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Feeding practices and other factors associated with faecal consistency and the frequencies of vomiting and diarrhoea in captive tigers (*Panthera tigris*)

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Abstract

Gastrointestinal (GI) health is important to the welfare of captive tigers, and diet is considered a likely influencing factor. A survey was performed to collect information on GI health indicators and diet of tigers housed in zoological facilities across the globe. Completed surveys were received for one tiger from each of 32 facilities. Three (9%) tigers were reported as being diagnosed as having current GI disease; 24 (75%) had ideal (soft with shape) faeces 'often' to 'always' during the four weeks before survey completion. Potential associations between current GI disease and other variables could not be explored because of the low disease prevalence. Commercial raw meat diets were the most commonly fed diet type, and the most common food source was horse. Upon multivariate analysis, including country as a covariate, the odds and frequency of vomiting during the previous six months increased with the frequency of feeding muscle meat and chicken, and decreased as the frequency of feeding long bones increased. The odds and frequency of diarrhoea over the previous six months increased with the frequency of feeding beef and muscle meat; and the frequency of liquid faeces in the previous four weeks increased with oral antimicrobial treatment and increasing frequency of feeding beef. Although limited by the small sample size, these findings characterised the nutritional care that captive tigers currently receive and provided preliminary insight into dietary associations with indicators of GI health. The findings support the need to consider species-specific dietary adaptations and for further investigations into the health impact of diet in captive tigers.

Keywords: felid, husbandry, diet, captive management, nutrition

1. Introduction

Animal welfare is important to the success of *ex situ* species conservation efforts (Swaisgood, 2007). However, certain elements of the captive environment may compromise the welfare and, thus, reproductive success of the animals it is designed to protect. A classic example of this is the role that the captive environment is thought to play in the health and welfare of captive cheetahs (*Acinonyx jubatus*) (Terio *et al.*, 2004; Wells *et al.*, 2004), as may be reflected in their gastrointestinal (GI) health (Munson *et al.*, 2005; Whitehouse-Tedd *et al.*, 2015).

Various GI problems have been recorded for both wild and captive tigers (*Panthera tigris*), including trematode and nematode infection (Anderson *et al.*, 2018; González *et al.*, 2007), haemorrhagic enterocolitis as a result of *Clostridium perfringens* infection (Zhang *et al.*, 2012), gastric dilatation with or without enterotoxaemia associated with *C. perfringens* (Anderson *et al.*, 2018), and inflammatory bowel disease (Crook and Carpenter, 2014). Multiple historic reports exist of captivity-associated 'tiger disease,' first observed in the early 1960s in a German zoo, which is generally believed to be caused by pancreatic dysfunction or disturbance of GI microbiota (Kloss and Lang, 1976). This

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disease is now largely recognised as chronic inflammatory bowel disease, for which treatment has included dietary modifications (Crook and Carpenter, 2014).

The link between diet and GI health has been explored in several carnivore species, including felids. Differences in faecal characteristics have been evaluated among five captive felid species, including Amur (*P. t. altaica*) and Indochinese (*P. t. corbetti*) tigers fed a beef-based raw diet (Vester *et al.*, 2008). Results indicated that Indochinese tigers had significantly looser faeces than other felid groups, suggesting that species or subspecies can differ in their sensitivities to the same diet. In a separate study (Vester *et al.*, 2010), two subspecies of tiger – Malayan (*P. t. jacksoni*) and Amur – had an ideal faecal score (mean score, 2.8/5; soft, moist, formed faeces) when consuming a horse-based diet, whereas jaguars (*Panthera onca*), cheetahs, and domestic cats (*Felis catus*) had ideal faecal scores when consuming a beef-based diet. The investigators suggested that tigers may be better suited than other species to a diet containing a non-fermentable fibre source, greater crude protein digestibility, or less collagen (Vester *et al.*, 2010). Work by Kerr *et al.* (2013) confirmed that the differences reported by Vester *et al.* (2010) were linked to fibre and meat source, but no significant difference was identified between mean faecal scores for Malayan tigers fed horse (3.6/5) vs beef (3.0/5) when plant fibre source was the same. However, mean faecal score (based on a 5-point scale, where 1=dry, 3=ideal, and 5=liquid) was significantly greater when Malayan and Siberian tigers were fed a beef-based diet with beet pulp (3.8/5 and 4.1/5, respectively) vs cellulose (3.0/5 and 3.3/5, respectively) as a fibre source. Additionally, faecal consistency improved when beet pulp inclusion in the diet increased from 2 to 4% (Kerr *et al.*, 2013). In these studies, faecal scores, which were assigned per the Association of Zoos and Aquariums (AZA) Felid Taxon Advisory Group (TAG) faecal scoring system (AZA Tiger Species Survival Plan®, 2016), were greater (wetter, looser) than ideal (>3.5) for tigers fed higher levels of more fermentable fibre (beet pulp), compared with those fed less fermentable cellulose, hence both type and amount of dietary fibre – possibly more so than meat type – must be considered linked with faecal consistency in this species.

