

Fermo-Food for Diarrheic or Constipated Cats: Fermented Turmeric (*Curcuma longa* L.) Alleviated Fecal Consistency

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This study was conducted to detect the probable efficacy of home prepared, by our scientific and clinical receipt, fermented turmeric (*Curcuma longa* L.) for treatment of diarrhea or constipation among cats with gastrointestinal issues. Turmeric, derived from *Curcuma longa*, and *Lactobacillus paracasei*, a lactic acid bacteria, have thoroughly been investigated for their potential gastrointestinal health promoting efficacy. A 6-marker scale was preferred for describing diarrhoea ≤ 3 (Group I – $n = 14$ cats), severe diarrhoea ≤ 2 (Group II – $n = 9$ cats) and constipation as 6 Group III – 7 cats. Raw, unfiltered, unpasteurized apple cider/hawthorn and grape vinegars were boosted with fermented turmeric with 3 probiotic strains [*L. plantarum* LP6595, *L. plantarum* HEAL9 and *S. boulardii*], were preferred as starter cultures for exhibition. They all were combined with sterile water 250 ml and vinegars combination (25 mL each) at the level of 2.5×10^9 CFU/mL each, respectively. Finally, curcumin obtained from a commercial manufacturer (fermented turmeric was standardized to 0.86 mg curcumin per 1.0 gr powder) was added onto final solution. Fermentation process took 14 days at 20 °C. Obtained fermented curcumin, as fermo-food was prescribed orally at a dose of 1.7 mL per cat per day for 10 consecutive days. The primary efficacy endpoint was changes in fecal scoring. Within the comparison between the pre- and post-treatment scores of each group, a significant difference was found in Group 1 ($p = 0.001$), Group 2 ($p = 0.05$) and Group 3 ($p = 0.015$). Moreover, a statistically significant difference was found between Group 1 and Group 2 ($p = 0.006$) and between Group 1 and Group 3 ($p = 0.001$), but the difference between Group 2 and Group 3 was not found to be statistically significant ($p = 0.214$). This clinical case control study promptly showed that fermo-food, as fermented curcumin prepared as a clinical receipt should have helped maintaining gastrointestinal health among cats with diarrhea and constipation.

Keywords: fermented turmeric, fermo-food, gastrointestinal issues, cats, *Lactobacillus paracasei*

1 Introduction

Specifically within the last era, growing body of evidence focused on traditional food material in entire world. Traditional fermented foods are selectively very important for health and well-being (Hesseltine & Wang, 1980). Moreover several researches regarding traditional foods have been linked to modifying health issues, life quality, satiety, safety and processing methods of these products.

Turmeric (*Curcuma longa* L.) exhibited an important participation as being an ayurvedic spice (Chumroenphat et al., 2021). As its well-recognized yellow texture, the turmeric plant itself involved 3 curcuminoid ingredients: curcumin (77%), demethoxycurcumin (17%), and bisdemethoxycurcumin (3%) accompanied by

essential oils (Lee et al., 2014). It serves as a feed additive against inflammatory state for obese cats (Leary et al., 2011), anti-proliferative and apoptotic effects against feline mammary gland tumor (Deveci Ozkan et al., 2021), in cats with osteoarthritis (Lefort-Holguin et al., 2024), feline head and neck dermatitis (Ural et al., 2019), feline infectious peritonitis (Ng et al., 2020) curcumin trials were evident. On the other hand, a study performed in laying hens investigated the efficacy of fermented turmeric on the phytobiotic ingredient and performance for production. In the latter research turmeric undergone anaerobic fermentation at 24°C for 7. Afterwards this fermented was fed, in which feed consumption was sixty-four percent increased. Available data in that study claimed that fermented turmeric efficaciously established its feed additive values (Kasri et al., 2024b), whereas this

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issue has been hindered by its low bioavailability (Adli, 2021; Adli et al., 2023), probably splitting beneficiary efficacy even administered directly to animals.

