Diet digestibility and palatability and intestinal fermentative products in dogs fed yeast extract

Gislaine Cristina Bill Kaelle, Camilla Mariane Menezes Souza, Taís Silvino Bastos, Ricardo Souza Vasconcellos, Simone Gisele de Oliveira and Ananda Portella Felix

Department of Animal Science, Federal University of Paraná, Curitiba, Brazil; Department of Animal Science, State University of Maringá, Maringá, Brazil

ABSTRACT
This study aimed to evaluate the effects of yeast extract on the apparent total tract digestibility (ATTD) of nutrients, metabolisable energy (ME), and palatability of diet, and on intestinal fermentative products in dogs. In addition, we also evaluated the ATTD and ME of yeast extract in dogs. Four diets containing 0, 40, 80, and 120 g/kg of yeast extract were evaluated. Diet digestibility, faecal characteristics, and intestinal fermentative products were evaluated in 12 adult Beagle dogs distributed in a randomised block design, with two periods of 10 days each. Yeast extract digestibility was obtained by regression analysis. Sixteen adult dogs were used for the palatability test to compare the diets: 0 vs. 40 g/kg yeast extract. The yeast extract presented ATTD of dry matter = 77.3%, crude protein (CP) = 55.7%, acid-hydrolysed ether extract (AHEE) = 73.8%, and ME = 4947.3 kcal/kg. There was a linear reduction in ATTD of CP and AHEE in the diets with yeast extract inclusion (p <.05). There was a linear increase in faecal concentration of total short-chain fatty acids (SCFA) and a linear reduction in faecal pH with the dietary inclusion of yeast extract (p <.05). Dogs preferred the 40 g/kg over the 0 g/kg yeast extract diet (p <.001).

HIGHLIGHTS
- The apparent digestibility of crude protein of the yeast extract was 55.7% in dogs;
- Yeast extract had beneficial effects on intestinal functionality in dogs;
- Palatability for dogs was higher for the diet with yeast extract.

INTRODUCTION

Yeast products from the sugar-alcohol or brewery industries can be used in dog and cat nutrition as a protein source or included as a functional ingredient, with potential benefits for intestinal functionality or diet palatability (Santos et al. 2018; Lin et al. 2019). Inactive yeast can be in the intact or autolysed form, in which the cell wall is broken (Borchani et al. 2014). After further centrifugation it is possible to obtain cell wall fractions, such as mannan oligosaccharides and β-glucans, as well as the extract.

Although some studies have evaluated the effects of yeast wall fractions in diets for dogs (Swanson et al. 2002; Theodoro et al. 2019; Lin et al. 2019), there is a lack of information about the effects of yeast extract. Yeast extract has approximately 34–48% of protein (in dry matter (DM)) and a relatively high concentration of essential amino acids, including the limiting amino acids lysine (3.61%), methionine (0.78%), and tryptophan (0.61%) (Wu et al. 2018). Thus, it can be classified as an alternative protein source of microbial origin and can contribute to meeting dogs’ nutritional requirements.

In addition to essential amino acids, yeast extract also has a high concentration of glutamic acid—approximately 5% in DM (Oliveira et al. 2011). Yeast extract has approximately 34–48% of protein (in dry matter (DM)) and a relatively high concentration of essential amino acids, including the limiting amino acids lysine (3.61%), methionine (0.78%), and tryptophan (0.61%) (Wu et al. 2018). Thus, it can be classified as an alternative protein source of microbial origin and can contribute to meeting dogs’ nutritional requirements.

Yeast extract has a high concentration of glutamic acid—approximately 5% in DM (Oliveira et al. 2011). Thus, due to the flavouring effect of glutamic acid, the inclusion of yeast extract can positively influence diet palatability (Tibbetts 2002; Costa 2004; Souza et al. 2011). Moreover, glutamic acid is involved in rapid cell
division, being an energy substrate for enterocytes and playing an important role in the integrity of the intestinal mucosa (Berres et al. 2010).

Other components present in yeast extract that can contribute to intestinal functionality are nucleotides—approximately 5.5% in DM (Fegan 2006; Souza et al. 2011). Nucleotides can contribute to tissue growth and recovery by participating in DNA synthesis (Lopes et al. 2011). Studies in piglets showed that the use of yeast extract in their diet contributed to the development of intestinal villi, immunomodulation, modulation of the intestinal microbiota, and reduction of diarrhoea (Waititu et al. 2017; Gao et al. 2021).

In addition to these components, yeast extract also has B vitamins, enzymes, short-chain fatty acids (SCFA), and chelated minerals (Butolo 2010). Thus, besides presenting an interesting nutritional profile, yeast extract can also be considered a functional ingredient. However, only one study evaluated the digestibility of brewer’s yeast in dogs (Martins et al. 2014), but none that evaluated specifically yeast extract digestibility in dogs was found. This study aimed to evaluate the effects of yeast extract on the apparent total tract digestibility (ATTD) of nutrients, metabolisable energy (ME), and palatability of the diet, and on intestinal fermentative products in dogs. We also aimed to evaluate the ATTD and ME of yeast extract in dogs.

**Material and methods**

The experiment was approved by the Animal Use Ethic Committee of the Agrarian Sciences Sector of the Federal University of Paraná, Curitiba, PR, Brazil, under the protocol number 054/2017.

**Digestibility assay**

**Animals and housing**

Six male and six female adult dogs (2 years old), weighing 10.3 ± 1.07 kg, were used. The body condition score (BCS) of the dogs was analysed at the beginning and at the end of the trial, on a scale from one to nine, according to Laflamme (1997). The dogs had a mean BCS of 5.2 ± 0.1. The animals underwent clinical and physical examination before and after the experimental period.

