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Antidiarrheal evaluation of guava leaf infusion in pediatric rats using an intestinal transit model

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ABSTRACT: Diarrhoea is a major gastrointestinal disorder in children and remains a significant cause of mortality, particularly in toddlers. Guava leaves (*Psidium guajava* L.) have long been used in traditional medicine as an antidiarrhoeal remedy. This study aimed to determine the effective dose of guava leaf infusion in weaned rats using the intestinal transit method. Thirty healthy rats were randomly assigned to six groups: a negative control (distilled water), a positive control (atropine sulfate 2.5 mg/kg BW), and four treatment groups receiving guava leaf infusion at doses of 100, 200, 400, and 800 mg/kg BW. Among the treatment groups, the 200 mg/kg BW dose resulted in the greatest reduction in intestinal transit. However, no statistically significant differences were observed between any treatment group and negative control. These results suggest that within the examined dosage range, guava leaf infusion does not demonstrate a significant antidiarrheal effect in paediatric rat models.

Keywords:
antidiarrhoea, infusion, guava leaves, pediatric rats

■ INTRODUCTION

Diarrhoea is one of the most prevalent gastrointestinal disorders in children and remains a major cause of mortality in low- and middle-income countries (Florez *et al.* 2020). In Indonesia, according to the Ministry of Health (2022), diarrhoea accounts for 14% of deaths in neonates (29 days to 11 months) and 10.3% of deaths among toddlers (12 to 59 months), causing a significant health burden.

Guava leaves (*Psidium guajava* L.) have been used in traditional medicine as a natural remedy for diarrhoea. Guava leaves show strong antidiarrhoeal properties (Zulfiana & Fatmawati 2023). Their therapeutic potential has been shown in studies using mouse and rat models (Ojewole *et al.* 2008), providing a scientific foundation for their further development as plant-derived antidiarrheal agents.

Despite promising findings, evidence on guava leaf infusion efficacy in paediatric animal models undergoing gastrointestinal maturation remains lacking. This knowledge gap is crucial for understanding guava leaf therapies in young populations that are distinct from adults. This study aimed to determine the effective dose of guava leaf infusion in weaned rats using the intestinal transit method.

■ MATERIALS AND METHODS

The experimental procedures were approved by the Ethics Committee of School of Veterinary Medicine and Biomedical Sciences (163/KEH/SKE/I/2024). Thirty healthy male Sprague–Dawley rats (*Rattus norvegicus*), three weeks old and 23–40 g, were used. All animals were fasted for 18 h to standardise gastrointestinal conditions.

The rats were allocated to six groups of five animals each. Group 1 received distilled water (1 mL) orally as a negative control, and Group 2 received atropine sulfate (2.5 mg/kg BW, intraperitoneally) as a positive control. Groups 3-6 received 1 mL *Psidium guajava* leaf infusion at extract-equivalent doses of 100, 200, 400, and 800 mg/kg BW. All animals were then administered castor oil (3 mL/kg BW) to induce diarrhoea.

After 30 min, each rat received 0.1 mL/kg BW Chinese ink orally to assess intestinal transit. Forty minutes later, the animals were euthanised and the intestine from the pylorus to the rectum was excised. The total intestinal length and ink

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marker distance were measured. The gastrointestinal transit ratio was calculated as the ink-travel distance relative to the total intestinal length.

Data are expressed as group means and analysed using one-way analysis of variance (ANOVA), followed by Tukey's post hoc test to determine significant differences among treatments.

■ RESULTS AND DISCUSSION

After castor oil administration, the negative control group showed severe diarrhoeal symptoms, confirming its efficacy in inducing diarrhoea in paediatric rats. In the intestinal transit assay, the key parameter was the ratio of the marker distance travelled to the total intestinal length. Atropine sulfate (2.5 mg/kg BW) showed the strongest antimotility effect, with the lowest transit ratio. Guava leaf infusion at doses from 100 to 800 mg/kg BW produced only a modest reduction in intestinal transit (0–12.51%), with a peak effect at 200 mg/kg BW, though not statistically significant compared to other doses or the control group. These results indicate that guava leaf infusion did not exert a significant antidiarrheal effect in paediatric rats. The mean transit ratios for all experimental groups are presented in Table 1.

Castor oil is widely used as a diarrhoeal inducer in experimental models. Its active component, ricinoleic acid, disrupts the intestinal mucosa, induces inflammation, and stimulates prostaglandin release. These mediators increase intestinal motility and secretion, ultimately causing diarrhoea (Bahekar & Kale 2015). Consistent with previous findings, a dose of 3 mL/kg BW effectively induced diarrhoea in the three-week-old rats used in this study.

Studies have shown that guava leaves contain bioactive compounds, such as tannins, flavonoids, and saponins, which are associated with antidiarrheal activity. Guava leaf infusion reduces diarrhoeal symptoms in adult rats at 100–400 mg/kg BW (Ojewole *et al.* 2008) but lacks efficacy in paediatric rats. Quercetin, a major flavonoid in guava leaves, inhibits intestinal contractions through calcium channel antagonism (Kumar *et al.* 2021) and mitigates post-weaning diarrhoea in piglets by modulating NF-κB signalling (Mao *et al.* 2024). These pathways may contribute to the limited effectiveness in paediatric rat models, where acute motility modulation dominates the response.

Atropine administration had a pronounced antidiarrheal effect in paediatric rats. As a competitive antagonist of acetylcholine at muscarinic receptors, atropine reduces gastrointestinal motility and secretion (Khare *et al.*, 2024). In three-week-old rats, this period involves transitioning from maternal milk to solid food and maturing gastrointestinal structures, including cholinergic phenotypes and neuromuscular transmission (de Vries *et al.* 2010; Xu *et al.* 2012). These processes may explain the heightened responsiveness of paediatric rats to atropine compared to guava leaf infusion.

Table 1 Effect of Psidium guajava leaves extract (100-800 mg/kg BW, p.o.) on castor oil-induced intestinal transit in rats.

Treatment group	Doses (mg/kg BW)	Ratio (%)
Negative control (distilled water)	0.0	88.65 ± 6.60[a]
Positive control (atropine sulfate)	2.5	50.94 ± 7.01[b]
Guava leaves extract	100	85.17 ± 7.40[a]
Guava leaves extract	200	76.14 ± 13.13[a]
Guava leaves extract	400	81.82 ± 14.85[a]
Guava leaves extract	800	92.07 ± 5.32[a]

Note: different superscript letters in the same column indicate significant differences ($p<0.05$)

■ CONCLUSION

In summary, guava leaf tea had a small effect on slowing movement in young rats, with the best result at 200 mg/kg body weight. However, it did not protect against diarrhoea caused by castor oil. Atropine works better to stop diarrhoea by blocking certain receptors and slowing gut movement.

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