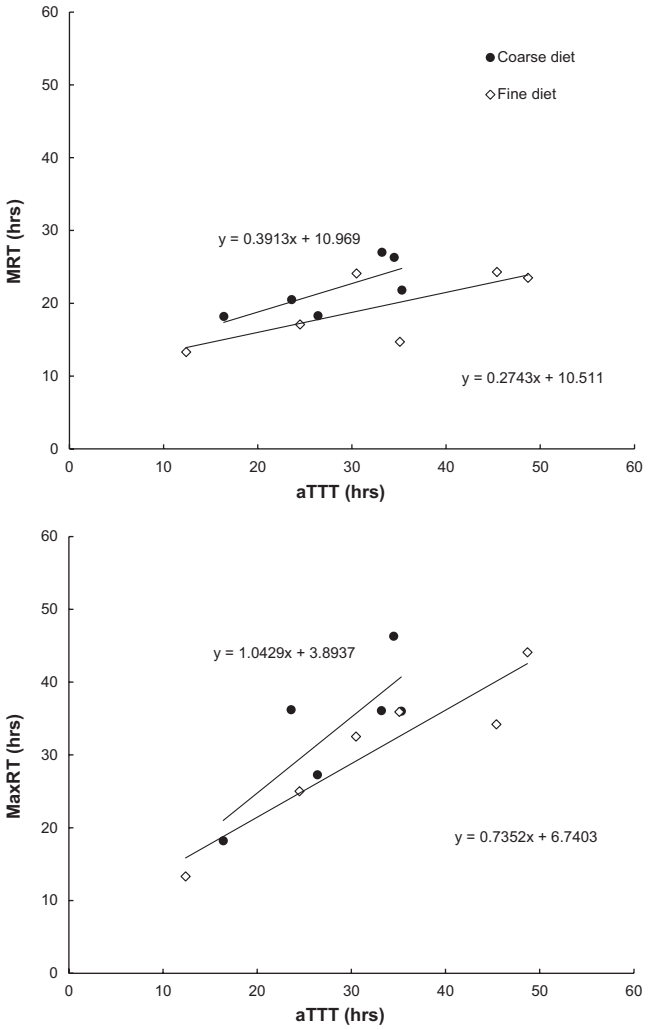

FIGURE 3 Correlations between marker retention times and capsule total transit time MRT, mean retention time (TiO_2); Max RT, time point of last marker excretion or maximum retention time (TiO_2); aTTT, total transit time (capsule); n = 6 dogs

Although aGRT, aCTT and aTTT were numerically lower on the coarse diet, there were no capsule transit time differences between diets ($p > .05$) (Table 2). The average total transit time of the capsule was 30.5 ± 10.6 h across diets. This aTTT consisted of constant proportions of 0.50 rGRT, 0.09 rSBTT and 0.42 rCTT that did not differ between diets. Both aGRT ($R = 0.825$; $p = .001$) and aCTT ($R = 0.913$; $p < .001$) but not aSBTT ($R = 0.344$; $p > .05$) were positively correlated with aTTT. As expected, rGRT was negatively correlated with rCTT ($R = -0.950$; $p < .001$), but neither one was correlated with rSBTT.

The mean retention time (MRT) did not differ between dietary treatments ($p > .05$) (Table 2). The average MRT was 20.8 ± 4.5 hr across diets. However, the time point of last marker excretion (MaxRT) differed significantly ($p = .013$) between diets, with the coarse diet exceeding the average value of the fine. Both MRT ($R = 0.617$, $p = .032$) and MaxRT ($R = 0.814$; $p = .001$) were positively correlated to the aTTT (Figure 3). The slope of the respective regression equations was <1 (0.136, 0.497) for the MRT-aTTT relationship, but included 1 in the confidence interval (0.368, 1.113) for the MaxRT-aTTT relationship. Within diets, MRT and aTTT differed (fine diet: $p = .026$; coarse diet: $p = .032$). The difference between MaxRT and aTTT was not significant within the fine diet ($p = .4$) but tended towards significance in the coarse diet ($p = .074$).