During most of the diet adaptation period, dogs had free access to an outdoor area of 1.137.84 m² under supervision for 4 h/day for voluntary exercise and socialisation with other experimental dogs and people. During the faecal collection period, the dogs were individually housed in covered masonry kennels (5 metres long × 2 metres wide) with a bed and access to water *ad libitum*. The facilities had side wall grates allowing visual and limited interaction with neighbouring dogs. The ambient temperature ranged from 15°C to 26°C with a 12 h light–dark cycle (light from 6 a.m. to 6 p.m.).

**Ingredient and experimental diets**

Four diets containing increasing concentrations of yeast extract (Nupro®, Alltech Brasil Ltda., Maringá, PR, Brazil) — 0, 40, 80, and 120 g/kg—were evaluated. Yeast extract was added at the expense of the 0 g/kg diet formulation. The 0 g/kg diet was formulated according to the nutritional recommendations of the Association of American Feed Control Officials (AAFCO 2016) for adult dogs.

The yeast extract used had a powdered form and consisted of dry brewer’s yeast extract (*Saccharomyces cerevisiae*). The ingredients and chemical composition of the diets are shown in Table 1, and the chemical composition of the yeast extract is shown in Table 2.

The ingredients were ground in a hammer mill equipped with a 1 mm sieve and extruded in a single screw extruder (Ferraz, E-130; Ribeirão Preto, SP, Brazil). After extrusion, diets were dried in a triple-deck drier (100–110°C), sprayed with poultry fat, and then cooled.

**Digestibility and metabolisable energy determination**

The digestibility assay followed the total faecal collection method described by the Association of American Feed Control Officials (AAFCO 2016). The diets were provided during a 5-day adaptation phase, followed by 5 days of total faecal collection per period. Each three dogs were fed with one of the experimental diets per period, totalling six repetitions/diet after the two periods.

Food was offered twice a day (8:30 a.m. and 4:00 p.m.) in sufficient amounts to meet the animal ME requirement according to the National Research Council (2006): ME (kcal/day) = 130 × Body weight (kg)0.75. Water was provided *ad libitum*.

During the faecal collection phase of each period, faeces were collected at least twice a day, weighed, identified by animal and period, and stored in a freezer at −20°C. At the end of each collection period, the faeces were thawed, homogenised separately, forming a composite sample from each animal, and dried in a forced ventilation oven at 55°C (320-SE, Fanem, São Paulo, Brazil) for 72 h. After drying, diets
and faeces were ground through a 1 mm screen in a hammer mill (Arthur H. Thomas Co., Philadelphia, PA, USA) for chemical analysis.

Faeces and diets were analysed for DM at 105°C for 12 h. Nitrogen (N, method 954.01) and then crude protein (CP) were calculated as N x 6.25, crude fibre (FB, method 962.10), acid-hydrolysed ether extract (AHEE, method 954.02), and ash (method 942.05), according to the Association of the Official Analytical Chemists (AOAC 1995). Organic matter (OM) was obtained as: 100 – % ash. Gross energy (GE) was determined by an isoperibol calorimeter (Parr Instrument Co., model 1261, Moline, IL, USA). Chemical analyses were conducted in duplicate and repeated when the results varied more than 5%.

Faecal characteristics and intestinal fermentative products
At the 10th day of each period, fresh faecal samples (within 15 min of defecation) were collected and analysed for DM content (DMf), pH, ammonia, SCFA, branched-chain fatty acids (BCFA), and sialic acid. The DMf was calculated as: (DM at 55°C × DM at 105°C)/100. Faecal score was also evaluated considering a 5-point scale: 1 = pasty, shapeless faeces; 2 = soft and unshaped faeces; 3 = soft, shaped, and moist faeces; 4 = well-formed and consistent faeces; 5 = well-formed, hard, and dry faeces (Carciofi et al. 2009).

Faecal output was calculated as g faeces/g DM intake/day.

Faecal pH was measured with a digital pH metre (331, Politeste Instrumentos de Teste Ltda, São Paulo, SP, Brazil) using 3.0 g of fresh faeces diluted in 30 mL of distilled water. The ammonia concentration was determined according to Brito et al. (2010).

For SCFA and BCFA analyses, 10 g of faecal sample was mixed with 30 mL of 16% formic acid. This solution was homogenised and stored at 4°C for 3 to 5 days. Before the analysis, these solutions were centrifuged at 5000 rpm (2K15, Sigma, Osterode am Hans, NI, Germany) for 15 min. After centrifugation, the supernatant was separated and subjected to further centrifugation. Each sample underwent three centrifugations and, at the end of the last one, part of the supernatant was transferred to an identified Eppendorff tube for subsequent freezing at −20°C.

Table 1. Ingredients and analysed chemical composition (g/kg in dry matter) of the experimental diets.

| Item                  | Dry matter | Crude protein | Acid-hydrolysed ether extract | Ash | Crude fibre | Calcium | Phosphorus | Non-protein nitrogen | Gross energy (kcal/kg) |
|-----------------------|------------|---------------|-------------------------------|-----|-------------|---------|------------|----------------------|------------------------|
| Yeast extract         | 0.0        | 0.4           | 0.4                           | 0.4 | 0.4         | 0.4     | 0.4         | 0.4                  | 0.4                    |
| Yeast extract 0.0     | 40.0       | 80.0          | 120.0                         | 120.0 | 120.0       | 120.0   | 120.0       | 120.0                | 120.0                  |

Table 2. Analysed chemical composition (g/kg, dry matter) of yeast extract.