This study was designed to evaluate the potential regulatory effects of a supplement containing fermented turmeric obtained by traditional fermentation methods and enriched with probiotic microorganisms on fecal consistency in cats showing gastrointestinal system disorders (especially diarrhea and constipation).

2 Material and Methods

2.1 Demographic Distribution

All cats were admitted to Aydin Adnan Menderes University, Faculty of Veterinary, Department of Internal Medicine. First inclusion criteria was diarrhea or constipation at referral. All animal owners gave written consent for enrollment at the study. A total of 30 cats were enrolled. Demographic data involving group classification of cats were as shown below.

- Group I – $n = 14$ cats, 5 female 9 male, 1.5–7 years of age, 11 pure breed, 3 crossbred with faecal scoring of ≤ 3 (mild diarrhoea).
- Group II – $n = 9$ cats, 6 female, 3 male, 9 months to 9 years of age, 5 purebred, 4 crossbred with fecal scoring of ≤ 2 (severe diarrhoea).
- Group III – 7 cats, 5 female, 2 male, 3–10 years of age, 6 purebreds, 1 crossbred with fecal scoring 6 (as constipation).

2.2 Sample Collection and Scoring

Serum biochemistry and hematological analytes were deemed available (data not necessarily shown). Faecal samples were collected from each cat's litter tray on the first and 10th days of the trial. Even if faeces were not present, cat owners were advised to take precautions and keep interest till defecation. Animal owners were instructed to take relevant photographs of fecal samples and forwarded to researchers of this manuscript as soon as possible for scoring. Faecal consistency was classified 1 (watery), 2 (the vast majority unformed), 3 (nearly 50% formed), 4 (mostly formed with a small scale softer), 5 (well formed, entirely firmed) and 6 (hard, dry) through interpretation with the Cats Protection Faecal Scoring System (Allison et al., 2015), an altered version of the Bristol Stool Scale (Meyers Scale) (Lewis & Heaton, 1997). Previously determined classification involved and allowed us to further classify subgroups: faecal scoring of ≤ 3 (mild diarrhoea), ≤ 2 (severe diarrhoea) and 6 as constipation, respectively.

2.3 Fermentation Process

Raw, unfiltered, unpasteurized apple cider/hawthorn and grape vinegars were boosted with fermented turmeric. Three probiotic strains, *L. plantarum* LP6595 (2.5 mg, 1.5 milliard cfu), *L. plantarum* HEAL9 (2.5 mg, 1.5 milliard cfu) and *S. boulardii* (125 mg, 2.5 milliard cfu), were preferred as starter cultures for exhibition. Commercially available (Enterobiotik® Capsule) and well-known strains were deemed available and purchased by B. Ural during his visit to Montenegro. Colony forming units were known, as declared by the company (Enterobiotik® Capsule, the cell number reached 2.5×10^9 CFU/mL), allowed us to skip strain inoculation. The cells in ready capsules were pelleted through centrifuge, at 5000 x g for 10 minutes at 20 °C, and the obtained suspension were washed in phosphate-buffered saline solution (PBS, pH 7.4) twice, similar to what have been reported previously (Başyigit Kılıç et al., 2016). Available probiotic chains was poured on sterile water 250 ml and grape/apple/hawthorn vinegars combination (25 ml each) at the level of 2.5×10^9 CFU/mL each, respectively for *L. plantarum* and *S. boulardii*. Finally, curcumin obtained from a commercial manufacturer (fermented turmeric was standardized to 0.86 mg curcumin per 1.0 gr powder) was added onto final solution. Fermentation process took 14 days at 20 °C (Başyigit Kılıç et al., 2016). At a final step of fermentation, turmeric liquid extract (Kasri et al., 2024a) was deemed available. Each finally obtained turmeric liquid extract was subjected to preparation at 100% the same ingredients in same route and methodology, for standardization. In the present study characteristics of microbiological antibacterial and chemical activities of turmeric were not analyzed. Therefore, at the time of writing and performing the study other than pH total acid total phenol total sugar total microbial CFU/mL and inhibition zone diameters for pathogenic bacteria remained unclear. However, our next and subsequent study would thus be aimed for this issue.

