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Full Paper

Continuous intake of galacto-oligosaccharides containing syrup contributes to maintaining the health of household cats by modulating their gut microbiota

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The study of the relationships between the microbiota and intestinal environment of companion animals has gained increasing attention, particularly concerning health and disease. Previously, we demonstrated that continuous intake of galacto-oligosaccharides (GOS), a prebiotic, can improve the health of household dogs by modulating their gut microbiota. Given the potential health benefits of GOS in cats, we conducted a single-arm open-label study to evaluate the effects of a GOS-containing syrup on the gut microbiota and its metabolites in healthy cats. The study included 25 household cats and was conducted over 12 weeks. Each cat was fed 1.2 g of a GOS-containing syrup per day, equivalent to 0.5 g of GOS. Before the start of the study (week 0), during the 8-week intake period (weeks 4 and 8), and 4 weeks after the intake period (week 12), fecal microbiota, fecal organic acid and putrefactive product concentrations, fecal odor, and serum uremic toxin concentrations were assessed. The results showed that the levels of acetic acid-producing *Bifidobacteriaceae* significantly increased as a result of GOS intake. Additionally, *Peptostreptococcaceae* and *Eggerthellaceae* levels significantly decreased and increased, respectively, due to GOS intake. Furthermore, the concentrations of acetic, propionic, and *n*-butyric acids in feces significantly increased, whereas serum phenyl sulfate levels decreased significantly. These findings suggested that continuous GOS intake may contribute to the health of household cats.

Key words: galacto-oligosaccharides, household cats, microbiota, organic acids, uremic toxins, *Peptostreptococcaceae*, *Bifidobacteriaceae*

INTRODUCTION

Advances in genomic analysis technologies have shown that microbiomes or bacterial populations in various parts of the body play crucial roles in functions such as digestion and immunity, as well as in host health and disease [1]. Microbiome research has gained considerable attention in the field of veterinary medicine, where associations between the microbiome and various diseases have been identified, including inflammatory bowel disease [2, 3], diabetes [4], small cell lymphoma [5], obesity [6], feline immunodeficiency virus infection [7], and chronic kidney disease [8]. In cats, as in humans, probiotics, prebiotics, and synbiotics are used to regulate the gut microbiome [9, 10]. Prebiotics are dietary components that selectively stimulate the growth and activity of beneficial gut bacteria, such as bifidobacteria and lactobacilli, which help maintain the gut microbiota balance and promote host health [11]. Oligosaccharides are commonly used prebiotics that are added to pet foods for their functional, heat-resistance, and easy-to-process properties [12, 13]. Upon reaching the large intestine, oligosaccharides are metabolized by gut bacteria into organic acids such as acetic, propionic, and *n*-butyric acids [14]. Organic acids considerably contribute to the preservation of intestinal homeostasis by inhibiting the proliferation of pathogenic bacteria, preventing infections, preserving the integrity of the intestinal mucosa, and modulating the immune system [15]. Carbohydrate-derived metabolites such as oligosaccharides of gut bacteria are beneficial for host health, whereas protein-derived enterobacterial metabolites such as ammonia, phenols, and hydrogen sulfide are associated with the development of intestinal diseases such as inflammatory

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bowel disease (IBD) and colon cancer [16–18]. Phenol, indole, and *p*-cresol undergo sulfate conjugation in the liver and colonic mucosa, causing damage to the kidneys after being transported via the bloodstream, and contribute to the development of chronic kidney disease (CKD) [19–21]. As mentioned above, several studies have established strong relationships between the gut microbiota and their metabolites and various diseases. It is crucial to maintain a healthy intestinal environment by balancing the gut microbiota for disease prevention and overall health maintenance [19–21].

Galacto-oligosaccharides (GOS), a type of prebiotic, are composed of isomerized sugars with glucose termini and multiple galactose molecules [22]. Clinical studies on humans have shown that GOS increase the number of bifidobacteria in the intestine and improve defecation frequency [23, 24]. Similarly, previous clinical trials conducted by our group on household dogs showed that GOS intake provides various benefits by modulating the gut microbiota, in a manner similar to that in humans [25]. GOS may have similar effects on cats and has been evaluated in several clinical trials. However, these studies were conducted on cats housed in research facilities, which may have resulted in a biased initial microbiota composition due to similar rearing conditions and diets [26–28]. Furthermore, the studies evaluated changes in bacterial counts by culture or quantitative polymerase chain reaction (PCR) for a few specific bacteria and did not evaluate the overall gut microbiota composition [29].

Thus, this study aimed to evaluate the effects of continuous GOS intake on the overall gut microbiota profile and metabolites in healthy household cats from diverse backgrounds.

