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Laxative Effects of Aqueous *Senna siamea* Leaf Extract (Yellow Cassia) on the Colon following Loperamide-induced Constipation in Adult Female Wistar Rats

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ABSTRACT

This study investigated the laxative effects of aqueous *Senna siamea* leaf extract (Yellow Cassia), an Asian medicinal plant widely used in Nigerian ethno-medicine, on loperamide-induced constipation in thirty female Wistar rats. The rats were separated into five groups (n=6) to evaluate excretion parameters, gastrointestinal transit time, and intestinal motility. The treatments were carried out as follows; Group I received distilled water (1 ml/500 g bw), Group II received Loperamide (3 mg/kg), Group III received Loperamide (3 mg/kg) and *Senna siamea* (500 mg/kg), Group IV received Loperamide (3 mg/kg) and *Senna siamea* (750 mg/kg), Group V received Loperamide (3 mg/kg) and Bisacodyl 5 mg/kg (Stimulant laxative). Constipation was successfully induced using loperamide for four days, resulting in dry, hard, and reduced rat droppings. Treatment with the *Senna siamea* extract at doses of 500 mg/kg and 750 mg/kg significantly improved the frequency, weight, and water content of the rat droppings, comparable to the reference stimulant laxative Bisacodyl. Consequently, the aqueous *Senna siamea* leaf extract effectively ameliorates the constipative effects of loperamide, supporting its traditional use for gastrointestinal ailments.

Keywords: *Senna siamea*, Loperamide, Bisacodyl, Constipation, GITT (gastrointestinal transit time)

INTRODUCTION

Cassia (now *Senna*) comes from Greek meaning spice, while *siamea* originated from Siam, now Thailand[1]. *Senna siamea* is a non-nitrogen-fixing leguminous tree in the family Leguminosae and sub-family Caesalpinoidea[2,3]. The genus *Cassia*, historically noted for comprising hundreds of species within the Caesalpiniaceae family, holds major economic and dietary value [4]. Specifically, varieties like *Cassia siamea* are indigenous to Southeast Asia, where their edible leaves, seeds, and blossoms are commonly prepared as vegetables in traditional Thai curries [5].

Senna siamea is widely distributed in Africa (Nigeria, Côte d'Ivoire, Eritrea, Ethiopia, Ghana, Kenya, Malaysia, Sierra Leone, South Africa, Tanzania, Togo, Uganda and Zambia) [6]. Globally cultivated and naturalized, *Senna siamea* is known by various names including Bombay blackwood and *Siamese cassia* [7]. Within Nigeria, the plant is predominantly located throughout the southwestern territories and select northern regions, where it is frequently incorporated into local ethnomedicine[8]. *Senna siamea* features prominently in regional Nigerian ethnomedicine and agroforestry. Southwestern communities utilize the leaves (known locally as "ewe kasia" or "Odan") for antipyretic therapies [8], whereas Northern regions utilize the tree, termed "Malga" or "Bakin raskata," for environmental shelterbelts [9]. In particular, Yobe State practices a multi-herbal leaf decoction blending *S. siamea* with citrus, guava, and mango leaves, which is commonly prescribed to combat typhoid and malaria infections [10]. This therapeutic efficacy is supported by compositional analyses, which demonstrate significant concentrations of crude proteins, saponins, alkaloids, anthraquinones, and phytobatannins within the plant matrix [11].

The therapeutic spectrum of *Senna siamea* encompasses the traditional management of

hypertension, diabetes, asthma, insomnia, cancer, and typhoid [12], with historical data validating its leaves and bark as regional antimalarials [8,13]. Its application in treating constipation addresses a global public health concern. This gastrointestinal disorder spans all age groups and socioeconomic classes without demographic boundaries [14]. Because it manifests as irregular and obstructed bowel evacuation, persistent constipation dramatically compromises patient quality of life and introduces risks of broader systemic complications when neglected [14].