2.4 Statistical Analyses

SPSS 29.0 (IBM, USA) program was used to analyze the data obtained in the study. Stool score data were evaluated separately for pre- and post-treatment periods. The normality distribution of the data was examined with the Shapiro-Wilk test; non-parametric test methods were used because the data belonging to the groups did not show normal distribution. The changes in stool scores of each group before and after treatment were analyzed with the Wilcoxon signed ranks test, which is appropriate for paired samples. Kruskal-Wallis variance analysis was applied separately for the pre- and post-treatment periods in comparing the differences in stool scores between the groups. In cases where a significant

difference was detected as a result of the Kruskal-Wallis test, pairwise group comparisons were evaluated with Dunn's multiple comparison test. In all analyses, $p < 0.05$ was considered statistically significant.

3 Results and Discussion

Available data regarding Cats Protection Faecal Scoring System (Allison et al., 2015), a modified version of the Bristol Stool Scale (Meyers Scale) (Lewis & Heaton, 1997) was used, were shown below in table 1.

The primary efficacy endpoint was changes in fecal scoring. Before treatment, fecal scores in Group 1 cats ranged from 1 to 2, with an average of 1.50, a median of 1.5, a minimum of 1, and a maximum of 2. In Group 2 cats, the scores were only 3, with an average and median of 3.0. In all Group 3 cats, the fecal score was recorded as 6, with an average, median, minimum, and maximum values of 6.0 in this group. After treatment, fecal scores in Group 1 cats ranged from 2 to 5, with an average of 4.14 and a median of 4.0. In Group 2 cats, the scores ranged from 2 to 5, with an average of 3.89 and a median of 4.0. In Group 3, fecal scores ranged from 3 to 5, with an average of 4.14 and a median of 4.0. In the comparison between the pre- and post-treatment scores of each group, a significant difference was found in Group 1 ($p = 0.001$), Group 2 ($p = 0.05$) and Group 3 ($p = 0.015$) with the Wilcoxon signed rank test and it was determined that the changes in stool scores were significant. In the comparison of pre-treatment stool scores between the groups, a significant difference was determined as a result of the Kruskal-Wallis test ($p < 0.0001$). In the pairwise comparisons made with the post-hoc Dunn test, a statistically significant difference was found between Group 1 and Group 2 ($p = 0.006$) and between Group 1 and Group 3 ($p = 0.001$), but the difference between Group 2 and Group 3 was not found to be statistically significant ($p = 0.214$). When the stool scores between the groups after treatment were evaluated with the Kruskal-Wallis test, no statistically significant difference was found between the groups ($p = 0.8303$).

4.1 Fermented Turmeric: Novel fermo-food

Fermentation procedure composed of microorganism's usage for to transmogryfy available curcuminoids and other related compounds exhibited turmeric into further bio accessible and probably favorable forms. During turmeric fermentation several microorganisms [bacteria, yeast etc.] might be preferred and could be exhibited in various routes [i.e. solid-state and submerged fermentation]. The superlative circumstances for fermentation could be exhibited through trials, which might be depended on the target product (Salve et al., 2023). Fermented turmeric exhibited elevated antioxidant action, total phenolic ingredient and bio accessibility of curcuminoids in contrast to unfermented turmeric, creating a propitious provenance of functional ingredient with probable health beneficiary (Salve et al., 2023). The first author of this manuscript (K.U.) described it as fermo-food, in which in his book of 'Curcumin Collection' (2023) he full filly denoted several cases were cured with curcumin. Regarding solid-state fermentation, the microorganisms extend on exterior side of a solid substrate [i.e. rice bran/ wheat bran], to those of shielded with the microorganisms. Following adjoint of turmeric to the substrate, fermentation might be started (Martgrita et al., 2022). This kind of solid-state fermentation has been linked to modify manufacturing curcuminoids [tetrahydrocurcumin/ demethoxycurcumin/bisdemethoxycurcumin], providing antioxidant and anti-inflammatory activity. On the other hand, for submerged fermentation, available turmeric has been pour on a liquid channel, previously shielded with necessary microorganisms. This kind of fermentation has been performed in a bioreactor, establishing necessary conditions for microorganism growth. The later type of fermentation supported elevated bioaccessibility of the curcuminoids in turmeric and to exhibit ferulic acid occurence, that has been full of antioxidant and anti-inflammatory issues (Singgih & Julianti, 2015). Fermentation period could change due to preferred microorganisms, the substrate/medium, and final compound targetted. Peak conditions to perform fermentation involve pH, temperature and