The difference between the diets in the ratio MRT/aTTT tended towards significance ($p = .059$). The ratio showed higher values (closer to 1) for the coarse diet compared to the fine diet meaning that the MRT lies closer to the aTTT for the coarse diet compared to the fine diet. The MaxRT/aTTT ratio was 0.97 vs. 1.19 for the fine and coarse diet, respectively ($p = .17$), indicating that on the coarse diet, the capsule tended to be excreted sooner than the last titanium marker.

4 | DISCUSSION

Overall, dietary particle size did not affect gastric emptying time and did not affect other transit parameters with the exception of MaxRT; however, dietary particle size appeared to influence how the two marker systems (a powder applied to the diet, or a larger capsule) compared with each other. Therefore, although our data do not indicate major changes in transit parameters, it indicates that the difference in particle size between diet and marker can have a relevant effect on measurements.

4.1 | Effects of dietary particle size with constant marker size

4.1.1 | Gastric emptying

Our results show that dietary particle size does not seem to have an effect on gastric emptying time (aGRT). We would have expected the coarse diet to slow down the gastric emptying compared to the fine diet. In humans, Vincent et al. (1995) showed that coarse bran compared to fine bran slowed down gastric emptying, although this concerned fibre particle size and not complete dietary particle size as is the case in our study. In general, the effect of dietary particle size on gastrointestinal transit in carnivores has not been studied. Adding texture to a canine diet—not by increasing particle size but through the inclusion of insoluble fibre (which might unknowingly have lead to the addition of more coarse particles)—delayed gastric emptying (Pedreira et al., 2013). However, we could not observe this effect in this study where texture was varied through particle size variation. The main reason probably lies in the particle size difference between the two test diets which was only 5.2 mm and too small to provoke significant differences. Additionally, particle size variation within one diet might have undone the particle size difference between diets (Figure 1). Unfortunately, no greater difference could be obtained due to limitations of the food processing equipment. It might be that large particles from both diets were retained in the stomach until the interdigestive migratory myoelectric complex (IMMC) occurred as they both exceeded 5 mm of particle size (see below). However, the latter threshold sizes concern non-food indigestible particles which differ

from dietary particle kinetics. Dietary particles that are too large to pass the pylorus will be propulsed back into the stomach and will be reduced in size through mechanical breakdown via repetitive muscular contractions until they are able to pass the pylorus (Martinez & Papich, 2009; Wyse et al., 2003). Additionally, due to the plasticity and form of food particles, it is still possible that larger particles are "moulded" through the pylorus and thereby react differently than indigestible solids (Carré, 2000). From this point of view, the fine diet (7.8 mm) still might have left the stomach earlier than the coarse diet (13 mm) although the aGRT was not affected by the diet type. However, any small difference in aGRT between the fine and coarse diet might have been missed mainly due to the fact that the capsule reacts as an indigestible solid with the aGRT reflecting the time at which the whole solid meal has left the stomach and not the "average" food particle residence time.

4.1.2 | Small bowel transit, colon transit and total transit

Neither of the other capsule-measured transit times (aSBTT, aCTT, aTTT, rSBTT and rCTT) nor the TiO_2-MRT were affected by dietary particle size. Only the MaxRT was affected by dietary particle size. The effect of particle size on small bowel transit has hardly been studied. Our results show similar SBTT values (2.5 hr ± 0.56) as found by Boillat, Gaschen, Gaschen, et al. (2010), Boillat, Gaschen, and Hosgood (2010) that also used a wireless motility capsule to study gastrointestinal transit times in dogs. The SBTT found in this study reflects the typical speed at which the IMMC propulses through the small bowel (Code & Marlett, 1975) and does not show any significant effect of particle size. As aSBTT (in contrast to aGRT and aCTT) was not correlated to aTTT, small intestinal transit appears to be particularly consistent, whereas transit through the other sections of the gastrointestinal tract is subject to more variation.