In contrast, the presence of animal fibre (e.g. fur, cartilage, bone, and connective tissue) can significantly lower putrefaction of digesta in the colon of cheetahs, suggesting that this may be a more beneficial source of fermentation in this and possibly other felid species (Depauw *et al.*, 2011). Using an epidemiological approach, Whitehouse-Tedd *et al.* (2015) identified feeding horse meat as a significant risk factor for GI disease in captive cheetahs, whereas feeding muscle meat and the inclusion of skeletal components in the diet were identified as protective factors.

Compared with cheetahs, the effect of diet on GI health and disease in tigers has been largely unexplored, but it remains important to understand to ensure their nutritional and, hence, welfare needs are met (AZA, 2012). The aim of this study was to address this knowledge gap by conducting an initial epidemiological survey of potential indicators of GI health in the captive tiger population. This included characterising the nutritional and other husbandry factors that may be associated with faecal consistency and clinical signs of GI disease or digestive disorders (i.e. vomiting or diarrhoea) as previously described for the species (Bush *et al.*, 1987; Seidel and Wisser, 1987; Seifert and Muller, 1987).

2. Materials and methods

Survey design

A survey was designed to collect data on several variables hypothesised *a priori* as potentially associated with GI health and disease: facility of origin (country and number of tigers in collection), tiger characteristics (age, sex, and body condition score (BCS)) and health status (current and prior diagnoses), preventive or clinical treatments and health monitoring frequency, and diet characteristics (composition, feeding frequencies and amounts, and supplements provided). It was designed to take no more than ten minutes to complete and was primarily modelled on the survey used in a similar study of cheetah GI health and disease (Whitehouse-Tedd *et al.*, 2015). After the first draft of the survey was complete, it was reviewed by two independent nutrition experts and the project supervisor. The main focus of the review was to ensure questions would be interpreted consistently by respondents, and to remove any redundant queries. The revised survey was then sent to the Species Survival Plan (SSP) coordinator and tiger management group of the AZA for further review. The major concern raised was the time the survey would take, as the original request was for each facility to complete questions for up to three animals (as per Whitehouse-Tedd *et al.*, 2015). The decision was therefore made to request completion of only one survey per facility, with the aim of increasing response rate, and to minimise the number of questions.

The survey (available from the corresponding author on request) included questions on five information categories; animal and facility details, potential indicators of GI health, veterinary information not related to GI disease, veterinary information related to GI disease and dietary information. The first section requested information on the job title of the respondent, the name and country of the facility, and the international studbook number and subspecies of the tiger. The second section examined how frequently the tiger had vomited and had diarrhoea over the previous six months, the proportion of tigers in the collection that had vomited within the previous week, and the observed consistency of the tigers' faeces per the standardised faecal scoring system developed by the Felid TAG in 2014 (AZA Tiger Species Survival Plan®, 2016). For this last question, respondents were asked to look at the photographs and descriptions provided, and then rate the frequency that they had observed each consistency within the previous four weeks. Briefly, the first sentence of each more detailed description that accompanied the photographs was as follows: 1 (i.e. extremely dry) = 'Hard, dry, multiple pellets that are easy to crumble or break apart into pieces', 2 (i.e. firm and dry) = 'Very firm, with some moisture. Segmentation is apparent and likely occurs as more than one faecal unit,' 3 (i.e. soft with shape) = 'Moist, surface that is pliable and formed,' 4 (i.e. soft without shape) = 'Very moist, has some texture, and occurs in piles or spots,' and 5 (i.e. liquid) = 'Watery liquid, that can be poured and occurs in puddles and flattens and may occur with splatter marks.'

The third section included details of veterinary care, health monitoring, and any issues unrelated to GI disease, such as canine distemper or feline immunodeficiency virus infection. Respondents were asked how frequently the animal had received certain types of veterinary care or health monitoring (e.g. vaccinations, weight checks, and faecal sample tests) over the previous year.

The fourth section required details of health issues related to GI disease and whether the tiger suffered from any conditions which, although not diet-related *per se,* might contribute to overall GI health (e.g. intestinal parasites). The fifth section asked respondents to estimate the proportions of various diet types provided to the tiger (e.g. carcass type, chunk muscle, or commercially produced product). Respondents were asked to describe the feeding frequencies of certain food sources (e.g. beef, horse, or chicken) and ingredients (e.g. bones, fur, or feathers) on the basis of a provided scale (less than 1%, 1 to 20%, 21 to 40%, 41 to 60%, 61 to 80%, and more than 80%), as well as whether any supplements were routinely administered, and, if so, brand names. Questions regarding the amount of food offered on a daily basis were included, as was the feeding schedule of the tiger (e.g. once daily or five days a week). Finally, respondents were asked to use the body condition scale based on the Felid TAG system (AZA, 2016) and assign a score to the tiger included in the survey (1=very thin, 2=underweight, 3=ideal, 4=overweight, and 5=obese). The survey was conducted in English, and no translation was provided.