| Item                  | Dry matter | Crude protein | Acid-hydrolysed ether extract | Ash | Crude fibre | Calcium | Phosphorus | Non-protein nitrogen | Gross energy (kcal/kg) |
|-----------------------|------------|---------------|-------------------------------|-----|-------------|---------|------------|----------------------|------------------------|
| Yeast extract         | 931.70     | 346.50        | 34.40                         | 75.30 | 7.60        | 2.10    | 10.80      | 37.70                | 5216.30                |

Histidine 8.30
Leucine 26.30
Isoleucine 16.10
Lysine 26.0
Methionine 5.70
Phenylalanine 15.30
Threonine 17.20
Tryptophan 4.20
Valine 19.0
Non-essential amino acids
Alanine 25.30
Aspartic acid 39.30
Cysteine 3.30
Glutamic acid 51.30
Glycine 21.10
Proline 17.80
Serine 20.90
Tyrosine 12.20
Arginine 23.50
Taurine 0.60
Nucleic acids 5.51
Manufacturer’s data.
Afterward, the samples were thawed and centrifuged again at 14,000 rpm for 15 min (Rotanta 460 Robotic, Hettich, Tuttingen, BW, Germany). Faecal SCFA and BCFA were analysed by gas chromatography (Shimadzu®, model GC2014, Kyoto, Honshu, Japan) using a 30-mm-long and 0.32-mm-wide glass column (Agilent Technologies, HP INNO wax-19091 N, Santa Clara, CA, USA). Nitrogen was used as the carrier gas with a flow rate of 3.18 mL/min. Working temperatures were 200°C at injection, 240°C at the column (at a 20°C/min rate), and 250°C at the flame ionisation detector.

For the sialic acid determination, faeces were lyophilised (Alpha 1–4 LO plus, Christ, Osterodeam Hans, Germany) and analysed according to Jourdian et al. (1971).

Calculation and statistics analysis
Based on the laboratory results, the ATTD and ME of diets were calculated according to the Association of American Feed Control Officials (AAFCO 2016):

\[
\text{ATTD} = \left( \frac{\text{g nutrient intake} - \text{g nutrient excretion}}{\text{g nutrient intake}} \right) \times 100.
\]

\[
\text{ME (kcal/g)} = \frac{\{\text{kcal/g GE intake} - \text{kcal/}
\text{g GE faecal excretion} - \{\text{g CP intake} - \text{g CP faecal excretion}\} \times (1.25 \text{kcal/g})\}}{\text{g feed intake}}.
\]

The ATTD of nutrients and ME of yeast extract were calculated according to Adeola (2001). The concentration of digestible nutrients (protein and fat) and digestible energy of yeast extract was calculated as: % nutrient or kcal of GE × ATTD of the nutrient or energy.

Data were analysed for normality using the Shapiro–Wilk test. When data and residues assumed normal distribution, they were analysed using the PROC MIXED of SAS statistical package (version 8, SAS Institute Inc., Cary, NC, USA), considering the effects of diets, blocks (periods), and sex. Effects of periods and sex were not observed; data were then submitted to regression analysis using the PROC REG of SAS statistical package (version 8, SAS Institute Inc., Cary, NC, USA), with six repetitions per treatment. Data that did not show normal distribution were analysed using the Kruskal Wallis test at 5% probability (SAS Institute Inc 2011).

Palatability assay

Animals and experimental procedures
Sixteen adult Beagle dogs (eight males and eight females), with 2 years of age, weighing 10.3 ± 1.07 kg, were used to determine yeast extract palatability. The animals were kept under the same conditions previously described.

The palatability test was conducted for two days and compared the diets 0 g/kg vs. 40 g/kg yeast extract. Once a day (8:00 a.m.) each dog received two bowls containing the diets to be compared. Each diet was offered in an amount 30% higher than the ME requirements recommended by the National Research Council (NRC 2006) for adult laboratory dogs. As soon as one of the diets was completely consumed, both bowls were removed, and the leftovers were quantified. On the second day, the position of feeders was changed to avoid bias.

The first bowl approached by the dog was computed as the first choice, whether the dog ate from it or not. We also calculated the intake ratio of each diet by the following equation:

\[
\text{Intake ratio} = \frac{\text{g ingested of the diet A or B}}{\text{total g consumed (A + B)}}
\]

Statistics analysis

The experiment followed a completely randomised design, totalling 32 repetitions (16 dogs × 2 days). The first-choice data were analysed by the chi-square test and the intake ratio by the paired Student’s t test, both using a significance level of 5%.

Results

Digestibility assay

All dogs remained healthy throughout the study. No episodes of food refusal, weight loss, vomiting, or diarrhea were observed. There were no differences (p = .98) in DM intake among the diets (mean = 173.8 ± 9.46 g/dog/day).

The ATTD of CP and AHEE declined linearly as the level of yeast extract in the diet increased (p < .05). ATTD of other nutrients was not different among the dietary treatments (p > .05, Table 3). The results of ATTD, ME, and digestible nutrient content of the yeast extract are shown in Table 4.

Faecal pH and isovalerate linearly decreased with higher levels of yeast extract in the diet. Faecal
propionate and total SCFA concentrations rose linearly with the dietary inclusion of yeast extract ($p < 0.05$, Table 5). Faecal concentration of butyrate showed a quadratic rise with the dietary inclusion of yeast extract ($p < 0.05$, Table 5). The other faecal variables did not differ among the treatments ($p > 0.05$).