The identified risk factors for constipation include: regular use of opioids, family history, public school attendance, poor dietary fiber intake, obesity, and sedentary lifestyles [15]. Symptoms of constipation are reported by 10% to 20% of adults worldwide[16]. Classically, the term "constipation" refers to infrequent bowel motions or hard feces. However, the disorder is heterogeneous; patients report a variety of symptoms including reduced bowel motion frequency, straining, hard stools, the sensation of incomplete emptying, the sensation of anal blockage, or the use of digitation or positioning to aid defecation[17]. In affected individuals, physical health, mental health, and social functioning are reduced [18,19]. Despite this, only one-fifth of constipated individuals seek health-care advice [20]. The high prevalence rate, the healthcare cost and the adverse effect on the quality of life of the sufferers make constipation a huge socio-economic burden on both the individual and the society [21]. Therefore, this research targeted the laxative potential of aqueous yellow cassia (*Senna siamea*) leaf extract, specifically monitoring its therapeutic impact on loperamide-induced colonic constipation using Wistar rats.

MATERIALS AND METHODS

Experimental animals

Thirty non-pregnant female Wistar rats (6-7 weeks old; 150- 200 g) were obtained from the animal house of a local dealer at Narayi community of Kaduna South, Kaduna. Nigeria. The animals were transported down to Zaria and "housed individually in clean metabolic cages placed in a well-ventilated house" (Human Anatomy Department animal house, ABU, Zaria). The animals were acclimatized for 7 days and fed with commercial pellet rat chow and water *ad libitum*.

Plant material sourcing and botanical identification

Yellow cassia leaves were obtained from locally grown shrubs inside Akenzua Hall at Zaria block, Ahmadu Bello University, in April 2024. The plant was verified in Ahmadu Bello University's Botany Department Herbarium, Faculty of Life Sciences, Zaria, by Namadi Sunusi using the ABU06863 voucher number.

Extraction of plant material

The authenticated leaves were washed with clean tap water to remove impurities before being air-dried for 7 days in an open room under primitive conditions at ambient room temperature with air circulation around the plant material to avoid heat and moisture[22]. The dried leaves weighed 350 g, after which they were ground into fine powder with the help of a clean pestle and mortar. It was taken to the Department of Pharmacognosy and Drug Development, ABU, Zaria, where it was macerated by soaking in distilled water for 24 hours and then filtered through filter paper using a funnel, and allowed to settle. The aqueous extract was transferred into an evaporating dish and placed inside the water bath in order to evaporate it to dryness[23].

Drugs and chemicals

Loperamide Capsule (2 mg) (Pramo Life Science Pharmaceuticals, Mumbai, India), Bisacodyl Tablets (5 mg) (Medrel Pharmaceuticals, India, PVT.LTD.), Chloroform, Distilled water, Ethanol, Xylene, Carmine red dye and methylcellulose "from Sigma Chemical Co. (St. Louis, MO, USA)" were purchased from "a" pharmacy store in Samaru, Zaria, Nigeria.

Instruments/Equipment

Glass jar, Glass slides, Cover slips, Beakers (10 ml), measuring cylinder (250 ml). Thirty plastic containers, Plastic test-tubes (Afro-Asia Automobile and Plastics Limited, China), Scalp Vein set (Anhui Kangda Medical Products CO., LTD., China), Ceramic triangle food basins, and improvised metabolic cages were obtained from an equipment store in Samaru, Zaria, Nigeria.

Statement of ethics

Ethical authorization for the use of animal models in this study was granted by the Ahmadu Bello University Ethics Committee on Animal Use and Care, designated by approval code ABUCAUC/2025/116.

Determination of doses and acute toxicity (LD_{50})

Following Lorke's method[24], we conducted the acute toxicity test and selected two extract doses corresponding to 10% and 15% of the LD_{50}.

Experimental design

Constipation induction

To establish the constipation model, Wistar rats received an oral administration of loperamide at a

dosage of 3 mg/kg daily over a 4-day period, following the protocol described by Wintola [25], whereas the control rats received distilled water only. The rats' reduced, hard, and dry droppings showed signs of constipation on the 4th day.