Additionally, no effect of particle size was seen on the aCTT. However, colonic transit times tend to show high intra-individual variability (Boillat, Gaschen, Gaschen, et al. (2010)) which was also observed in our study and might be due to the control of defecation by the dog for reasons unrelated to digestive physiology. Therefore, it might be difficult to observe differences in CTT provoked by particle size. Total transit time (aTTT) was not affected by particle size as well which is something we would have expected based on other species. Van Weyenberg et al. (2006) for example reviewed passage rate in horses and its influencing factors. Whenever feed particle size is reduced, the mean retention time is increased, particularly in the colon. By adding more texture to horse diets by for example long hay, the passage rate is increased compared to smaller particles in for example pelleted diets. The authors do differentiate between total dietary particle size and fibre length. Reducing the fibre length can shorten the mean retention time in the gut although this can vary according to the fibre source. Similarly as for the aGRT, it might still be that the particle size difference between diets in this study was too small to evoke any difference in aSBTT, aCTT and aTTT. However, the most plausible explanation for the aTTT is the size of the marker. In contrast to the aTTT, the MaxRT did experience a significant effect of diet type, that is particle size with the coarse diet having a longer MaxRT than the fine diet suggesting that the TiO_2 marker was more sensitive than the wireless motility capsule and that the diet might have had some effect on digesta transit.

4.2 | Effects of marker size with constant diet

Marker size seemed to influence transit measurements. Our results showed that within diets, TiO_2-MRT values and aTTT capsule values were significantly different with the aTTT exceeding the MRT value. The MaxRT did not significantly differ from aTTT within the fine diet but showed a tendency towards significance within the coarse diet. Clearly, the difference in marker size (i.e., powder vs. large inert capsule) seemed to affect transit parameters, and this was probably mainly at the level of the stomach. In dogs, Itoh, Higuchi, Gardner, and Caldwell (1986) reported that increasing the particle size of inert radio-opaque markers slowed down gastric emptying. Nelson, Jergens, Miles, and Christensen (2001) reported that increasing particle size (of barium impregnated spheres) increased the duration of the time needed to reach a certain percentage of gastric emptying but it also increased the interindividual variability in gastric emptying time. It is known for dogs that objects of different size—as in non-food accidentally ingested—differ in the time at which they leave stomach. Dressman (1986) reviewed and reported that particles of ≤1.6 mm leave the stomach sooner than the meal, and once particles exceed 2.4 mm, the particles are expelled later than the meal. Martinez and Papich (2009) state that particles ≤2-3 mm should be able to pass the canine pylorus immediately. Others state that the threshold lies at ≤5 mm diameter (Itoh et al., 1986; Wyse et al., 2003). Once exceeding the previously mentioned diameters, non-food particles are retained in the stomach until the interdigestive migratory myoelectric complex (IMMC) occurs, which propels large particles towards the duodenum (Itoh et al., 1986; Wyse et al., 2003). Once the stomach is passed, there does not seem to be an effect of marker size on the rest of the transit through the gastrointestinal tract. Also Bruce, Guilford, Hedderley, and McCauley (1999) did not see any difference between the CTTs in dogs of small inert radiopaque polyethylene spheres (1 mm) and large spheres (5 mm).

In our study, capsule sizes were 11 mm diameter by 26.7 mm long. Consequently, the capsule was not able to pass the pylorus and will have left the stomach with the IMMC for both diets. By contrast, the powder of the TiO_2 marker will have left the stomach earlier together with the food. Without subdivision in dietary treatments, the MRT (obtained by the TiO_2 marker) however did positively correlate with the aTTT (capsule) (Figure 3) although the aTTT was always higher than the MRT. The latter is logical as the MRT represents the mean retention time of the food in the gut (Thielemans et al., 1978) whereas the capsule aTTT represents the transit time of the last food that left the stomach (Martinez & Papich, 2009). Therefore, the MaxRT is a more comparable measure for aTTT, and indeed the MaxRT was more strongly correlated to the aTTT ($R = 0.814$ compared to $R = 0.617$) (Figure 3). However, the important question might be how the marker

size compares to the particle size of the diet fed with that particular marker.

4.3 | Difference in particle size between diet and marker

The dietary difference for the ratio MRT/aTTT tended towards significance (p = .059). The MRT/aTTT ratio for the coarse diet was significantly higher and closer to one than the fine diet ratio meaning that the aTTT and MRT were more similar for the coarse than the fine diet. One could speculate that the large particles from the coarse diet resided longer in the stomach (like the capsule) than the particles of the fine diet. The MaxRT/aTTT ratio was 0.97 vs. 1.19 for the fine diet and the coarse diet, respectively, indicating that on the coarse diet, the capsule tended to be even excreted sooner than the last titanium marker. Also, the higher MaxRT of the coarse diet (33.3 hr vs. 30.8 hr) might indicate that some parts of the coarse diet are not passed as fast as the fine diet.