Table 3. Means of apparent total tract digestibility (ATTD, %) and metabolisable energy (ME, kcal/kg) of diets containing increasing concentrations of yeast extract.

| Item                  | Yeast extract (g/kg) | p Value* |
|----------------------|----------------------|----------|
|                      | 0        | 40       | 80       | 120      | SEM | L | Q |
| ATTD                 |          |          |          |          |     |   |   |
| Dry matter           | 79.41    | 79.83    | 80.80    | 79.16    | 0.39 | 0.999 | 0.827 |
| Organic matter       | 84.52    | 84.95    | 84.66    | 84.20    | 0.24 | 0.827 | 0.697 |
| Crude protein        | 83.70    | 82.29    | 80.80    | 79.51    | 0.51 | <0.001 | 0.982 |
| Ether extract        | 91.47    | 91.50    | 90.10    | 89.72    | 0.20 | <0.001 | 0.367 |
| Gross energy         | 93.73    | 94.05    | 94.34    | 93.88    | 0.18 | 0.974 | 0.736 |
| Organic matter       | 84.52    | 84.95    | 84.66    | 84.20    | 0.24 | 0.827 | 0.697 |
| ME                   | 4100.24  | 4109.40  | 4202.73  | 4194.48  | 18.60 | 0.121 | 0.995 |

SEM: standard error of the mean.
*Probabilities for linear (L) and Quadratic (Q) effects; Mean ATTD Digestibility of Crude Protein (%): 83.635 $\pm$ 0.333 $\times$ % yeast inclusion ($R^2$=0.657); Mean ATTD Digestibility of Crude Protein of Ether Extract: 91.720 $\pm$ 0.171 $\times$ % yeast inclusion ($R^2$=0.813).

Table 4. Means of apparent total tract digestibility (ATTD, %), metabolisable energy (ME), and digestible nutrients of yeast extract in dogs.

| Item                  | ATTD      | Digestible nutrient (%) |
|----------------------|-----------|-------------------------|
| Dry matter           | 77.30     | –                       |
| Organic matter       | 81.72     | –                       |
| Crude protein        | 55.70     | 19.30                   |
| Acid-hydrolysed ether extract | 73.80 | 2.54                    |
| Gross energy (kcal/kg) | 94.24    | 4913.70                 |
| ME (kcal/kg)         | 4947.29   | –                       |

Table 5. Means of faecal characteristics and intestinal fermentative products of dogs fed with diets containing yeast extract.

| Item                  | Yeast extract (g/kg) | p       |
|----------------------|----------------------|---------|
|                      | 0        | 40       | 80       | 120      | SEM | L | Q |
| Dry matter (%)       | 34.06    | 33.36    | 33.19    | 31.82    | 0.39 | 0.297 | 0.978 |
| pH                   | 6.71     | 6.72     | 6.55     | 6.35     | 0.06 | 0.021 | 0.351 |
| Faecal output*       | 0.61     | 0.61     | 0.58     | 0.64     | 0.02 | 0.954 | 0.781 |
| Ammonia (g/kg)       | 0.43     | 0.53     | 0.54     | 0.46     | 0.10 | 0.963 | 0.074 |
| Sialic acid (mmol/g) | 1.47     | 1.47     | 1.33     | 1.47     | 0.06 | 0.607 | 0.197 |
| Scoreb               | 4        | 4        | 4        | 4        | –    | –    | –    |
| Short-chain fatty acids (SCFA, μmol/g) |          |          |          |          |     |   |   |
| Acetate              | 248.78   | 304.66   | 300.33   | 300.62   | 1.13 | 0.152 | 0.251 |
| Propionate           | 146.70   | 178.76   | 176.62   | 184.41   | 0.477| 0.005 | 0.142 |
| Butyrate             | 35.46    | 43.96    | 43.75    | 43.13    | 0.114| 0.013 | 0.019 |
| Valerate             | 5.73     | 5.56     | 4.81     | 4.27     | 0.136| 0.564 | 0.838 |
| Total SCFA           | 436.67   | 532.94   | 525.51   | 532.43   | 1.555| 0.037 | 0.141 |
| Branched-chain fatty acids (BCFA, μmol/g) |          |          |          |          |     |   |   |
| Isovalerate          | 10.35    | 10.74    | 9.92     | 5.69     | 0.094| 0.030 | 0.105 |
| Isobutyrate          | 6.54     | 6.26     | 6.74     | 5.76     | 0.021| 0.362 | 0.373 |
| Total BCFA           | 16.89    | 17.00    | 16.66    | 11.45    | 0.212| 0.168 | 0.373 |

SEM: standard error of the mean; L: linear; Q: quadratic.
*Faecal output $=$ faeces produced as-is/g dry matter consumed/day.
Median faecal consistency score analysed by Kruskal–Wallis ($p > 0.05$); pH $= 6.770 \pm 0.031 \times %$ yeast inclusion ($R^2$=0.86); propionate (μmol/g) = 156.01 $\pm$ 2.75x ($R^2$=0.779); butyrate (μmol/g) = 36.05 $\pm$ 2.32x $\times 0.14$x ($R^2$=0.931); isovalerate (μmol/g) = 12.53 $\pm$ 0.45x ($R^2$=0.662); total SCFA (μmol/g) = 464.91 $\pm$ 6.99x ($R^2$=0.592).

**Palatability**

The intake ratio was greater for the diet containing 40 g/kg of yeast extract when compared to the 0 g/kg diet ($p < 0.001$). However, there was no difference ($p > 0.05$) for the first choice (Table 6).

**Discussion**

In the present study we calculated the ATTD of yeast extract in dogs, which presented relatively low CP ATTD (55.7%) when compared to common vegetable protein sources used in extruded diets for dogs (69–88%, Kawauchi et al. 2011; Félix et al. 2013). In pigs, yeast extract presented a greater apparent CP ileal digestibility (81.3%, Wu et al. 2018), which may be explained in part due to the measure of the digestibility in the ileum and not in the total tract. We did not find any study that evaluated specifically the digestibility of yeast extract in dogs; however, a study evaluating the digestibility of brewer’s and sugarcane yeast observed greater ATTD of CP values, which varied from 88.8% (integral brewer’s yeast) to 63% (autolyzed sugarcane yeast) in dogs (Martins et al. 2014).