METHODS

Animals

This study was conducted on healthy cats living in households and was supported by the Kawasaki Veterinary Medical Association. The Kawasaki Veterinary Medical Association approved the study protocol. The inclusion criteria for the study were as follows: no antibiotic usage within the previous 2 weeks, no medical treatment needed during the study period, no health risks associated with fecal or blood collection, age of at least 1 year, owner's consent for sample collection, availability of the cat's medical history and other relevant information, and discontinuation of probiotic or prebiotic supplements before the start of the study. Owners were instructed not to alter the diets or lifestyles of their cats throughout the study and not to administer any probiotic, prebiotic, or antibiotic supplements. Written informed consent was obtained from all cat owners before enrolment.

Study protocol

The study was a single-arm open-label trial. A sugar syrup with at least 55% GOS, in which 4'-galactosyllactose (galactose β1-4 galactose β1-4 glucose) was the primary component in solids, was obtained from Yakult Pharmaceutical Industry Co., Ltd. [30]. This syrup was used as the test food, and parameters (fecal microbiota, fecal organic acid and putrefactive product concentrations, fecal odor score, and serum uremic toxin concentrations) were compared at prescribed time points before, during, and after intake of the test food. An outline of the study is presented in Fig. 1. Twenty-five cats received the GOS-containing syrup, amounting to 1.2 g (equivalent to 0.5 g GOS) per day, for 8 weeks. Fresh feces and blood samples were collected before (0 weeks [w], baseline), during (4 w and 8 w), and after (12 w) intake of the GOS-containing syrup. Feces were immediately stored at −20°C after collection and subsequently transferred to a storage unit kept at −80°C within 3 days. Blood was immediately transferred into a blood collection tube containing a serum separator. The solutions were mixed, and subsequently allowed to stand at 20–25°C for approximately 30 min. Serum was then separated by centrifugation at 1,300 g for 10 min and stored at −80°C. Blood samples were collected after a minimum fast of 8 hr.

Fecal processing and DNA extraction

Fecal processing and DNA extraction were performed as described previously [25].

Amplicon analysis of the 16S rRNA gene

The composition of the fecal microbiome was determined via amplicon sequencing of the 16S rRNA gene using the procedure described in our previous study [25]. Briefly, DNA extracted from feces was amplified using forward (515F: 5′-GTGCCAGCMGCCGCGGGTAA-3′) and reverse (806R: 5′-GGACTACHVGGGGTWTCTAAT-3′) primers targeting the V4 region of the 16S rRNA gene [31]. Amplified DNA was purified, quantified, and sequenced on a MiniSeq system using a MiniSeq Mid Output Kit (Illumina Inc., San Diego, CA, USA). Sequencing reads were imported into the QIIME 2 software (ver.

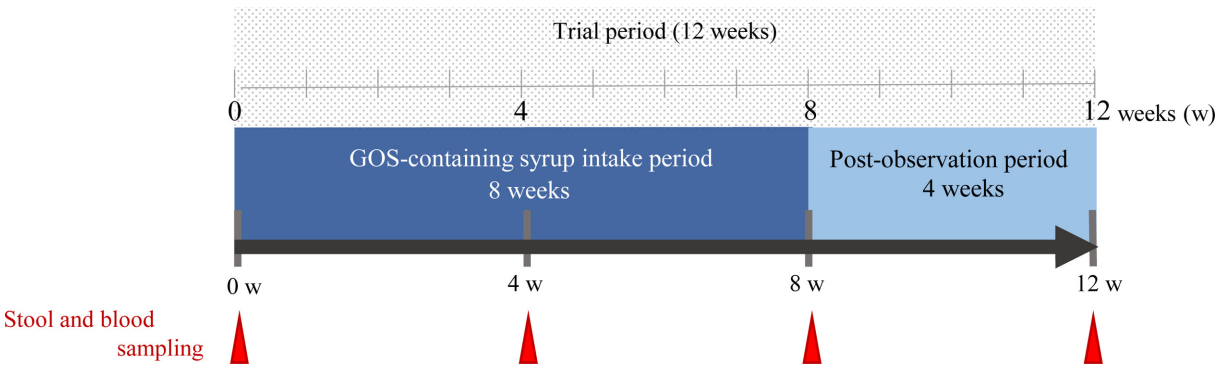

Fig. 1. Study outline. GOS: galacto-oligosaccharides.

2022.2) to process the sequencing data and were subsequently assigned to taxonomic groups using the SILVA 138 database [32]. Weighted UniFrac distance and α-diversity indices (Faith's phylogenic diversity [PD], observed amplicon sequence variants [ASVs], and Shannon index) were calculated using the minimum number of reads in the analyzed samples (depth, 10,587).