Ethical approval for the study was granted by Nottingham Trent University's School of Animal, Rural and Environmental Sciences Ethics Review Group (ARE576).

Survey distribution

The survey was conducted by use of an online survey tool (Bristol Online Survey tool (https://www.onlinesurveys.ac.uk)), and the aim was to receive as many responses as possible over the study period (i.e. no *a priori* sample size calculation was performed) through convenience sampling (the use of an opportunistically available sample, as opposed to random sampling). Interested parties were provided with a PDF copy of the survey from the author, if preferred, to complete the survey by hand, or if they wished to assess what information would be needed prior to completing the survey.

For facilities located in North America, an invitation to participate in the survey via a weblink was distributed on behalf of the authors by the tiger SSP coordinator. To access information on the large population of tigers held by numerous small private collectors in North America (which the authors believed might have more restricted diet options than larger collections), a representative from one of the major suppliers of commercially-prepared carnivore diets was contacted, who then distributed the weblink to the survey on behalf of the authors, accompanied by a covering letter briefly explaining the purpose of the survey.

For survey distribution in Europe, the European Endangered Species Programme declined the invitation to participate due to other surveys taking place simultaneously, hence, only facilities not participating in that program were contacted directly by the authors. In the rest of the world, facilities that held tigers were identified using the International Tiger Studbook. Because no email addresses were provided in the studbook, each facility's website was then used to contact zoo personnel to invite participation. Completed surveys were accepted from May 18 until December 31, 2017, by which point no additional responses were included.

Animals

Respondents were asked to complete a survey for only one tiger, regardless of the number of tigers in their collection, to reduce response fatigue and eliminate potential bias introduced by larger collections being represented more than once. To qualify for inclusion, tigers were required to be over 12 months of age at the time of survey, and females were required to be in a non-reproductive stage (i.e. not pregnant or lactating). To ensure random selection within each facility, respondents with more than one tiger in their collection were asked to select a tiger with the longest house name or, in the event of a tie, the tiger with the most vowels in its name in order. This randomised approach, rather than selection of tigers with the most signs of GI disease, was chosen to allow independent identification of factors associated with outcome (Cockcroft and Holmes, 2003).

Statistical analysis

Survey responses were exported into a spreadsheet application (Microsoft Excel, Microsoft Corp, Redmond, WA, USA) and inspected for validity. Data were then imported into statistical software (Stata/IC, version 11.1, Stata Corp, College Station, TX, USA) for analysis. For tigers with a reported studbook number (n=29), age at the time of survey completion was calculated from the birth date. For three other tigers, age was obtained directly from the facility soon after survey completion. Age was then assessed for normality of distribution with the Shapiro-Wilk test and histogram creation and, because of the non-normal distribution, was reported as median (range).

Univariate analysis was performed to identify variables for potential inclusion in multivariate models; variables with *P*-values <0.20 were used in this manner (Dohoo *et al.*, 2003a). Specifically, logistic regression was performed to examine associations between variables hypothesised *a priori* to be risk or protective factors (i.e. various health, health care, facility, tiger, and dietary characteristics) and three dichotomous outcomes: current GI disease, vomiting in the previous six months, and diarrhoea in the previous six months (per the survey question and not the scoring system). Ordinal logistic regression was performed to examine associations between putative protective or risk factors and four ordinal outcomes: frequencies of vomiting and diarrhoea in the previous six months as well as occurrence of liquid faeces (faecal score of 5) and ideal faeces (soft with shape; faecal score of 3) in the previous four weeks. A manual selection process was used for multivariate model building, and Akaike information criterion values and estimated confidence intervals (CIs; i.e. stability of estimates) were used to identify the optimal multivariate model for each outcome variable. Country was included as a random effect in all multivariate models, regardless of its significance, in an attempt to control for environmental variables (e.g. climate or resources) that were otherwise unaccounted for. Because each tiger represented a different facility, no controlling for facility was required. Associations in the multivariate models were considered significant when the *P*-value was <0.05. Only the odds ratios (ORs) derived from multivariate (and not univariate) analyses were reported.