Table 1 Alteration in fecal score before and after fermo-food application

| Fecal Score | Group I mild diarrhea | | Group II severe diarrhea | | Group III constipation | |
|-------------------------|-----------------------|-------|--------------------------|-------|------------------------|-------|
| | before | after | before | after | before | after |
| Mean | 1.5 | 4.14 | 3.0 | 3.89 | 6.0 | 4.14 |
| Median | 1.5 | 4.0 | 3.0 | 4.0 | 6.0 | 4.0 |
| SD | 0.52 | 0.83 | 0.00 | 0.78 | 0.00 | 0.69 |
| Min | 1.0 | 2.0 | 3.0 | 2.0 | 6.0 | 6.0 |
| Max | 2.0 | 5.0 | 3.0 | 5.0 | 3.0 | 5.0 |
| P-value (Wilcoxon test) | 0.001 | | 0.05 | | 0.015 | |

oxygen concentration. Variable fermentation issues have thoroughly been dedicated to influence exhibition of curcuminoids and relevant ingredients in turmeric, along with total antioxidant capacity and total phenolic ingredient belonging to fermented turmeric (Mohd Azhar et al., 2017). In the present study we preferred submerged fermentation through grape/hawthorn/apple cider vinegars combination along with *L. plantarum* and *S. boulardii*.

In a prior investigation dual efficacy of wine yeast (*S. cerevisiae* SY) and autochthonous bacteria (*L. plantarum* SCFF107 and *L. plantarum* SCFF200) on cider quality through inoculation methodologies for inducing malolactic fermentation were established. Regarding antioxidant ability, data denoted that apple ciders fermented through simultaneous inoculation method exhibited an elevated radical scavenging rate and total antioxidant capacity. Obtained data showed that ciders exhibited by mixed inoculation with *L. plantarum* modified flavor due to volatiles, non-volatiles and polyphenols ingredient (Chen et al., 2023). In the present study as a standard procedure all fermented curcumin suspensions were uniquely prepared and as was also aforementioned above *L. plantarum* and *S. cerevisiae* strains were preferred. In all cats this fermented curcumin was well tolerated and none of the cats rejected to consume it. This may be partly explained with the data above, as flavor was well accepted probably due to *L. plantarum* ingredient, as reported previously (Chen et al., 2023). At this study all fermented curcumin suspension were given to cats with a 14° degree temperature, which is a preferred temperature for water consumption.

4.2 Proof of Evidence for Human and Animal Studies with or Without Fermentation

In a prior study with 8 obese cats 2 diets prescribed: with hesperidin (0.05%) and naringin (0.1%) from citrus extract vs. with highly bioavailable curcumin from *Curcuma longa* extract (0.09%) for 8-weeks periods. In that study in comparison to pre-research data, citrus polyphenols added diet caused lower plasma AGP and haptoglobin levels, whereas curcumin caused lower plasma AGP concentration. No significant differences were found between groups. In that study it was suggested that hesperidin/naringin or highly bioavailable curcumin extract exhibited have beneficiary effects, treat to target in liver and clinical remission with the obesity-linked inflammation (Leray et al., 2011).