Total bacterial count

Quantification of total bacterial counts in feces was performed via qPCR using an ABI PRISM 7900HT Sequence Detection System (Life Technologies Japan Ltd.), according to the method described by Shima *et al.* [33]. The total number of bacteria was determined using forward (UniF:5′-GTGSTGCAYGGGYYGTCGTCA) and reverse (UniR: 5′-ACGTCRTCCMCNCCTTCCTC) primers. The standard strain (*Faecalibacterium prausnitzii* ATCC 27768T) was quantified via qPCR.

Measurement of fecal organic acid concentrations

The fecal concentrations of organic acids (succinic, lactic formic, acetic, propionic, *n*-butyric, isobutyric, *n*-valeric, and isovaleric acids) were determined using high-performance liquid chromatography (HPLC) following a method described previously [25].

Measurement of fecal putrefactive product concentrations

The fecal concentrations of putrefactive products (phenol, indole, and *p*-cresol) were determined using HPLC following a method described previously [25].

Measurement of serum uremic toxin concentrations

Concentrations of phenyl sulfate, indoxyl sulfate, and *p*-cresyl sulfate were determined via HPLC mass spectrometry, using the method described by Kawase *et al.* [34]. The trimethylamine N-oxide (TMAO) concentration was determined using a modified version of the method described by Wu *et al.* [35]. Briefly, the analytical equipment included an ACQUITY TQD MS system (Waters, Milford, MA, USA) and Inertsil HILIC column (3.0 µm × 150 × 2.1 mm, GL Sciences, Tokyo, Japan). The measurement conditions were as follows: solution A, 0.1% formic acid (10 mM ammonium acetate); solution B, MS grade acetonitrile (10 mM ammonium acetate); column temperature, 40°C; flow rate, 0.25 mL/min; injection volume, 3 µL; and gradient procedure, 0–3 min (solution A, 100%), 3–4 min (solution A, from 100 to 60%), 4–7 min (solution A, 60%), 7–8 min (solution A, from 60 to 0%), and 8–14 min (solution A, 0%). The system was controlled using the Waters MassLynx mass spectrometry software (Waters). Serum uremic toxin concentrations were measured at Kyoto Institute of Nutrition & Pathology, Inc. (Kyoto, Japan).

Questionnaire survey on fecal odor score

A questionnaire survey was conducted in which the owners were asked to evaluate the fecal odor experienced during fecal collection on a 5-point scale, with a score of 5 indicating "very smelly" and a score of 1 indicating "not smelly".

Stratified analysis

To evaluate the cats' responses to the GOS-containing syrup based on age, a stratified analysis was performed. The cats were divided into a young-adult group (under 7 years old, n=18) and a mature-adult/senior group (>7 years old, n=7) according to the 2021 AAHA/AAFP Feline Life Stage Guidelines [36].

Statistical analyses

All statistical analyses were performed using the EZR (ver. 1.61) and R (ver. 4.2.2) software. The relative abundances of bacterial taxa at the family level were analyzed using the ALDEx2 package [37]. Only bacterial taxa identified in >10% of all fecal samples were analyzed from the 16S rRNA amplicon sequencing [37]. Alpha diversity indices (Faith's PD, observed ASVs, and Shannon index), total bacteria and bacterium counts, fecal concentrations of organic acids and putrefactive products, serum uremic toxin concentrations, and fecal odor score were analyzed using the Wilcoxon signed-rank test. The composition of the microbiota was determined using principal coordinate analysis based on the weighted UniFrac distance and compared using permutational multivariate analysis of variance. These analyses involved pre- and post-intake comparisons using pre-intake data (0 w) as the baseline. Multiplicity correction was applied using the Benjamini-Hochberg method for weighted UniFrac distance and the Bonferroni method for the remaining analyses. A p-value with multiplicity correction or we.eBH (the expected Benjamini-Hochberg corrected p-value for the Welch's t-test) of <0.05 was considered to indicate statistical significance, and a p-value with multiplicity correction or we.eBH of ≥ 0.05 and <0.1 was considered to indicate marginal significance.

In the stratified analysis, fecal concentrations of organic acids, putrefactive products, and serum uremic toxin levels were analyzed using the Mann–Whitney U test, with $p<0.05$ indicating statistical significance.

RESULTS

Clinical profiles of the studied animals

A total of 26 cats were enrolled in this study. During the second week of the study, one cat was withdrawn from the study by its owner because of a disease unrelated to the intake of the test food. The remaining 25 cats completed the study and were included in the analysis. Background information of the cats is presented in Supplementary Table 1. Fecal samples were obtained from all cats. Blood samples were collected at all time points from 21 of the 25 cats. The GOS-containing syrup intake rate among the 25 cats in the analysis was 98 ± 4%. All cats were determined to be in good health based on blood biochemistry and veterinary examinations. Notably, no adverse reactions (e.g., diarrhea) to the GOS-containing syrup were observed.