3. Results and discussion

Tigers and facilities

From July to December 2017, completed surveys representing 35 tigers were received from 32 facilities that housed captive tigers. Of these, 27 had been invited to participate by the SSP, and the remaining eight had been contacted directly by one of the authors. In situations where a facility provided details for more than one animal, one tiger was randomly selected (by die roll) from among the multiple tigers from the same facility (one tiger each) to avoid overrepresentation of certain facilities and in keeping with the original study plan. Consequently, 32 tigers from 32 facilities were included in the study. Countries of origin included the United States (n=25), Australia (n=2), New Zealand (n=2), Canada (n=1), South Africa (n=1), and South Korea (n=1). The reported total number of tigers housed at these facilities ranged from one to nine (median=3). Overall, the included tigers represented approximately 1.7% of all tigers enumerated in the Species360 Zoological Information Management Software (version 1.7; available at http://zims.Species360.org; accessed June 5, 2017) at the time of the study. The region with the greatest representation was Oceania, with data collected on 6.2% of all tigers and 26.7% of all facilities enumerated in that area. A large proportion of tigers were from North America as well, representing 5.8% of all tigers and 19.6% of all facilities there. Only 0.17% and 2.2% of all tigers enumerated in Asia and Africa, respectively, were represented, and there was no representation from South America or Europe. The small sample size, lack of random selection of facilities, inclusion of only one tiger per facility, and general lack of representation of tigers from regions other than Oceania and North America limited the generalisation of all descriptive findings to the global captive tiger population.

Twenty-one of the 32 (66%) tigers were male, and 11 (34%) were female. Subspecies was reported as Siberian or Amur (n=13 (41%)), Sumatran (*P. t. sumatrae*; n=9 (28%)), Malayan (n=8 (25%)), Bengal (*P. t. tigris*; n=1 (3%)), and hybrid (n=1 (3%)). Median age was 8.4 years (range 2.8 to 21.8 years). All included tigers were confirmed *via* studbook entries to have been born in captivity; therefore, tiger origin (captive or wild birth) could not be evaluated for associations with GI health or disease.

General health and health care

Twenty-four (75%) tigers had an ideal BCS (3/5), five (16%) were scored as underweight (BCS, 2/5), and three (9%) were scored as overweight (BCS, 4/5). At the time of the survey completion, two tigers had kidney disease and one had lameness. The tiger with kidney disease recently had a soft tissue sarcoma removed. Three tigers had received a diagnosis of dental disease (of any type) at some point in the past. None of the tigers had received a diagnosis of rabies, haemobartonellosis, canine distemper, or feline immunodeficiency virus, feline leukaemia virus, calicivirus, or herpesvirus infection at any time in the past.

Medications provided during the previous four weeks included dewormers (n=12), antimicrobials (six (five orally administered; one parenterally administered)), heartworm preventive (three), gastroprotectants (two), probiotics (two), corticosteroids (two), stool softener (one), analgesics (one), and blood pressure medication (one). The frequencies with which tigers received various types of veterinary examinations are summarised in Figure 1.

The most common type of veterinary care or health monitoring received was a weight check, with 69% of tigers weighed more than three times a year. A total of 82% of tigers underwent faecal parasite testing at least once a year.

Diet

Percentages and frequencies of feeding various dietary components are summarised in Table 1, 2 and 3. The most common diet type was commercially produced raw meat blends, with 69% of tigers fed this more than 60% of the time. In the authors' experience, such commercial blends typically contain other ingredients in addition to solely

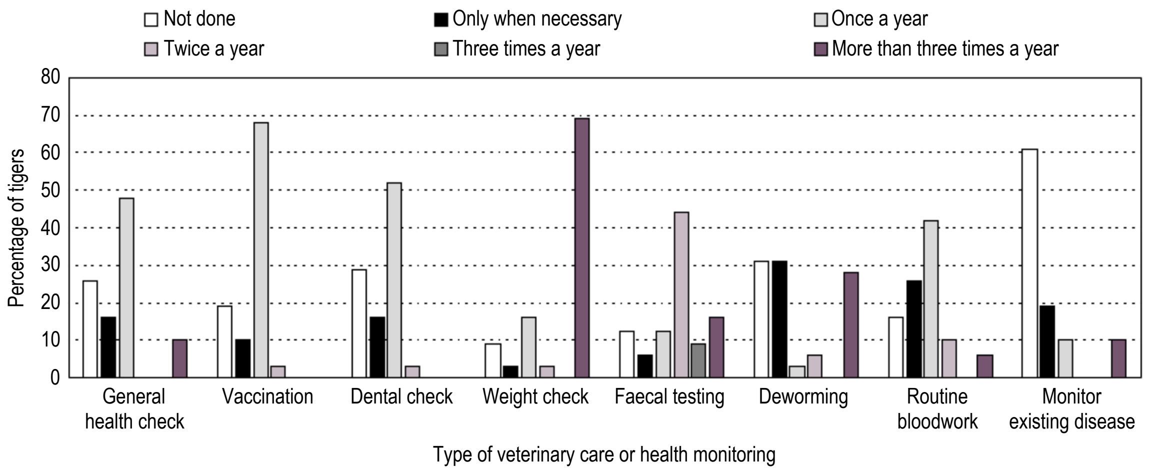

Figure 1. Frequency with which captive tigers (n=32) received various types of veterinary care or health monitoring in the previous year.