4.4 | Biological implications and conclusions

In general, we could not prove any substantial effect of particle size on the transit characteristics obtained by the IntelliCap capsule. However, MaxRT values obtained through the second marker system (TiO_2) differed between diets. The MaxRT difference might not be big enough to cause any physiological consequences but indicates that particle size might affect the mechanics in the gut. One could state that particle size might have been undone due to chewing on the food although this was very unlikely. Wolves, the dog's wild ancestor (Axelsson et al., 2013), are known to gorge feed (Bosch et al., 2015) and this is a characteristic still to be found in dogs. During this experiment, a similar behaviour was observed with all dogs barely chewing their food. Overall, analyses show that the titanium marker and the capsule differ more on the fine diet and less on the coarse diet. For the coarse diet, it could even happen that the capsule is excreted sooner than the last titanium marker. One could state that for a wild carnivore or a carnivore fed a natural diet (whole prey, coarse), the capsule could be an adequate reflection of how passage happens through the gut. However, for artificial diets with fine particle sizes and less texture, the capsule might not be a representative marker for the gastrointestinal passage with the passage probably being faster than measured by the capsule.

Although we could only prove a small MaxRT difference between diets (33.3 hr vs. 30.8 hr, coarse and fine diet respectively), there is reason to believe that adding texture to a carnivore's natural diet influences transit time substantially. This might not only be through the factor particle size but even more likely through the addition of animal fibre (i.e., poorly digestible animal tissues [glyco]protein-rich matter such as raw bones, tendons, cartilage, skin, hair or feathers), which might play a more crucial role in guiding digestive processes such as transit time. Depauw et al. (2013) showed that feeding whole rabbit to cheetahs compared to supplemented beef resulted in a lower amount of putrefactive fermentation products and a better faecal consistency. The mechanism underlying the improved gut health was not completely clear, although it was speculated that the presence of more animal fibre in the whole rabbit diet might have influenced gastric emptying, passage rate, motility and absorption. In dogs, it is shown that by the inclusion of plant-derived insoluble fibre in the diet, transit is affected. According to Burrows et al. (1982), the inclusion of cellulose in a canine diet decreases total transit time. Pedreira et al. (2013) showed that the inclusion of 10% insoluble fibre (sugarcane fibre) in a dog's diet delays the gastric emptying and colonic filling time. It might be that animal fibre exerts similar effects on transit parameter as the plant-derived analog and that through extended gastric fill, satiety is prolonged (Pappas et al., 1989). However, more research is warranted concerning the effect of texture—by varying the dietary animal fibre content, or maybe equally important, the particle size of animal fibre in the diet—on transit parameters in carnivores.

ACKNOWLEDGEMENTS

The authors sincerely thank Kiezebrink Putten B.V. for the provision of experimental diets. The authors also gratefully acknowledge Daniel Tensy involved in the caretaking of the dogs and sample collection. This work was supported by the Institute for Promotion of Innovation through Science and Technology in Flanders (IWT) (grant number 131448).

ORCID

A. De Cuyper http://orcid.org/0000-0003-2057-8283

M. Clauss http://orcid.org/0000-0003-3841-6207

REFERENCES

Axelsson, E., Ratnakumar, A., Arendt, M. L., Maqbool, K., Webster, M. T., Perloski, M., … Lindblad-Toh, K. (2013). The genomic signature of dog domestication reveals adaptation to a starch-rich diet. *Nature*, *495*, 360–364.

Boillat, C. S., Gaschen, F. P., Gaschen, L., Stout, R. W., & Hosgood, G. L. (2010). Variability associated with repeated measurements of gastrointestinal tract motility in dogs obtained by use of a wireless motility capsule system and scintigraphy. *American Journal of Veterinary Research*, *71*, 903–908.

Boillat, C. S., Gaschen, F. P., & Hosgood, G. L. (2010). Assessment of the relationship between body weight and gastrointestinal transit times measured by use of a wireless motility capsule system in dogs. *American Journal of Veterinary Research*, *71*, 898–902.

Boisen, S., & Fernández, J. A. (1995). Prediction of the apparent ileal digestibility of protein and amino acids in feedstuffs and feed mixtures for pigs by in vitro analyses. *Animal Feed Science and Technology*, *51*, 29–43.