Effects on fecal microbiota

One fecal sample collected at one of the time points was excluded from the microbiota analyses owing to an insufficient number of reads obtained during sequencing. Therefore, the analysis was performed using 24 samples instead of 25. There was a significant decrease in the observed ASVs during the intake period (4 and 8 w) compared with the baseline (0 w), whereas no significant changes were identified in Faith's PD or the Shannon Index (Fig. 2A). Figure 2B shows the changes in microbiota based on weighted UniFrac distance from baseline (0 w) through the intake period (4 and 8 w) to 4 weeks after the end of intake (12 w), and the results indicated that the composition at 8 and 12 w was marginally different from that at baseline (0

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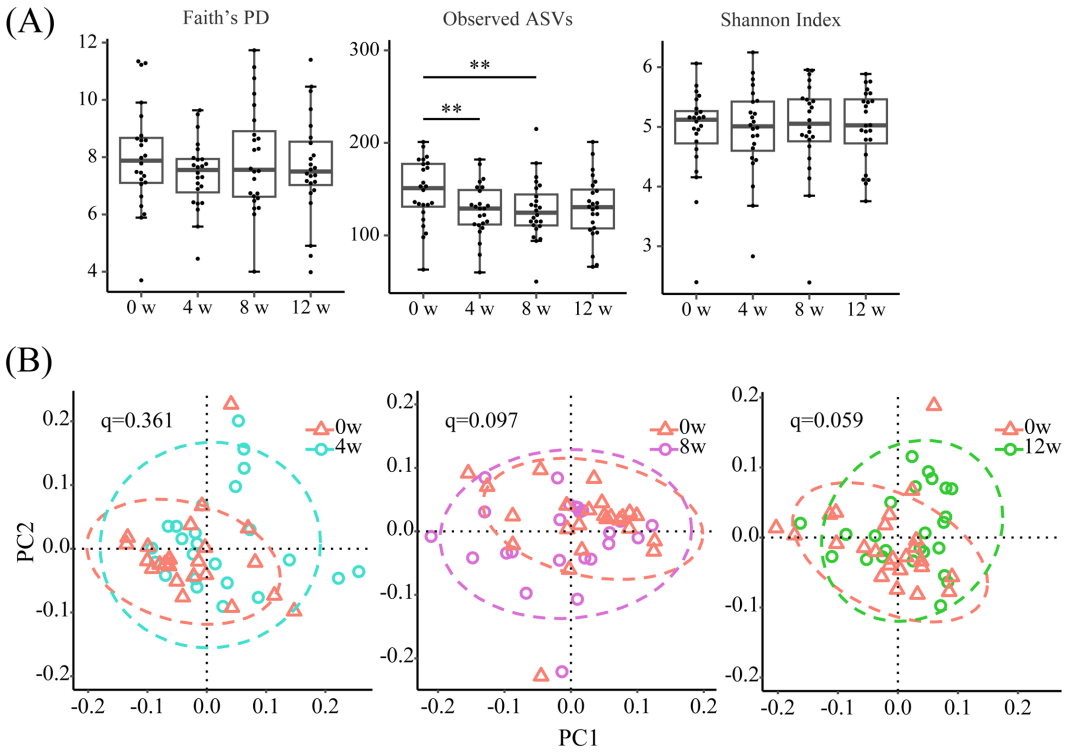

Fig. 2. Changes in diversity of fecal microbiome during the study period. (A) α-diversity. (B) β-diversity. α-diversity data are expressed as box plots, β-diversity is expressed as PCoA plots based on weighted UniFrac distance. Dashed lines indicate 95% confidence intervals. One sample had very few reads in the MiSeq output, and therefore, the corresponding sample was excluded from all microbiota analyses (n=24). Significant differences in α- and β-diversity were determined using the Wilcoxon signed-rank test with Bonferroni correction, and PERMANOVA with Benjamini-Hochberg correction, respectively (vs. 0 weeks). **p<0.01. PCoA: principal coordinate analysis; PERMANOVA: Permutational multivariate analysis of variance.

w). The changes in the abundances of the top bacterial families are summarized in Fig. 3A. During the GOS intake period (4 and 8 w), the abundances of *Selenomodaceae*, *Veillonellaceae*, and *Bifidobacteriaceae* markedly increased compared with the baseline (0 w), whereas the abundance of *Peptostreptococcaceae* decreased. After the end of the GOS-containing syrup intake period (12 w), the abundances of the families with observed changes returned to their baseline levels.