This relatively low value of the ATTD of CP may be explained by two main possibilities: (1) the high concentration of nucleotides (about 5%) in the yeast extract may have accounted for the total nitrogen present in the faeces, interfering in the results; (2) the limitation of the regression method for estimating the digestibility of ingredients. The extrapolation of the ingredient dietary inclusion to 1000 g/kg can increase the errors in determining its digestibility, since it extrapolates the concentrations evaluated and may not represent exactly what would happen in the animal’s gastrointestinal system (Kawauchi et al. 2011). This method assumes that all numerical variation in the digestibility of the diet occurred as a function of the ingredient’s inclusion concentration. The same can be explained for the AHEE result.

These hypotheses are supported by the results of faecal ammonia, BCFA, and pH. The inclusion of autolysed yeast extract in the present study did not increase the faecal concentration of ammonia and BCFA but reduced the faecal concentration of isovaleric acid and pH. These results demonstrate that

Table 6. Number of visits to the food bowl (first choice) and intake ratio of diets.

|                  | Diets (g/kg yeast extract) | p       |
|------------------|---------------------------|---------|
| First choice     | 17                        | 15      | .892  |
| Intake ratio     | 0.39                      | 0.61    | <.001 |
increasing the dietary concentration of yeast extract probably did not affect the indigestible protein content in the colon.

Fermentation of undigested proteins in the colon by microbiota results in the formation of BCFA (formed from branched-chain amino acids), ammonia, indole and phenolic compounds, biogenic amines, hydrogen sulphide, and nitric oxide (Windey et al. 2012; Ápajalhti and Viinola 2016). These protein fermentation metabolites have been associated with possible impairment of intestinal functionality and worsening of faecal odour in dogs (Bastos et al. 2020).

Isovaleric and isobutyric are the main BCFA originated from nitrogen compounds fermentation (Rios-Covian et al. 2015). A study evidenced that humans consuming a high-protein diet had higher faecal concentrations of isovaleric and isobutyric BCFA when compared to humans consuming a lower protein diet (Russell et al. 2011). In dogs, increases in faecal pH and metabolites such as BCFA and indoles were observed in animals fed with high protein diets (Hang et al. 2013; Herstad et al. 2017; Jackson and Jewell 2019). Faecal pH is also considered an indicator of the fermentative activity of the intestinal microbiota, with the highest pH corresponding to the proteolytic metabolism (Ephraim et al. 2020).

We also observed higher faecal concentrations of total SCFA and of propionic and butyric acids with the inclusion of yeast extract, which may be indicative of positive effects on intestinal functionality (Zentek et al. 2002; Swanson et al. 2002; Pinna et al. 2017; Félix et al., 2022). SCFA produced from microbial fermentative activity in the intestine are essential for intestinal homeostasis (Tramontano et al. 2018). SCFA act as secondary messengers that regulate gene expression and stimulate hormone and gut peptide synthesis and initiate other signal transduction pathways in peripheral tissues (Scholz-Ahrens et al. 2007; Koh et al. 2016; Alexander et al. 2019). Butyric acid has been recognised as having an important role in reducing inflammation and improving the epithelial barrier (Hamer et al. 2008; Huang et al. 2015). Besides, butyrate is the main energy source for colonocytes (Markowiak-Kopec and Slizewska 2020). The role of propionate in gut functionality has been studied less often than that of butyrate, but an in vitro study observed that propionate has an anti-inflammatory effect on the gut by inhibiting an accessory protein (CD14) of the toll-like receptor 4 (Hoyles et al. 2018). Besides, a recent meta-analysis (Félix et al. 2022) described that the faecal concentration of propionate is reduced in dogs with gastrointestinal diseases, indicating that it is a biomarker of gut functionality in dogs.

SCFA are also important to reduce the intestinal pH (Pieper et al. 2014; Chen et al. 2015). This reduction helps to improve gut functionality by inhibiting the proliferation of some potentially pathogenic bacteria (Bovera et al. 2010; Zentek et al. 2013; Tramontano et al. 2018; Alexander et al. 2019). Thus, faecal pH can be correlated with SCFA production (Do et al. 2021).

No studies evaluating the relationship of nucleotides from yeast extract on SCFA production and microbial populations in dogs were found. However, in studies with broiler chickens and piglets supplemented with nucleotides from yeast extract, increases in bacterial diversity and on SCFA concentrations were observed (Waititu et al. 2017; Leung et al. 2019). The benefits in these studies have been attributed to the increase in intestinal villi and villus: crypt ratio and immunomodulatory effect of yeast extract on the production of inflammatory cytokines in the intestine (Waititu et al. 2017; Leung et al. 2019; Gao et al. 2021). This is probably due to the action of yeast extract nucleotides on mitotic cell division in the intestine, as well as to glutamic acid, which is an important energy source for intestinal cell metabolism (Lacey and Wilmore 1990; Fegan 2006). Thus, the supplementation of yeast extract in the diet may contribute to the gut functionality and integrity of dogs, and it is important that new studies evaluating the microbiota, immune response, and intestinal permeability be carried out.

Regarding faecal consistency, the maximum inclusion of 12% yeast extract did not affect faecal DM and score, which remained at 4, and is considered within the normal range (Félix et al. 2009).

The lack of change in the concentration of faecal sialic acid indicates that yeast extract did not have an adverse effect on the intestinal mucosa. As an important component of mucin, the concentration of sialic acid tends to be greater when there are bacterial infections or osmotic fragility in the intestine (Pirgozliev et al. 2008). Thus, it is possible to infer that the inclusion of autolysed yeast extract did not alter the mucin production in the dog gut.