Bosch, G., Hagen-Plantinga, E. A., & Hendriks, W. H. (2015). Dietary nutrient profiles of wild wolves: insights for optimal dog nutrition? *British Journal of Nutrition*, *113*, S40–S54.

Bruce, S. J., Guilford, W. G., Hedderley, D. I., & McCauley, M. (1999). Development of reference intervals for the large intestinal transit of radiopaque markers in dogs. *Veterinary Radiology & Ultrasound*, *40*, 472–476.

Burrows, C. F., Kronfeld, D. S., Banta, C. A., & Merritt, A. M. (1982). Effects of fiber on digestibility and transit time in dogs. *The Journal of Nutrition*, *112*, 1726–1732.

Carré, B. (2000). Effets de la taille des particules alimentaires sur les processus digestifs chez les oiseaux d'élevage. *Productions Animales - Paris - Institut National de la Recherche Agronomique*, *13*, 131–136.

Chen, J., & Rosenthal, A. (Eds.), 2015. *Modifying food texture Volume 2: Sensory analysis, consumer requirements and preferences*. Cambridge: Woodhead Publishing, Elsevier.

Code, C. F., & Marlett, J. A. (1975). The interdigestive myo-electric complex of the stomach and small bowel of dogs. *Journal of Physiology*, *246*, 289–309.

Cools, A., De Cuyper, A., Pauwels, J., & Janssens, G. P. (2015). Animal fibre: A key nutrient to carnivores but how to determine this dietary fraction analytically? In: M. Clauss, T. Huisman, F. vanAltena, & D. Vermeulen (Eds.), *8th European zoo nutrition conference* (p. 14). Arnhem: EAZA Nutrition Group.

Depauw, S., Hesta, M., Whitehouse-Tedd, K., Vanhaecke, L., Verbrugghe, A., & Janssens, G. P. J. (2013). Animal fibre: The forgotten nutrient in strict carnivores? First insights in the cheetah. *Journal of Animal Physiology and Animal Nutrition*, *97*, 146–154.

Dressman, J. B. (1986). Comparison of canine and human gastrointestinal physiology. *Pharmaceutical Research*, *3*, 123–131.

Ferguson, L. R., & Harris, P. J. (1997). Particle size of wheat bran in relation to colonic function in rats. *LWT - Food Science and Technology*, *30*, 735–742.

Gidenne, T., Carré, B., Segura, M., Lapanouse, A., & Gomez, J. (1991). Fibre digestion and rate of passage in the rabbit: Effect of particle size and level of lucerne meal. *Animal Feed Science and Technology*, *32*, 215–221.

Heller, S. N., Hackler, L. R., Rivers, J. M., Van Soest, P. J., Roe, D. A., Lewis, B. A., & Robertson, J. (1980). Dietary fiber: The effect of wheat bran on colonic function in young adult men. *The American Journal of Clinical Nutrition*, *33*, 1734–1744.

Hervera, M., Baucells, M. D., Blanch, F., & Castrillo, C. (2007). Prediction of digestible energy content of extruded dog food by in vitro analyses. *Journal of Animal Physiology and Animal Nutrition*, *91*, 205–209.

ISO. (1973). *Meat and meat products – Determination of total fat content (ISO 1443)*. Geneva, Switzerland: International Organisation for Standardization.

ISO. (2005a). *Animal feeding stuffs-determination of nitrogen content and calculation of crude protein content - Part 1: Kjeldahl method (ISO 5983–1)*. Geneva, Switzerland: International Organisation for Standardization.

ISO. (2005b). *Animal feeding stuffs – Determination of amino acids content ISO 13903*. Geneva, Switzerland: International Organisation for Standardization.

Itoh, T., Higuchi, T., Gardner, C. R., & Caldwell, L. (1986). Effect of particle size and food on gastric residence time of non-disintegrating solids in beagle dogs. *Journal of Pharmacy and Pharmacology*, *38*, 801–806.

Martinez, M. N., & Papich, M. G. (2009). Factors influencing the gastric residence of dosage forms in dogs. *Journal of Pharmaceutical Sciences*, *98*, 844–860.