To further investigate the impact of the GOS-containing syrup on microbiota, a compositional differential abundance analysis was conducted on each family using ALDEx2. The results indicated a significant increase in the abundance of *Eggerthellaceae* during the intake period (4 and 8 w) compared with that at baseline (0 w; Fig. 3B). Additionally, a significant increase in *Bifidobacteriaceae* and a significant decrease in *Peptostreptococcaceae* were observed at 8 w compared with baseline (0 w). *Selenomodaceae* and *Veillonellaceae* did not show significant changes in the ALDEx2 analysis.

Total bacterial counts in feces were measured via qPCR and did not vary significantly throughout the study period. The bacterial counts in each family were calculated by multiplying the total bacterial counts by the abundances of the bacteria in each sample. The total bacterial counts multiplied by the abundances of the three bacterial families that showed significant variations in ALDEx2 showed significant variations in the bacterial counts for all three bacterial families. Compared with the baseline (0 w), the counts of *Peptostreptococcaceae* and the counts of *Eggerthellaceae* and *Bifidobacteriaceae* decreased significantly and increased significantly, respectively, during the GOS-containing syrup intake period (4 and 8 w). The *Bifidobacteriaceae* counts showed a significant decrease after the end of the GOS-containing syrup intake (12 w) compared with baseline (0 w).

Effects on fecal metabolites and fecal order

Two of the collected samples lacked sufficient quantities to determine the concentrations of putrefactive products, so the remaining 23 samples were analyzed. The changes in the concentrations of the three major organic acids in feces (acetic, propionic, and *n*-butyric acids) are shown in Fig. 4A. Significant increases in the concentrations of acetic acid, propionic acid, and *n*-butyric acid were observed during the intake period (4 w) compared with baseline (0 w). There were also significant increases in the concentrations of succinic, lactic, formic, isobutyric, *n*-valeric, and isovaleric acid concentrations during the intake period (Supplementary Table 2). In contrast, the concentrations of putrefactive products (phenol, indole, and *p*-cresol) showed no significant changes compared with those at baseline (0 w), during (4 and 8 w), and after the end of intake (12 w; Fig. 4B). In addition, no noticeable changes were recorded in fecal odor scores during the study (data not shown).

Effects on serum uremic toxin concentrations

Serum phenyl sulfate levels were significantly lower during the intake period (8 w) and at 4 weeks after the end of intake (12 w)

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than at baseline (0 w). There were no significant differences in the indoxyl sulfate, *p*-cresyl sulfate, or TMAO levels during the study period (Fig. 5).

Stratified analysis

No significant differences were observed in the effects of GOS-containing syrup intake between the groups, except for *p*-cresyl sulfate (Supplementary Table 3). The *p*-cresyl sulfate concentration showed a significantly greater decrease in the young-adult group than in the mature-adult/senior group during the intake period (4 w) and at 4 weeks post-intake (12 w).

DISCUSSION

In this study, we evaluated the effects of continuous intake of a GOS-containing syrup on the gut microbiota and its metabolites in healthy household cats. Continuous GOS intake for 8 weeks altered gut microbiota, increased fecal organic acid