Table 1. Distribution of the frequency of feeding various diet types as a proportion of the total diet fed to captive tigers (n=32) in the previous month.[1]

Diet type	0%	1-20%	21-41%	41-60%	61-80%	81-100%
Whole-body prey or carcasses	3 (9)	21 (66)	1 (3)	2 (6)	2 (6)	3 (9)
Skeletal muscle diet (no bones)	4 (13)	21 (66)	3 (9)	0 (0)	1 (3)	3 (9)
Commercial raw meat blend	6 (19)	0 (0)	2 (6)	2 (6)	6 (19)	16 (50)
Commercial canned diet	30 (94)	2 (6)	0 (0)	0 (0)	0 (0)	0 (0)

[1] Data represent number (%) of tigers fed the indicated diet type at the indicated proportion of their diet. No tigers were fed commercial kibble (dry or pelleted/ extruded food).

Table 2. Distribution of the frequency (percentage of the time) of feeding various food sources to captive tigers (n=32) in the previous month.[1]

Food source	0%	1-20%	21-41%	41-60%	61-80%	81-100%
Deer	22 (69)	9 (28)	1 (3)	0 (0)	0 (0)	0 (0)
Beef	7 (22)	17 (53)	2 (6)	1 (3)	1 (3)	4 (13)
Pork	24 (75)	7 (22)	0 (0)	0 (0)	0 (0)	1 (3)
Horse	5 (16)	4 (13)	2 (6)	2 (6)	5 (16)	14 (44)
Donkey	30 (94)	1 (3)	0 (0)	0 (0)	0 (0)	1 (3)
Goat	26 (81)	6 (19)	0 (0)	0 (0)	0 (0)	0 (0)
Other ruminant hoofstock	31 (97)	1 (3)	0 (0)	0 (0)	0 (0)	0 (0)
Other nonruminant hoofstock	30 (94)	2 (6)	0 (0)	0 (0)	0 (0)	0 (0)
Chicken	11 (34)	15 (47)	3 (9)	1 (3)	2 (6)	0 (0)
Turkey	25 (78)	6 (19)	1 (3)	0 (0)	0 (0)	0 (0)
Other poultry	23 (72)	8 (25)	1 (3)	0 (0)	0 (0)	0 (0)
Rabbit	6 (19)	25 (78)	1 (3)	0 (0)	0 (0)	0 (0)
Level one offal	17 (53)	15 (47)	0 (0)	0 (0)	0 (0)	0 (0)
Level two offal	30 (94)	2 (6)	0 (0)	0 (0)	0 (0)	0 (0)
Non-animal-derived ingredient	31 (97)	1 (3)	0 (0)	0 (0)	0 (0)	0 (0)

[1] Data represent number (%) of tigers fed the indicated food source at the indicated frequency.

Table 3. Distribution of the frequency (percentage of the time) of feeding various diet ingredients to captive tigers (n=32) in the previous month.[1]

Ingredient	0%	1-20%	21-41%	41-60%	61-80%	81-100%
Hides or skins	12 (38)	15 (47)	0 (0)	1 (3)	0 (0)	4 (12)
Long bones (limbs)	7 (22)	19 (59)	1 (3)	2 (6)	1 (3)	2 (6)
Thoracic bones (ribs)	15 (47)	12 (38)	0 (0)	1 (3)	1 (3)	3 (9)
Skulls	17 (53)	14 (44)	0 (0)	0 (0)	0 (0)	1 (3)
Feet or wings	13 (41)	17 (53)	0 (0)	0 (0)	0 (0)	2 (6)
Muscle meat	1 (3)	13 (41)	1 (3)	1 (3)	4 (12)	12 (38)
Viscera	14 (44)	12 (38)	2 (6)	0 (0)	0 (0)	4 (12)
Fur or feathers	11 (34)	17 (53)	1 (3)	0 (0)	0 (0)	3 (9)

[1] Data represent number (%) of tigers fed the indicated ingredient at the indicated frequency.

skeletal muscle meat, including plant-based fibre sources, organ tissues, other animal-based proteins (dried egg), and soybean meal, as well as appropriate levels of supplemental minerals, vitamins, sometimes with added amino acids and/or fatty acid sources. Only 6% of tigers received at least a portion of their diet as commercial canned food, and no tiger was fed commercial kibble (i.e. dry or pelleted/extruded food).

The most common food source was horse, with 60% of tigers fed this over 60% of the time. By contrast, only 16% of tigers were fed beef over 60% of the time. The median total amount of food fed on a daily basis was 4 to 5 kg (range, <3 to >8 kg), and the most common frequency of feeding was more than once per day (minimum reported frequency, five days/week).