Myers, W. D., Ludden, P. A., Nayigihugu, V., & Hess, B. W. (2004). Technical Note: A procedure for the preparation and quantitative analysis of samples for titanium dioxide. *Journal of Animal Science*, *82*, 179–183.

Nelson, O. L., Jergens, A. E., Miles, K. G., & Christensen, W. F. (2001). Gastric emptying as assessed by barium-impregnated polyethylene spheres in healthy dogs consuming a commercial kibble ration. *Journal of the American Animal Hospital Association*, *37*, 444–452.

NRC. (2006). *Nutrient requirements and dietary concentrations. Nutrient requirements for dogs and cats*. 2nd edn. Washington, D.C: National Academy Press. pp. 359.

Pappas, T. N., Melendez, R. L., & Debas, H. T. (1989). Gastric distension is a physiologic satiety signal in the dog. *Digestive Diseases and Sciences*, *34*, 1489–1493.

Pedreira, R., Silva, F., Avante, M., Loureiro, B., Matheus, L., Ferreira, C., … Carciofi, R., 2013: Insoluble fiber delays gastric emptying and colonic filling time of dogs fed kibble diets. *Proceedings of The WALTHAM International Nutritional Sciences Symposium* p. 48.

Plantinga, E. A., Bosch, G., & Hendriks, W. H. (2011). Estimation of the dietary nutrient profile of free-roaming feral cats: Possible implications for nutrition of domestic cats. *British Journal of Nutrition*, *106*, S35–S48.

Prosky, L., Asp, N. G., Furda, I., Devries, J. W., Schweizer, T. F., & Harland, B. F. (1985). Determination of total dietary fiber in foods and food-products - collaborative study. *Journal of the Association of Official Analytical Chemists*, *68*, 677–679.

Stanogias, G., & Pearce, G. R. (1985). The digestion of fibre by pigs 1. The effects of amount and type of fibre on apparent digestibility, nitrogen balance and rate of passage. *British Journal of Nutrition*, *53*, 513–530.

Svihus, B., Hetland, H., Choct, M., & Sundby, F. (2002). Passage rate through the anterior digestive tract of broiler chickens fed on diets with ground and whole wheat. *British Poultry Science*, *43*, 662–668.

Thielemans, M., Francois, E., Bodart, C., & Thewis, A. (1978). Mesure du transit gastrointestinal chez le porc à l'aide des radiolanthanides. Comparaison avec le mouton. *Annales de Biologie Animale Biochimie Biophysique EDP Sciences*, *18*, 237–247.

Udén, P. (1988). The effect of grinding and pelleting hay on digestibility, fermentation rate, digesta passage and rumen and faecal particle size in cows. *Animal Feed Science and Technology*, *19*, 145–157.

Van Weyenberg, S., Sales, J., & Janssens, G. P. J. (2006). Passage rate of digesta through the equine gastrointestinal tract: A review. *Livestock science*, *99*, 3–12.

Vincent, R., Roberts, A., Frier, M., Perkins, A. C., MacDonald, I. A., & Spiller, R. C. (1995). Effect of bran particle size on gastric emptying and small bowel transit in humans: A scintigraphic study. *Gut*, *37*, 216–219.

Weber, M., Bissot, T., Servet, E., Sergheraert, R., Biourge, V., & German, A. J. (2007). A high-protein, high-fiber diet designed for weight loss improves satiety in dogs. *Journal of Veterinary Internal Medicine*, *21*, 1203–1208.

Wyse, C. A., McLellan, J., Dickie, A. M., Sutton, D. G. M., Preston, T., & Yam, P. S. (2003). A review of methods for assessment of the rate of gastric emptying in the dog and cat: 1898–2002. *Journal of Veterinary Internal Medicine*, *17*, 609–621.

Zou, H., Shimizu, J., Wanke, C., & Iordanov, V. 2013: Transit and pH in fasted beagle dogs with the IntelliCap system. *Controlled Release Society Meeting* pp. 5–6.

How to cite this article: De Cuyper A, Hesta M, Tibosch S, Wanke C, Clauss M, Janssens GPJ. How does dietary particle size affect carnivore gastrointestinal transit: A dog model. *J Anim Physiol Anim Nutr*. 2018;102:e615–e622. https://doi.org/10.1111/jpn.12803