Rats with constipation: grouping and treatment

The rats were separated into five groups of 6 rats per group. The rats in Group I (control) and Group II (constipated control) received 1ml of distilled water orally. Groups III and IV had constipated rats that received 500 mg/kg and 750 mg/kg of *Senna siamea* leaf extract, respectively. Group V received bisacodyl (5 mg/kg body weight). Treatment with extract of yellow cassia and drugs was done via the oral route.

Behavioral/Physical observations

To evaluate physiological changes, the rats' body weights were measured before and after the administration of the aqueous leaf extract of *Senna siamea* (ALESS). Daily, individual rats' fresh droppings/wet droppings were collected, counted, and weighed. After air-drying the samples "at room temperature for 24 hours", they were "reweighed to" calculate "the water content" of droppings [26]. This was computed as follows;

% Water content of rat droppings = Weight of wet droppings − Weight of dry droppings / Weight of wet droppings × 100

In vitro* study on *Senna siamea

To determine its effect on gastrointestinal motility, aqueous leaf extract of *Senna siamea* (ALESS) was evaluated *in vitro* using an organ bath to measure changes in colon contractility. The pharmacological experiment took place in August 2024 at the Department of Pharmacology and Therapeutics, Ahmadu Bello University, Zaria, Nigeria, as outlined by Iyaniwura [27].

Gastrointestinal transit time (GITT)

The intestinal transit assay evaluates gastrointestinal motility and peristalsis [28]. On Day 8, rats received 1ml of an unabsorbable carmine red dye suspension (1.2 g carmine in 20 mL of 0.5% methylcellulose; Fujifilm Wako, Japan) via oral gavage. Gastrointestinal transit time (GITT) was recorded as the duration from administration to the first appearance of dyed rat droppings [29, 30].

Statistical evaluation

Statistical evaluations were executed utilizing GraphPad Prism software (version 10.4.2), with graphical data expressed as mean ± standard error of the mean (SEM). To determine differences across multiple experimental groups, a one-way analysis of variance (ANOVA) was performed, followed by Tukey's post-test to evaluate pairwise significance at a threshold of $p < 0.05$.

RESULTS

Extraction yield of aqueous *Senna siamea* leaf extract (ALESS)

In this study, a powdered sample of yellow cassia leaves (350 g) extracted using aqueous medium yielded 37.78 g corresponding to a percentage yield of 10.8%. This is a better yield compared to the 3.75% obtained from the methanol extract as reported by Ogbiko *et al*.[31] Esievo *et al*.[3] reported that 5.12% of *Senna siamea* was alcohol-soluble, while 16.71% was water-soluble. Gupta and Verma[33] reported the extractive values of *Senna siamea* leaves using the following medium as; Chloroform; 6.24%, Ethyl acetate; 6.92%, Ethanol; 6.04%, Methanol; 5.16%, Aqueous; 12.60%. This higher aqueous yield demonstrates a greater concentration of polar compounds within this *Senna* species, as they extract more effectively in water than in organic solvents.

Acute lethality (LD_{50}) and toxicological screening

Oral administration of aqueous *Senna siamea* leaf extract at a maximum dose of 5000 mg/kg resulted in no mortality or observable toxic effects. Consequently, the oral LD_{50} was determined to be >5000 mg/kg, as shown in Table 1.

Table 1: Oral LD50 of *Senna siamea* leaves

Phase	Dose (mg/kg)	Number of Wistar rats	Mortality rate (after 24 hr of observation)
I	10	**3**	0/3
	100	3	0/3
	1000	3	0/3
II	1600	1	0/1
	2900	1	0/1
	5000	1	0/1

Effects of aqueous leaf extract of *Senna siamea* on body weight and excretion parameters

We investigated the effects of aqueous leaf extract of *Senna siamea* on body weight and excretion parameters in loperamide-induced constipated rats. By evaluating body weight, fecal count, stool weight, and water content, we compared the administration of *Senna siamea* (500 mg/kg) and (750 mg/kg) with the standard laxative Bisacodyl (5 mg/kg). While body weights remained stable across all groups (Figure 1), the constipated rats experienced a decline in stool frequency, weight, and hydration, passing hard, round droppings. Treatment with *Senna siamea* significantly reversed these symptoms, performing comparably to the Bisacodyl group. Together, these findings demonstrate that *Senna siamea* effectively treats constipation by promoting stool excretion (Figures 2, 3, and 4).