Currently, the AZA guidelines recommend the provision of a 'nutritionally adequate diet' comprising appropriately supplemented meat and/or prey ingredients (including commercially prepared carnivore diets as an option to achieve this). Although stated as 'nutritionally complete', according to domestic cat requirements, commercial formulations may vary according to their sources of meat-based protein sources and added dietary fibre sources (i.e. beet pulp as a soluble fermentable fibre vs cellulose as an insoluble, less fermentable fibre). These differences may influence the impact within the GI tract and related physiologic responses (Kerr *et al.*, 2013). To limit response fatigue and minimise the risk of erroneous or missing data, respondents did not have to name the specific types or components of commercial diets fed, although such data, including the fibre and protein sources, could have helped to better explain the nature of associations identified in the study (below).

Gastrointestinal health

Twenty-eight of 32 zoological facilities responded to the question, 'How many of the tigers in your collection have shown signs of vomiting in the past week? Please present as a fraction (e.g. 3/7 tigers).' On the basis of the provided information, the overall prevalence of vomiting among all tigers at these 28 facilities was 11% (10/93). Prevalence among tigers at individual facilities ranged from 0% (zero out of eight tigers) to 100% (three out of three tigers).

With respect to GI disease at the time of survey completion, for veterinarian-diagnosed problems, one tiger had inflammatory bowel disease and another had gastroenteritis, inflammatory bowel disease, and dietary hypersensitivity. One had non-veterinarian-diagnosed GI disease of unspecified type and was one of the two tigers that had received a gastroprotectant product administered during the previous four weeks. This prevalence (2/32 (6%) or 3/32 (9%), including the tiger with non-veterinarian-diagnosed disease) was lower than that reported for cheetahs in a previous study (13% (24/184); Whitehouse-Tedd *et al.*, 2015), and ideally the diagnoses would have been confirmed by requesting additional information from the facilities. However, the purpose of the study was not to document the nature of GI disease in tigers. Because respondents were asked to select one tiger in a manner designed to avoid selection bias, there was low risk of tigers being selected on health status (e.g. only the healthiest tiger selected) and the prevalence statistic of 9% for current GI disease was deemed reliable for the surveyed group. Because of the low number of tigers with current GI disease, and because one of these tigers was lacking data for several variables, no modelling was performed to identify factors associated with current GI disease.

In the six months prior to the survey, seven (22%) tigers were reported as having had vomiting at some point but no diarrhoea, five (16%) had diarrhoea (per the survey question and not the scoring system) but no vomiting, and six (19%) had both vomiting and diarrhoea. Frequencies of vomiting and diarrhoea during this period ranged from every few months (nine tigers for both) to every few days (one tiger for both). It was noteworthy that 18 (56%) tigers had vomiting or diarrhoea observed in the week preceding the survey.

Faecal scoring

Twenty-four (75%) tigers had ideal (soft with shape) faeces (faecal score of 3) 'often' to 'always' during the previous four weeks, as defined by the photographic examples and descriptions in the survey. Sixteen (50%) tigers had no liquid faeces (faecal score of 5) or extremely dry (faecal score of 1) faeces during the previous four weeks. Nine tigers (28%) had no extremely dry faeces but 'occasional' liquid faeces, one (3%) had extremely dry faeces 'often' but no liquid faeces. One tiger (3%) experienced a range of both extremely dry and liquid faeces 'occasionally,' and five (16%) had liquid faeces 'occasionally' but no extremely dry faeces.

Although these responses were likely subject to recall bias and, hence, should not be interpreted as prevalence estimates, there was no expectation that the degree of recall would meaningfully bias the results of statistical analyses because the specific hypotheses being explored were not revealed to respondents (Dohoo *et al.*, 2003b). Consequently, it was considered that the responses were valid as a crude indicator of GI health in the statistical models.

Variables associated with the frequency of vomiting in the previous six months

Results of univariate and multivariate analyses to identify factors associated with vomiting ('yes' or 'no' responses) in the previous six months were similar to those associated with the frequency of vomiting in the previous six months. Hence, only the results for the latter analyses were reported here. In the univariate analyses to predict frequency of vomiting in the previous six months, variables with increased odds included gastroprotectant treatment (P=0.007) and increased frequency of monitoring for existing disease (P=0.15), increased percentage of diet consisting of canned food (P=0.09), and increased feeding other (i.e. not deer, beef, or goat) ruminant hoofstock (P=0.04), chicken (P=0.08), non-animal-derived ingredients (e.g. vegetables or grain; P=0.10), and muscle meat (P=0.14) in the previous four weeks. Variables with a decreased chance included increased BCS (P=0.047), increased frequency of feeding horse meat (P=0.16) and long bones (P=0.12) in the previous four weeks, and providing a separate vitamin or mineral supplement of any type (P=0.08). The final multivariate model, which included only three of these variables, indicated that the frequency of vomiting increased with feeding muscle meat (OR, 2.10; 95% CI, 1.17 to 3.79; P=0.01) and chicken (OR, 12.79; 95% CI, 2.26 to 72.49; P=0.004) increased and decreased as the frequency of feeding long bones increased (OR, 0.11; 95% CI, 0.03 to 0.51; P=0.003).