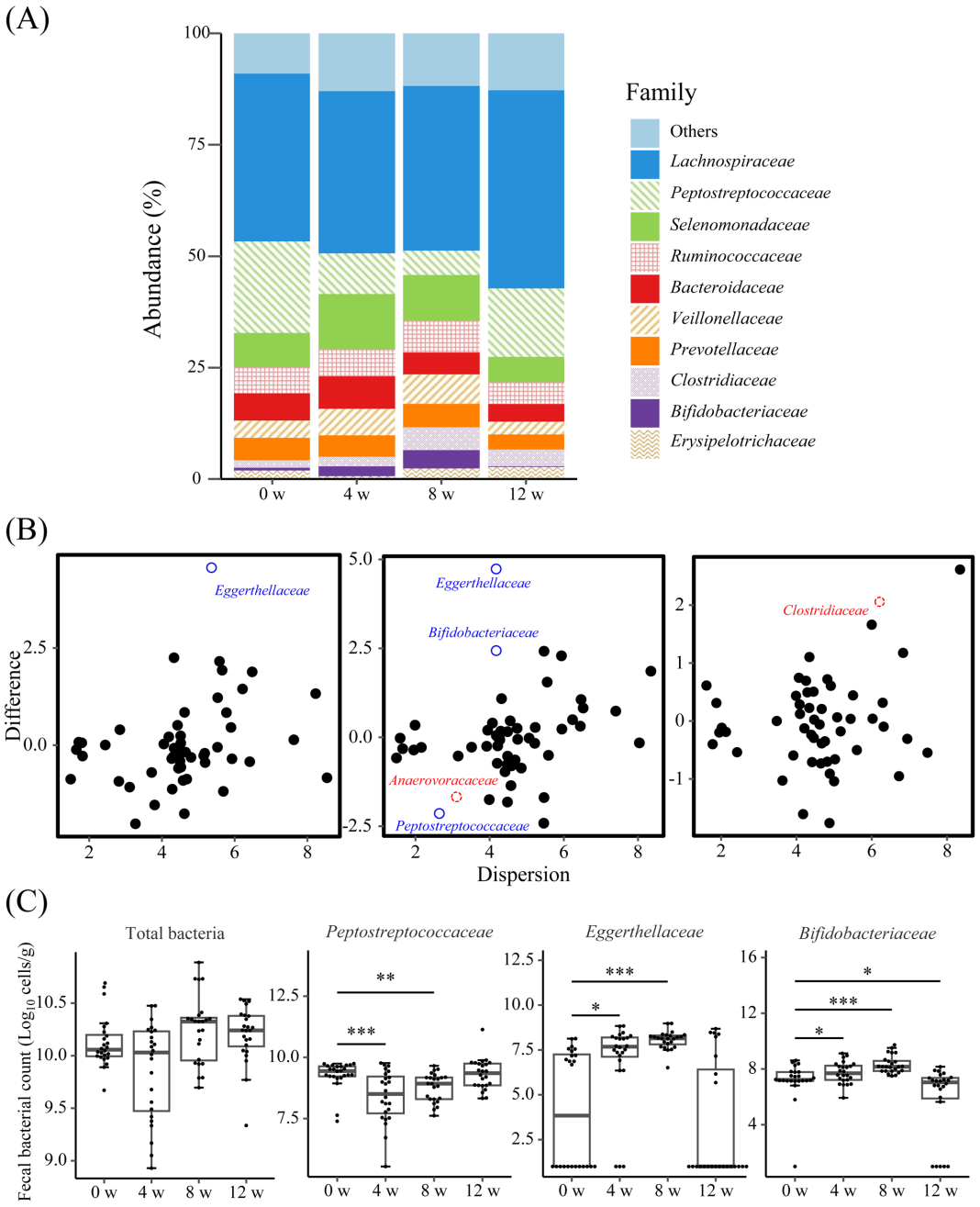

Fig. 3. Changes in abundance of fecal microbiota during the study period. (A) Intestinal abundance. (B) ALDEx2 analysis of differential abundance (Left: 0–4 weeks, Central: 0–8 weeks, Right: 0–12 weeks) (C) Fecal bacterial counts. Abundance data are expressed as the mean, fecal bacterial count data are expressed as box plots. One fecal sample collected at one of the time points was excluded from microbiota analyses owing to insufficient number of reads obtained during sequencing (n=24). Significant differences in ALDEx2 analysis of differential abundance were determined using the Welch's t-test with Benjamini–Hochberg correction (vs. 0 weeks). Blue circle p<0.05, red dashed circle p<0.1. Significant differences in bacterial counts were determined using the Wilcoxon signed-rank test with Bonferroni correction (vs. 0 weeks). *p<0.05; **p<0.01; ***p<0.001.

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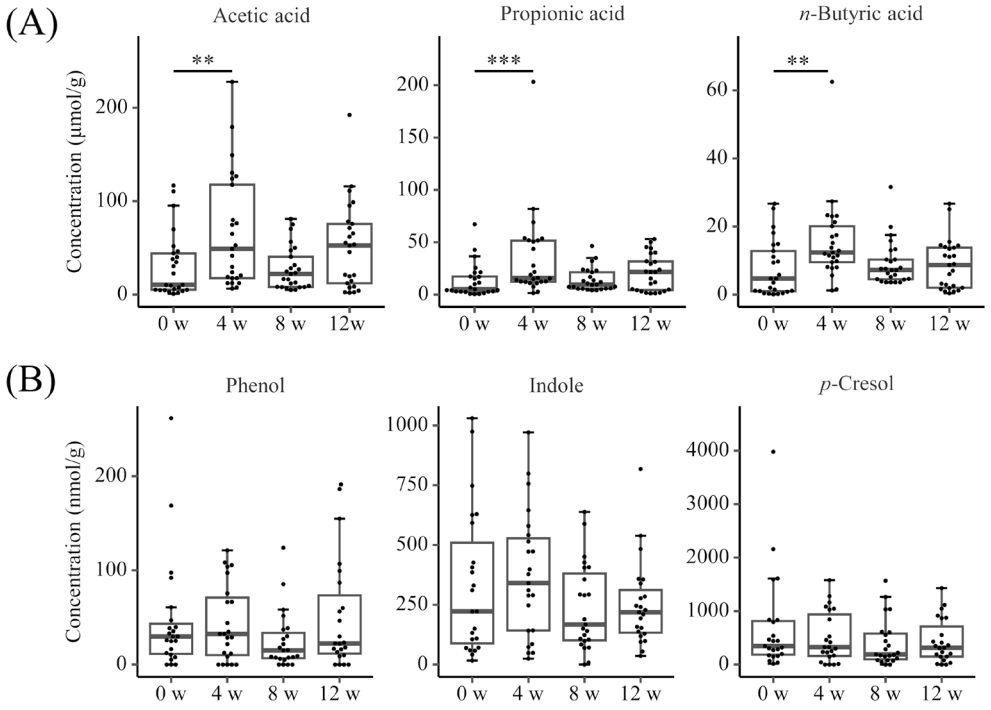