***In-Vitro* effects of *senna siamea* leaves on the colon**

Results showed that acetylcholine (0.1, 0.2, 0.4, 0.8 mL), which was the standard drug used, contracted the colon of experimental rats (Figure 5a). On the other hand, the extract (*Senna siamea*) at a concentration of 10mg/ml, 100mg/ml, 500mg/ml and 750mg/ml relaxed the contraction of the colon proportionally to the dosage, as observed in Figures 6 and 7. Introduction of loperamide (1mg/ml) into the organ-bath solution resulted in relaxation of the colon by reducing the amplitude of contraction by almost 50%. Responses elicited by the extract and loperamide are summarized in Figure 7.

Gastrointestinal transit time (GITT)

A significant difference in transit time was observed between the constipated and the control group. It showed that the administration of loperamide + distilled water had a significant effect on the gastrointestinal transit time. *Senna siamea,* on the other hand (at 500 mg/kg and 750 mg/kg), significantly decreased the gastrointestinal transit time proportionally to the dosage compared with the loperamide + distilled water-treated group, as seen in Figure 8.

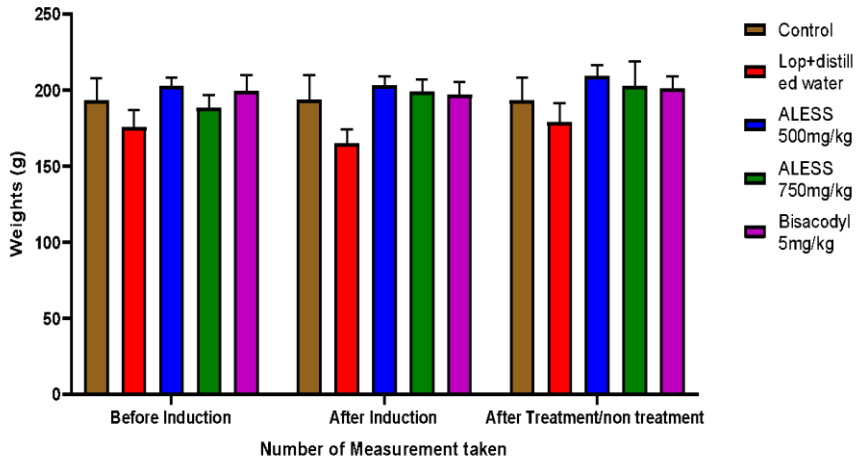

Figure 1: Change in body weights of experimental rats before and after treatment with extract and standard drugs