Yeast products have been used as palatability enhancers in cat and dog food industries for many years (Swanson and Fahey 2006). The results found in the palatability test are similar to the ones Martins et al. (2014) have found: a preference for food with 7.5% inclusion of brewer’s yeast or sugarcane autolysed yeast, with no difference between these two presentation forms. Even lower inclusions of yeast
such as 0.2% (Lin et al. 2019) and 0.5% (Oliveira et al. 2016) showed an increase in the palatability of diets for dogs and cats, respectively. In all studies, the authors argue that this preference can be attributed to the presence of glutamic acid in yeast extract, which sensitises umami receptors and makes the diet more palatable, as it has a flavor-enhancing effect that stimulates food intake. Furthermore, during the manufacturing process, glutamic acid reacts with sodium to form monosodium glutamate, which provides a great potential to favour the palatability of manufactured food. Likewise, the nucleotides present in this ingredient also enhance the effects of glutamic acid (European Association for Specially Yeast Products (EURASYP 2006).

Conclusion

The dietary inclusion of up to 120 g/kg of yeast extract did not change most ATTD of the nutritional fractions and ME of the diet, except for CP and AHEE. On the other hand, this study indicated beneficial effects on intestinal functionality, through higher faecal concentrations of total SCFA, propionate, and butyrate and lower faecal concentration of isovalerate and pH. Finally, the inclusion of 40 g/kg of yeast extract resulted in a more palatable diet. Considering those results, the dietary inclusion of 40 g/kg of yeast extract can be used in extruded diets for dogs to improve diet palatability and the faecal concentration of propionate and butyrate.

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Disclosure statement

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ORCID

Gislaine Cristina Bill Kaelle http://orcid.org/0000-0003-0171-741X
Camilla Mariane Menezes Souza http://orcid.org/0000-0003-2095-5835
Tais Silvino Bastos http://orcid.org/0000-0002-7741-715X
Ricardo Souza Vasconcellos http://orcid.org/0000-0002-0333-4300
Simone Gisele de Oliveira http://orcid.org/0000-0002-2913-1173
Ananda Portella Félix http://orcid.org/0000-0002-8570-5725

Data availability statement

The data of this study are available from the corresponding author, upon reasonable request.

References

Association of American Feed Control Officials (AAFCO). 2016. Official publications of the Association of American Feed Control Officials incorporated. Oxford (IN): Official Publication.

Association of Official Analytical Chemists (AOAC). 1995. Official methods of analysis. 16th ed. Arlington (VA): AOAC International.

Adeola O. 2001. Digestion and balance techniques in pigs. In: Lewis AJ, Southern LL, editors. Swine nutrition. 2nd ed. Washington, DC: CRC Press; p. 903–916.

Alexander C, Swanson KS, Fahey GC, Garleb KA. 2019. Perspective: physiologic importance of short-chain fatty acids from nondigestible carbohydrate fermentation. Adv Nutr. 10(4):576–589.

Apajalahti J, Vienola K. 2016. Interaction between chicken intestinal microbiota and protein digestion. Anim Feed Sci Technol. 221:323–330.

Bastos TS, de Lima DC, Souza CMM, Maiorka A, Oliveira SG, Bittencourt LC, Félix AP. 2020. Bacillus subtilis and Bacillus licheniformis reduce faecal protein catabolites concentration and odour in dogs. BMC Vet Res. 16(1):116.

Berres J, Vieira SL, Kidd MT, Taschetto D, Freitas DM, Barros R, Nogueira ET. 2010. Supplementing L-valine and L-isoleucine in low-protein corn and soybean meal all-vegetable diets for broilers. J Appl Poult. 19(4):373–379.

Borchani C, Fonteyn F, Jamin G, Paquot M, Blecker C, Thonart P. 2014. Enzymatic process for the fractionation of baker’s yeast cell wall (Saccharomyces cerevisiae). Food Chem. 163:108–113.

Bovera F, Marono S, Di Meo C, Piccolo G, Iannaccone F, Nizza A. 2010. Effect of mannanoligosaccharides supplementation on caecal microbial activity of rabbits. Animal. 4(9):1522–1527.

Brito CBM, Félix AP, Jesus RM, França MI, de Oliveira SG, Krabbe EL, Maiorka A. 2010. Digestibility and palatability of dog foods containing different moisture levels, and the inclusion of a mould inhibitor. Anim Feed Sci Technol. 159(3–4):150–155.
Butolo JE. 2010. Qualidade de ingredientes na alimentação animal [Quality of ingredients in animal feed]. Campinas, São Paulo: CBNA.

Carciofi AC, Oliveira L, Valério A, Borges LL, Carvalho F, Brunetto MA, Vasconcellos RS. 2009. Comparison of micronized whole soybeans to common protein sources in dry dog and cat diets. Anim Feed Sci Technol. 151(3–4): 251–260.

Chen H, Wang W, Degroote J, Possemiers S, Chen D, De Smet S, Michiels J. 2015. Arabinoxylan in wheat is more responsible than cellulose for promoting intestinal barrier function in weaned male piglets. J Nutr. 145(1):51–58.

Costa LF. 2004. Leveduras na nutrição animal [Yeast in animal nutrition]. NRE. 1:1–6.

Do S, Phungvivatnikul T, Godoy MRC, Swanson KS. 2021. Nutrient digestibility and fecal characteristics, microbiota, and metabolites in dogs fed human-grade foods. J Anim Sci. 99:1–13.