The protective association that feeding carcass components, such as long bones, appeared to have against vomiting frequency is similar to the association identified in captive cheetahs (OR=0.36; 95% CI, 0.19 to 0.70; P=0.039) *via* multivariate analysis in a previous study (Whitehouse-Tedd *et al.*, 2015). Furthermore, feeding whole animals in smaller meals multiple times per day has been part of a successful treatment regimen for tigers (Seidel and Wisser, 1987). Although frequent vomiting has been linked with GI diseases and specifically 'tiger disease' (with proposed, yet unknown, aetiologies reported variously as hairballs, oral problems, pancreatic dysfunction, disruption of gut microbiome, stress, infectious agents, or kidney or liver failure (Bush *et al.*, 1987; Seidel and Wisser, 1987), this study serves as the first, albeit limited and preliminary, report of vomiting frequency in tigers. It remains to be determined whether vomiting frequency is correlated with feeding management, and underlying predisposing factors require further study. In addition, because of the cross-sectional survey nature of the study, no conclusions can be drawn regarding causation or which came first: vomiting frequency and the other outcomes of interest or the investigated variables. Therefore, these results should be interpreted with this in mind.

Variables associated with the frequency of diarrhoea in the previous six months

Results of univariate and multivariate analyses to identify factors associated with diarrhoea ('yes' or 'no' responses) in the previous six months were similar to those associated with the frequency of diarrhoea over the same time frame. Therefore, only the results for the frequency analyses were reported here. In the univariate analysis regarding frequency of diarrhoea in the previous six months, variables with an increased odds of this outcome included oral antimicrobial treatment (P=0.02) or gastroprotectant treatment (P=0.02) within the previous four weeks; increased frequency of routine health examination (P=0.11), deworming (P=0.19), and monitoring for existing disease (P=0.02); increased percentage of diet consisting of canned food (P=0.13); and increased frequency of feeding beef (P=0.08) and muscle meat (P=0.08) within the previous four weeks. Variables with a decreased odds included increased number of tigers at the facility (P=0.14); increased frequency of feeding horse meat (P=0.10), hides or skins (P=0.14), long bones (P=0.10), thoracic bones (P=0.07), skulls (P=0.11), and feet or wings (P=0.18) over the previous four weeks; and increased BCS (P=0.08). The final multivariate model, which included two of these variables, revealed that the frequency of diarrhoea within the previous six months increased with the frequency of feeding beef (OR, 3.39; 95% CI, 1.43 to 8.03; P=0.006) and muscle meat (OR, 3.08; 95% CI, 1.28 to 7.37; P=0.01).

The adverse association between muscle meat feeding and liquid faeces in tigers contrasted with previously reported epidemiological findings for captive cheetahs. This suggested that cheetahs fed muscle meat at least once a week are less likely than others to have chronic gastritis or non-specific GI disease (Whitehouse-Tedd *et al.*, 2015). Although muscle meat was one category of ingredients fed (Table 3), it was not specified as to which species this meat originated from. Responses regarding beef and muscle meat were not correlated (correlation coefficient, -0.09), indicating that the related questions were not interpreted by survey respondents as meaning the same thing, and so it appears that the muscle meat category likely included meat from other animal species, as well as cattle.

Variables associated with frequency of liquid faeces in the previous four weeks

Several variables were identified on univariate analysis for consideration in the multivariate model to predict frequency of liquid faeces (faecal score of 5) within the previous four weeks. Variables which increased the chance of liquid faeces included oral antimicrobial treatment (P=0.04) or any antimicrobial treatment (P=0.04) over the previous four weeks, increased frequency of routine health examination (P=0.10) and monitoring for existing disease (P=0.02), history of dental disease (P=0.09), increased frequency of feeding beef (P=0.04) in the previous four weeks, and higher total amount of food fed per day (P=0.18). Variables which decreased the chance of liquid faeces scores included increased frequency of feeding horse meat (P=0.15), hides or skins (P=0.10), or skulls (P=0.09) in the previous four weeks, as well as higher BCS (P=0.19). The final multivariate model included two of these variables. Specifically, the frequency of liquid faeces increased with oral antimicrobial treatment (OR, 15.62; 95% CI, 1.03 to 236.53; P=0.047) and increasing frequency of feeding beef (OR, 2.15; 95% CI, 1.08 to 4.30; P=0.03).