Fig. 4. Changes in the concentration of fecal organic acids and putrefaction products during the study period. (A) Organic acids concentration, n=25. (B) Putrefaction product concentration. Two of the samples collected were found to be insufficient for measuring fecal putrefactive product concentrations due to the limited quantity of stool samples obtained (n=23). Data are expressed using box plots. Significant differences were determined using the Wilcoxon signed-rank test with Bonferroni correction (vs. 0 weeks). **p<0.01; ***p<0.001.

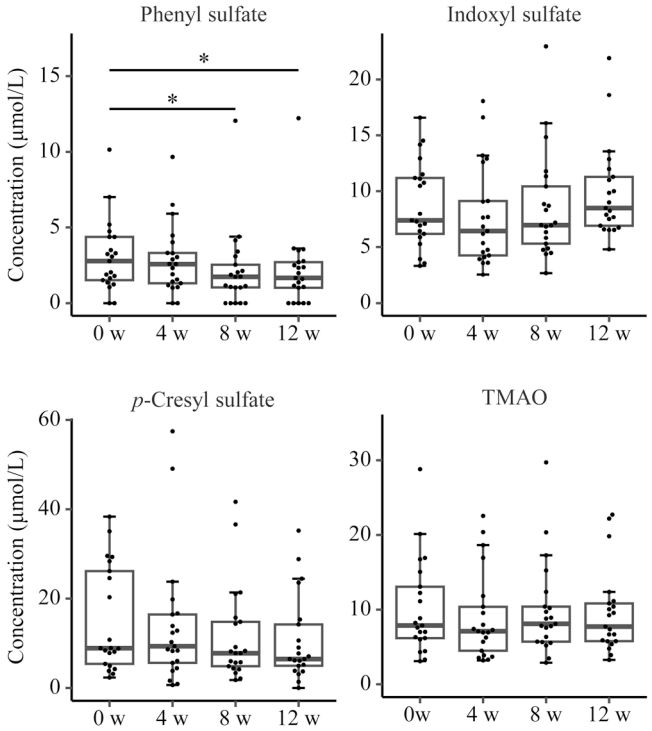

Fig. 5. Changes in serum levels of uremic toxins during the study period. Data are expressed using box plots. Blood samples were submitted for all points in 21 of the 25 cats (n=21). Significant differences were determined using the Wilcoxon signed-rank test with Bonferroni correction (vs. 0 weeks). *p<0.05.

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concentrations, and decreased serum uremic toxin concentrations. Previous studies on cats raised in similar facility environments have evaluated the effects of GOS intake on specific bacterial species, such as *Bifidobacterium*. However, these studies did not evaluate the impact of GOS intake on the overall gut microbiota or serum uremic toxins and had limited analyses of organic acids and putrefactive products. This study identified changes in these parameters and demonstrated the efficacy of GOS in healthy household cats from diverse rearing environments.

Changes in α- and β-diversity were observed during the GOS intake period (4 and 8 w), indicating that GOS intake changed the feline gut microbiota within the period (Fig. 2A, 2B). Specifically, significant changes in the abundances of several species from families such as *Peptostreptococcaceae*, *Eggerthellaceae*, and *Bifidobacteriaceae* were observed after GOS intake (Fig. 3A, 3B). Among these changes, the increase in *Bifidobacteriaceae* is presumed to be due to their β-galactosidase activity, which enables them to utilize GOS [38]. Conversely, most *Eggerthellaceae* lack β-galactosidase activity [39, 40], and their increase may have resulted from indirect effects, possibly by utilizing sugars hydrolyzed by GOS-utilizing bacteria such as *Bifidobacteriaceae* or in response to GOS-induced changes. The reason for the decrease in *Peptostreptococcaceae* remains unclear; however, similar to *Eggerthellaceae*, it may have been associated with indirect effects mediated by GOS, such as increased organic acid concentrations.