Ephraim E, Cochrane CY, Jewell D. 2020. Varying protein levels influence metabolomics and the gut microbiome in healthy adult dogs. Toxins. 12(8):517.

European Association for Specially Yeast Products (EURASYP). 2006. Yeast products: yeast cell wall. Paris: EURASYP.

Fegan F. 2006. Functional Foods for aquaculture: benefits of NuPro® and dietary nucleotides in aquaculture feeds. Bangkok: Alltech. [accessed 2017 May 13]. http://www.aquafeed.com/docs/papers/Fegan.pdf.

Félix AP, Zanatta CP, Brito CBM, Murakami FY, França MI, Maiorka A, Fleming JS. 2009. Suplementação de mananolígosacarídeos (MOS) e uma mistura de aluminossilícios na qualidade das fezes de cães adultos [Mannanoligosaccharide (MOS) supplementation and a mixture of aluminosilicates in the quality of faeces of adult dogs]. Arch Vet Sci. 14:31–35.

Félix AP, Zanatta CP, Brito CBM, Sá Fortes CML, Oliveira SG, Maiorka A. 2013. Digestibilidade e metabolizável energia de raw soybeans manufactured with different processing treatments and fed to adult dogs and puppies. J Anim Sci. 91(6):2794–2801.

Félix AP, Souza CMM, Oliveira SG. 2022. Biomarkers of gastrointestinal functionality in dogs: a systematic review and meta-analysis. Anim Feed Sci Technol. 283:115183.

Gao L, Xie C, Liang X, Li Z, Li B, Wu X, Yin Y. 2021. Yeast-based nucleotide supplementation in mother sows modifies the intestinal barrier function and immune response of neonatal pigs. Anim Nutr. 7(1):84–93.

Hang I, Heilmann RM, Grutzner N, Suchodolski JS, Steiner JM, Atrosi F, Sankari S, Kettunen A, De Vos WM, Zentek J, et al. 2013. Impact of diets with a high content of greaves-meal protein or carbohydrates on faecal characteristics, volatile fatty acids and faecal calprotectin concentrations in healthy dogs. BMC Vet Res. 9:201.

Hamr HM, Jonkers D, Venema K, Vanhoutvin S, Troost FJ, Brummer RJ. 2008. Review article: the role of butyrate on colonic function. Aliment Pharmacol Ther. 27(2):104–119.

Herstad KMV, Gajardo K, Bakke AM, Moe L, Ludvigsen J, Rudi K, Rud I, Sekelja M, Skancke E. 2017. A diet change from dry food to beef induces reversible changes on the faecal microbiota in healthy, adult client-owned dogs. BMC Vet Res. 13(1):147.

Hoyles L, Snelling T, Umlai UK, Nicholson JK, Carding SR, Glen RC, McArthur S. 2018. Microbiome–host systems interactions: protective effects of propionate upon the blood–brain barrier. Microbiome. 6(1):13.

Huang C, Song P, Fan P, Hou C, Thacker P, Ma X. 2015. Dietary sodium butyrate decreases postweaning diarrhea by modulating intestinal permeability and changing the bacterial communities in weaned piglets. J Nutr. 145(12): 2774–2780.

Jackson MI, Jewell DE. 2019. Balance of saccharolysis and proteolysis underpins improvements in stool quality induced by adding a fiber bundle containing bound polyphenols to either hydrolyzed meat or grain-rich foods. Gut Microbes. 10(3):298–320.

Jourdian GW, Dean L, Roseman S. 1971. A periodate-resorcin method for the quantitative estimation of free sialic acids and their glycosides. J Biol Chem. 246(2):430–435.

Kawauchi IM, Sakomura NK, Vasconcellos RS, De-Oliveira LD, Gomes MOS, Loureiro BA, Carciofi AC. 2011. Digestibility and metabolizable energy of maize gluten feed for dogs as measured by two different techniques. Anim Feed Sci Technol. 169(1–2):96–103.

Koh A, De Vadder F, Kovatcheva-Datchary P, Bäckhed F. 2016. From dietary fiber to host physiology: short-chain fatty acids as key bacterial metabolites. Cell. 165(6): 1332–1345.

Lacey JM, Wilmore DW. 1990. Is glutamine a conditionally essential amino acid?. Nutr Rev. 48(8):297–309. doi: 10.1111/j.1753-4887.1990.tb02067.x.2080048

Laflamme D. 1997. Development and validation of a body condition score system for dogs. Canine Pract. 22:10–15.

Leung H, Ytibarek A, Snyder R, Patterson R, Barta JR, Karrow N, Kiarie E. 2019. Responses of broiler chickens to Eimeria challenge when fed a nucleotide-rich yeast extract. Poult Sci J. 98(4):1622–1633.

Lin C-Y, Alexander C, Steelman AJ, Warzeca CM, de Godoy MRC, Swanson KS. 2019. Effects of a Saccharomyces cerevisiae fermentation product on fecal characteristics, nutrient digestibility, fecal fermentative end-products, fecal microbial populations, immune function, and diet palatability in adult dogs. J Anim Sci. 97(4):1586–1599.

Lopes CC, Rabello CBV, Silva VA, Jr, Holanda MCR, Arruda EMF, Silva JCR. 2011. Desempenho, digestibilidade, composição corporal e morfologia intestinal de pintos de corte recebendo dietas contendo levedura de cana-de-açúcar [Performance, digestibility, body composition, and intestinal morphology of broiler chicks fed diets containing sugarcane yeast]. Acta Sci Anim Sci. 33:33–40.

Markowiak-Kopiec P, Sliżewska K. 2020. The effect of probiotics on the production of short-chain fatty acids by human intestinal microbiome. Nutrients. 12(4):1107.