LOP- loperamide; ALESS- Aqueous leaf extract of *Senna siamea*

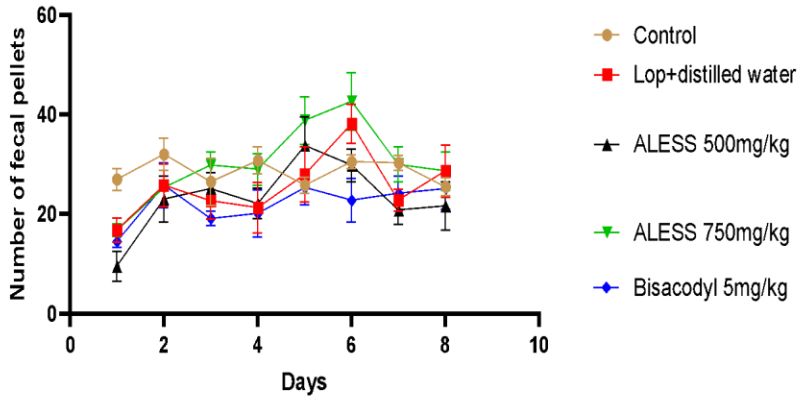

Figure 2: Number of rat droppings across groups for 8 days

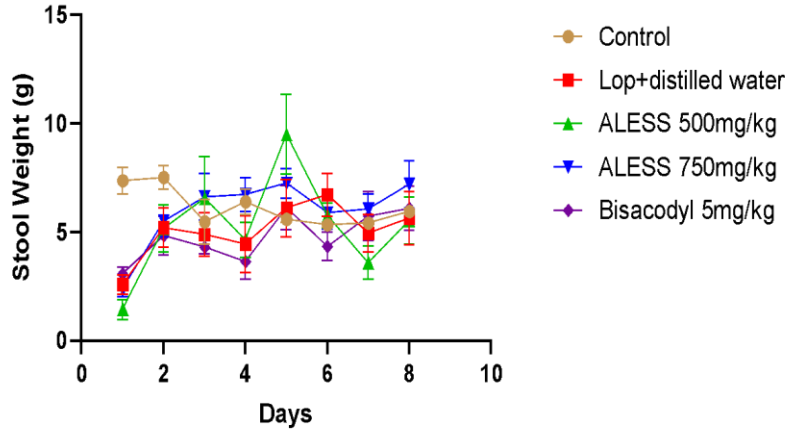

Figure 3: Weight of rat droppings across groups for 8 days

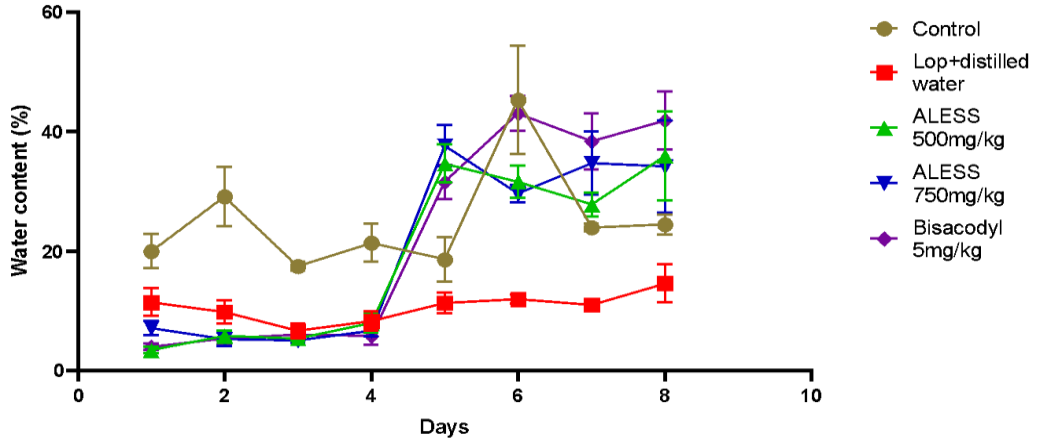

Figure 4: Water content of rat droppings across groups for 8 days.