A significant increase in acetic acid, which inhibits the proliferation of pathogenic bacteria and prevents infections, and its typical producer, *Bifidobacteriaceae*, was observed. Several strains of *Bifidobacteriaceae* are used as probiotics in cats and recognized as beneficial bacteria [41]. A significant decrease in *Bifidobacterium* abundance has been reported in cats with IBD compared with the abundance in healthy cats [28]. The significant increase in the abundance and counts of *Bifidobacteriaceae*, as well as the concentration of its metabolite acetic acid, suggests that continuous intake of GOS may be beneficial for the maintenance of gut health in cats by improving gut microbiota and the intestinal environment. However, no correlation was found between acetic acid concentrations and the number or abundance of *Bifidobacteriaceae*, suggesting that the utilization of acetic acid by the host and other gut bacteria may increase [42]. In addition, we found a significant increase in the levels of propionic acid, which has anti-inflammatory properties, and *n*-butyric acid, which is involved in inducing regulatory T-cell differentiation [15]. *Selenomodaceae* and *Veillonellaceae*, which showed highly increased, non-significant abundances among the top 10 families, are known to include propionic acid-producing bacteria (Fig. 3A) [43, 44]. *Selenomodaceae* includes *Megamonas*, a GOS-utilizing propionate-producing species. Our previous study in dogs showed an increase in *Megamonas* and propionic acid with continuous GOS intake [25]. Although cats and dogs are both typical companion animals, the gut microbiota of the two differ greatly [45]. However, in this study, similar changes in the concentrations of major intestinal organic acids were observed in cats as in dogs after continuous intake of a GOS-containing syrup. Thus, GOS have a beneficial effect on various animal species with different gut microbiota, similar to their effects in humans [15].

Intake of the GOS-containing syrup resulted in a significant decrease in the concentration of phenyl sulfate in the serum (Fig. 5). Additionally, significant reductions in TMAO and *p*-cresol were observed during the intake period (TMAO, p=0.038 for 0 vs. 4 w; *p*-cresol, p=0.038 for 0 vs. 8 w), although prior to multiple corrections. Phenyl sulfate is found in high concentrations in the serum of patients with CKD [46]. Phenyl sulfate reportedly decreases oxidative stress resistance and exhibits cytotoxic activity in LLC-PK1 cells (porcine renal tubule-derived cell line) *in vitro* [47]. Similarly, TMAO is associated with tubulointerstitial fibrosis and dysfunction in animal models, suggesting a link to the severity of feline CKD [48, 49]. TMAO is a urinary toxin metabolized in the liver from trimethylamine and is produced by *Peptostreptococcaceae*, a common feline bacterium [8, 50–52]. In this study, *Peptostreptococcaceae* abundance and counts were also significantly lower during the intake period (Fig. 3B and 3C). The metabolites of *p*-cresol reportedly have toxic effects and cause cardiovascular and renal damage, as well as effects on CKD progression [53]. Several methods of using probiotics and prebiotics to reduce *p*-cresol levels have been studied [53], and GOS have been reported to be effective in rats [54]. The results showing a reduction of *p*-cresol in cats after GOS intake suggested that GOS may have a preventive effect on CKD in cats. Therefore, the reduction in phenyl sulfate, *p*-cresol, and TMAO due to GOS intake may lead to a reduction in kidney damage and a further benefit for cats with a high incidence of CKD. However, the relationships between the reduction in these uremic toxins and changes in the microbiota were not clarified in this study. To elucidate these mechanisms, additional investigations, such as a metabolic pathway analysis using shotgun metagenomic analysis, are required.

The stratified analysis revealed that age has a minimal impact on the effects of GOS on fecal metabolites and serum uremic toxins (Supplementary Table 3). However, the baseline serum *p*-cresyl sulfate concentrations were notably higher in the young-adult group, which exhibited a particularly strong reduction effect, than those in the mature-adult/senior group. Additionally, similar trends were observed for the baseline levels of indole and *p*-cresol. These baseline differences are presumed to have been due to variations in gut microbiota composition and dietary differences between the two groups. The gut microbiota also changes with age in cats [55]. In this study, differences in baseline β-diversity were observed between the two groups (Supplementary Fig. 1). Although a detailed dietary survey was not conducted, the reduced food intake in older cats may result in lower protein intake, which serves as a substrate for putrefactive products and uremic toxins.

This study had several limitations. First, it was a single-arm, open-label study. A double-blind, placebo-controlled, parallel-group study is required to provide more evidence for the benefits of GOS in cats. Second, the GOS-containing syrup used in this study was a liquid sugar produced industrially by an enzymatic reaction, and 45% of its solid components consist of monosaccharides and other non-GOS ingredients. Sugars other than GOS may have contributed to the effects observed in this study. Third, the use of a purer GOS test product would be effective for clarifying its efficacy in improving the gut microbiome composition of cats.

In conclusion, 8 weeks of continuous GOS intake in household cats with diverse backgrounds and rearing environments altered the composition of the gut microbiota, increased organic acids, and decreased blood uremic toxin concentrations. Thus,

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continuous GOS intake may help maintain gut health in cats and prevent diseases.

CONFLICT OF INTEREST

None.

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