Martins MS, Sakomura NK, Souza DF, Filho FOR, Gomes MOS, Vasconcellos RS, Carciofi AC. 2014. Brewer’s yeast and sugarcane yeast as protein sources for dogs. J Anim Physiol Anim Nutr. 98(5):948–957.

National Research Council (NRC). 2006. Nutrient requirements of dogs and cats. Washington: National Academies Press.

Oliveira R T d, Haese D, Kill JL, Lima A, Malini PV, Thompson GR. 2016. Palatability of cat food with sodium pyrophosphate and yeast extract. Cienc Rural. 46(12):2202–2205.
Pieper R, Boudry C, Bindelle J, Vahjen W, Zentek J. 2014. Interaction between dietary protein content and the source of carbohydrates along the gastrointestinal tract of weaned piglets. Arch Anim Nutr. 68(4):263–280.

Pinna C, Vecchiato CG, Cardenia V, Rodriguez-Estrada MT, Stefaneli C, Grandi M, Gatta PP, Biagi G. 2017. An in vitro evaluation of the effects of a Yucca schidigera extract and chestnut tannins on composition and metabolic profiles of canine and feline faecal microbiota. Arch Anim Nutr. 71(5):395–412.

Pirgozliev V, Oduguwa O, Acamovic T, Bedford MR. 2008. Effects of dietary phytase on performance and nutrient metabolism in chickens. Br Poult Sci. 49(2):144–154.

Rios-Covian D, Gueimond M, Duncan SH, Flint HJ, Reyes-Gavilan AG. 2015. Enhanced butyrate formation by cross-feeding between Faecalibacterium prausnitzii and Bifidobacterium adolescentes. FEMS Microbiol Lett. 362:1–7.

Russell WR, Gratz SW, Duncan SH, Holtrop G, Ince J, Scobie L, Duncan G, Johnstone AM, Lobley GE, Wallace RJ, et al. 2011. High-protein, reduced-carbohydrate weight-loss diets promote metabolite profiles likely to be detrimental to colonic health. Am J Clin Nutr. 93(5):1062–1072.

Santos JPF, Aquino AA, Gloria M, Avila-Campos MJ, Oba PM, Santos KM, Vendramini THA, Carciofi AC, Junior AR, Brunetto MA. 2018. Effects of dietary yeast cell wall on faecal bacteria and fermentation products in adult cats. J Anim Physiol Anim Nutr. 102(4):1091–1101.

SAS Institute Inc. 2011. SAS/Stat® 9.3 user’s guide. Cary (NC): SAS Institute Inc.

Scholz-Ahrens KE, Ade P, Marten B, Weber P, Timm W, Açıl Y, Glüer C-C, Schrezenmeir J. 2007. Prebiotics, probiotics, and symbiotics affect mineral absorption, bone mineral content, and bone structure. J Nutr. 137(Suppl 2): 8385–8465.

Souza RB, Costa FGP, Lima MR, Pinheiro SG. 2011. Utilização de cana-de-açúcar (Saccharomyces cerevisiae) nas ração de aves [Use of sugar cane (Saccharomyces cerevisiae) in poultry feed]. NRE. 8:1632–1646.

Swanson KS, Grieshop CM, Flickinger EA, Bauer LL, Healy HP, Dawson KA, Merchen NR, Fahey GC. Jr. 2002. Supplemental fructooligosaccharides and mannanoligosaccharides influence immune function, ileal and total tract nutrients digestibilities, microbial populations and concentrations of protein catabolites in the large bowel of dogs. J Nutr. 132(5):980–989.

Swanson KS, Fahey GC. Jr. 2006. Potential role of yeast and yeast by-products foods. Recent advances in pet nutrition. In: Laue DK, Tucker LA, editors. Recent advances in pet nutrition. Nottingham: Nottingham University Press; p. 19–35.

Theodoro SS, Putarow TC, Tiemi C, Volpe LM, Oliveira CAF, Glória MBA, Carciofi AC. 2019. Effects of the solubility of yeast cell wall preparations on their potential prebiotic properties in dogs. PLoS One. 14(11):e0225659.

Tibbetts GW. 2002. Nucleotides from yeast extract: potential to replace animal protein sources in food animal diets. In: Lyons TP, Jaques, KA, editors. Proceedings of the 18th annual symposium of nutritional biotechnology in the feed and food industries. Nottinghan: Nottinghan University Press; p. 435–443.

Tramontano M, Andrejev S, Pruteanu M, Klünemann M, Kuhn M, Galardini M, Jouhten P, Zelezniaik A, Zeller G, Bork P, et al. 2018. Nutritional preferences of human gut bacteria reveal their metabolic idiosyncrasies. Nat Microbiol. 3(4):514–522.

Waititu SM, Yin F, Patterson R, Rodriguez-Lecompte JC, Nyachoti CM. 2017. Short-term effect of supplemental yeast extract without or with feed enzymes on growth performance, immune status and gut structure of weaned pigs challenged with Escherichia coli lipopolysaccharide. J Anim Sci Biotechnol. 90:179–181.

Windey K, De Preter V, Verbeke K. 2012. Relevance of protein fermentation to gut health. Mol Nutr Food Res. 56(1):184–196.

Wu Y, Pan L, Tian Q, Piao X. 2018. Comparative digestibility of energy and ileal amino acids in yeast extract and spray-dried porcine plasma fed to pigs. Arch Anim Nutr. 72(1):76–84.

Zentek J, Ferrara F, Pieper R, Tedin L, Meyer W, Vahjen W. 2013. Effects of dietary combinations of organic acids and medium chain fatty acids on the gastrointestinal microbial ecology and bacterial metabolites in the digestive tract of weaning piglets. J Anim Sci. 91(7):3200–3210.