In a prior animal study with the hypothesis of investigating antiobesity effects of turmeric fermented with *L. paracasei* against obesity, orally given 5% *L. paracasei*-fermented and unfermented turmeric accompanied to diet over 16 weeks were comparatively evaluated. In that research

although curcuminoid ingredient of turmeric diminished due to fermentation, it markedly limited weight gain and decreased plasma metabolic parameters. The efficacy of fermented turmeric were more observable in contrast to other relevant unfermented one. Moreover, expression of adipogenesis, lipogenesis, and inflammatory-related protein were all downregulated, with improved insulin and influenced gut microbiota (Lin et al., 2024). In the present study thoroughly for deep discussion on focus of purpose of this study fermod food may have helped metabolic correction (although not analyzed) among cats involved. Moreover, although not data was given decreased weight gain exhibited for all cats involved. On the other side although solely fecal parameters and its morphology were analyzed for both constipated and diarrheic cats we should claim that fermod food presented anti-inflammatory activity. Furthermore although not shown at this study prior to and there after hepatobiliary function analyses through Quantum pet bio resonance device exhibited significantly altered energy production function, detoxification function and the liver fat content of beneficiary to the treated cats.

To those of 60 people with elevated alanine transaminase (ALT) levels (40–200 IU/L, it was hypothesized that fermented turmeric powder might exhibit hepatoprotective efficacy. All 30 participants were administered 2 capsules of fermented turmeric powder 3.0 g/for 3 times daily or placebo ($n = 30$) 3.0 g/daily for 12 weeks. Following 12 weeks of therapy out of 48 subjects included, there was a significant decrease in ALT levels in comparison to with the placebo group ($p = 0.019$ (Kim et al., 2013)). In the present study apart from hepatological protection, gastrointestinal health was 'treat to target' at the focus. Taking into account the primary efficacy endpoint was fecal score alterations; mean and median values were increased in group I and II, whereas was decreased in Group III, as expected (table 1). Comparative interpretation between the pre- and post-treatment scores of each group, a significant difference was detected in Group I ($p = 0.001$), Group II ($p = 0.05$) and Group III ($p = 0.015$) along with marked stool scores. Furthermore, comparatively pre-treatment stool scores between the groups, revealed a significant difference as detected by Kruskal-Wallis test ($p < 0.0001$). regarding the pairwise comparisons (via post-hoc Dunn test), a marked difference was determined between Group I and Group II ($p = 0.006$) and between Group I and Group III ($p = 0.001$). When the stool scores between the groups after treatment were evaluated with the Kruskal-Wallis test, no statistically significant difference was found between the groups ($p = 0.8303$).

5 Conclusions

In conclusion it should not be unwise to draw preliminary conclusion that fermo-food, as submerged curcumin fermentation through grape/hawthorn/apple cider vinegars combination along with *L. plantarum* and *S. boulardii* should have helped protecting gastrointestinal health among cats at least enrolled herein at this study. Someone might also criticise how fermented curcumin be effective against both diarrhea and constipation, the north and the south poles. This was probably linked to the synergistic protection influence of both the curcumin along with the turmeric and the probiotic chains preferred for submerged fermentation (*Lactobacillus* and *Saccharomyces*) capable of diminishing the exhibition of oxidative stress and averting the long-term production of pro-inflammatory cytokines (Yong et al., 2019). Moreover, as a known fact curcumin presented beneficiary effects as demonstrated on diabetic gastroparesis model of rats, in which anti-gastroparetic properties through modification of ghrelin expression, with balanced energy and promoted gastrointestinal motility against oxidative stress (Xu et al., 2013). Aforementioned mechanism of action could reflect efficacy of fermo-food at this study both against diarrhea and constipation, by modification of gastrointestinal motility. Further studies are warranted, which would thus be aim of our subsequent investigation.

Conflict of Interest

The author(s) declare that there is no conflict of interest regarding the publication of this manuscript.

Author Contributions

The study was conceived and designed by K.U. All authors (K.U., H.E., S.E., and S.P.) contributed equally to data collection, analysis, and manuscript writing. All authors have read and approved the final version of the manuscript.

AI and AI-Assisted Technologies Use Declaration

No generative AI tools/AI-assisted technologies were used during the preparation of the manuscript.

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