The observed association between oral antimicrobial treatment and liquid faeces was not surprising, given that diarrhoea is a reported adverse effect of several orally administered antimicrobials in many species, including felids (Albarellos and Landoni, 2009). However, owing to the cross-sectional nature of the study, it was unknown whether antimicrobial treatment preceded the diarrhoea or, perhaps, was prompted by it. The reason for antimicrobial treatment was not requested in the survey. Given that an increased frequency of feeding beef was associated with an increased chance of diarrhoea or liquid faeces in the three multivariate models (data not reported for one model), it appeared that this association was not spurious. Despite the small sample size, the 95% CIs on the OR estimates regarding beef were fairly narrow, again adding credence to this result. Moreover, these findings aligned with those of previous research into the effect of diet source in captive tigers, in which faecal consistency was poorer (and dry matter concurrently decreased) when fed beef- vs horse-based diets. This was potentially explained by the higher collagen content or difference in added plant-based fibre types in the commercial formulations (Vester *et al.*, 2010). Feeding muscle meat, which was not associated with the frequency of liquid faeces in the previous four weeks, was associated with diarrhoea in the previous six months in the two related multivariate models (data not reported for one model). Combined, the impact of beef (or muscle meat, regardless of species of origin) may have been attributable to the relative lack of animal fibre provided from this diet, which has previously been demonstrated to reduce faecal quality in captive cheetahs (Depauw *et al.*, 2011). Alternatively, as reported by Kerr *et al.* (2010), this meat source-based difference in faecal quality may reflect differences in plant-based fibre sources that were not quantified in this survey. It may be that the drivers of faecal consistency in tigers are distinct from those contributing to loose faeces (Depauw *et al.*, 2011) or GI disease (Whitehouse-Tedd *et al.*, 2015) in cheetahs, or that other (unmeasured) factors explained the observed GI signs in the study tigers. Further research is therefore warranted.

Variables associated with the frequency of ideal faeces

Variables with increased odds identified in the univariate analysis to predict frequency of ideal (soft with shape) faeces (faecal score of 3) included tigers being treated with dewormer (P=0.07) and steroid medication (P=0.17) in the previous four weeks, as well as increased frequency of vaccination (P=0.02) and dental examination (P=0.11). In addition, ideal faecal scores were associated with the increased frequency of feeding horse meat (P=0.048), poultry other than chicken or turkey (P=0.02), and level two offal (internal organs other than liver, kidney and heart; P=0.17), increased overall feeding frequency (P=0.02), and higher BCS (P=0.04). The only variable with a decreased chance in univariate analysis was increasing frequency of feeding other ruminant hoofstock in the previous four weeks (P=0.17). The final multivariate model, which included two of these variables, indicated that the frequency of ideal faecal scores increased with increasing higher feeding (OR, 3.45; 95% CI, 1.10 to 10.90; P=0.04) and vaccination frequency (OR, 9.26; 95% CI, 2.20 to 39.00; P=0.01).

The observed association between vaccination (with responses ranging from 'not done' to 'twice a year') and ideal faecal score occurrence over the previous four weeks was interesting. No information was requested about the nature of the vaccines; however, this variable could be related to the immune status of the tigers which may have had reduced susceptibility to infectious causes of GI disease. Alternatively, or concurrently, this may have caused more veterinary or keeper vigilance in animal health care either at these facilities in general or within these facilities for this time point. However, no other veterinary examination variables were associated with any outcome.

Other variables

No significant associations with any single evaluated outcome were identified in the multivariate analysis for several variables hypothesised *a priori* to have a protective (e.g. deworming practices, nutritional supplements, and probiotics) or adverse association (e.g. tiger subspecies, number of tigers in the collection, and percentage of diet consisting of certain types of food) with vomiting, diarrhoea or faecal consistency. However, such associations cannot be ruled out, owing to the small sample size.

4. Conclusions

The present study characterised the nutrition and management that a small number of captive tigers currently receive, and enabled the identification of several husbandry factors associated with GI variables in captive tigers, taking into account the country of residence. Overall health of the tigers appeared good, as suggested by the BCS and faecal scores for a large proportion (75%), a general lack of non-GI-related diagnoses, and a low prevalence of GI-related diagnoses.

Multivariate analyses revealed a number of factors associated with increased odds and frequencies of vomiting and diarrhoea in the previous six months and increased occurrence of liquid (faecal score of 5) or ideal (faecal score of 3) faeces in the previous four weeks, which may be relevant to the general captive tiger population. These findings suggested that beef and muscle meat feeding may be involved in undesirable GI-related signs in tigers, whereas the provision of skeletal components, such as long bones, may be beneficial, although the authors acknowledge that no causal relationship was established.

As the present study was limited in sample size and representation, these results must be considered preliminary and additional investigation is needed to explore these possibilities further. Such studies should include evaluation of the specific commercial diet blends fed, given the established association between dietary fibre and faecal quality in tigers. Moreover, these findings provide additional support for the need to consider species-specific dietary adaptations and for further investigation into the GI health impact of dietary provision in captive tigers.

A general limitation of the present study (and all such studies) is that information was collected through a survey and, hence, retrospectively rather than through direct examination of medical records and prospective monitoring. This approach was expected to result in some misclassification of the analysed variables, and readers are encouraged to consider this potential bias when interpreting the findings.

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Declaration of interest

The authors declare no conflict of interest.

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