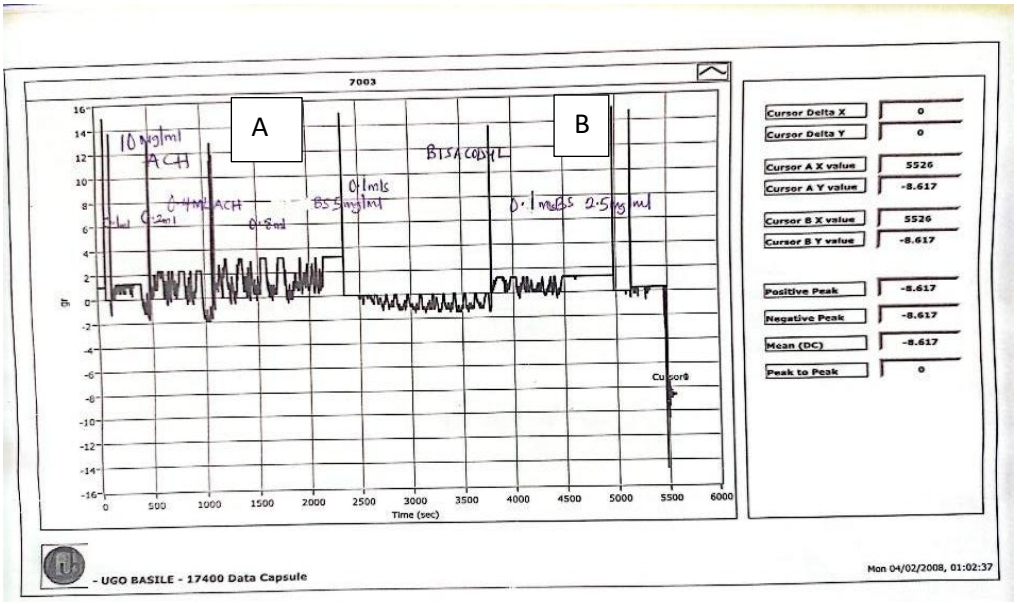

Figure 5a & b: a. Standard solution of ACH at 5 µg/ml concentration of 0.1 ml, 0.2 ml, 0.4 ml and 0.8 ml potentiated the contraction of the colon of an experimental rat in an organ bath. b. Solutions of Bisacodyl at 5 mg/ml and 2.5 mg/ml concentrations of 0.1 ml recorded a protracted phase of colon relaxation.

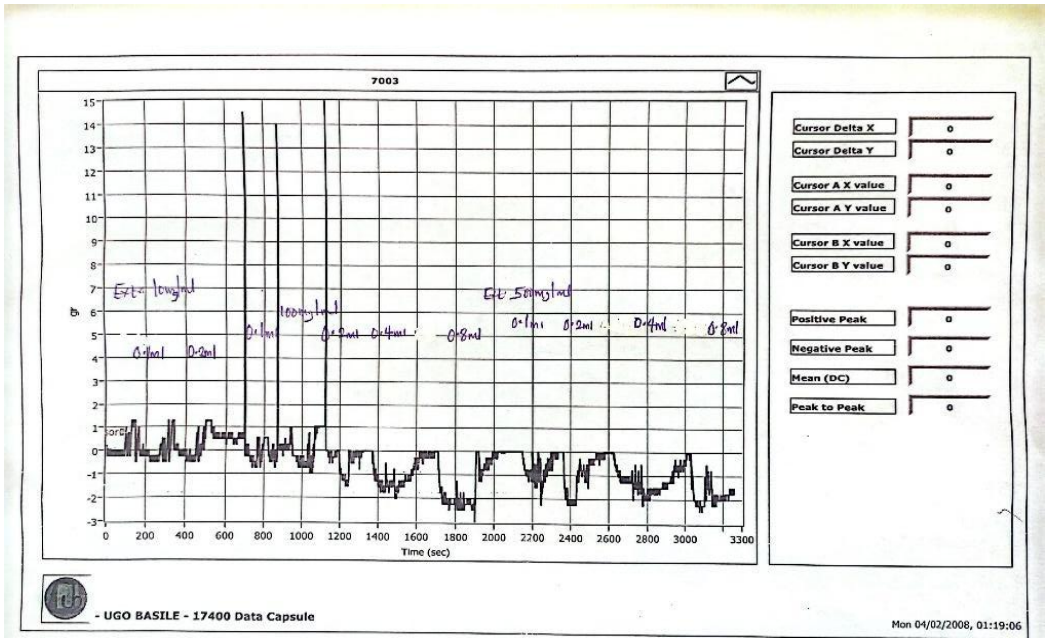

Figure 6: Smooth muscle relaxation of the colon at extract concentrations of 10 mg/ml, 100 mg/ml, and 500 mg/ml.

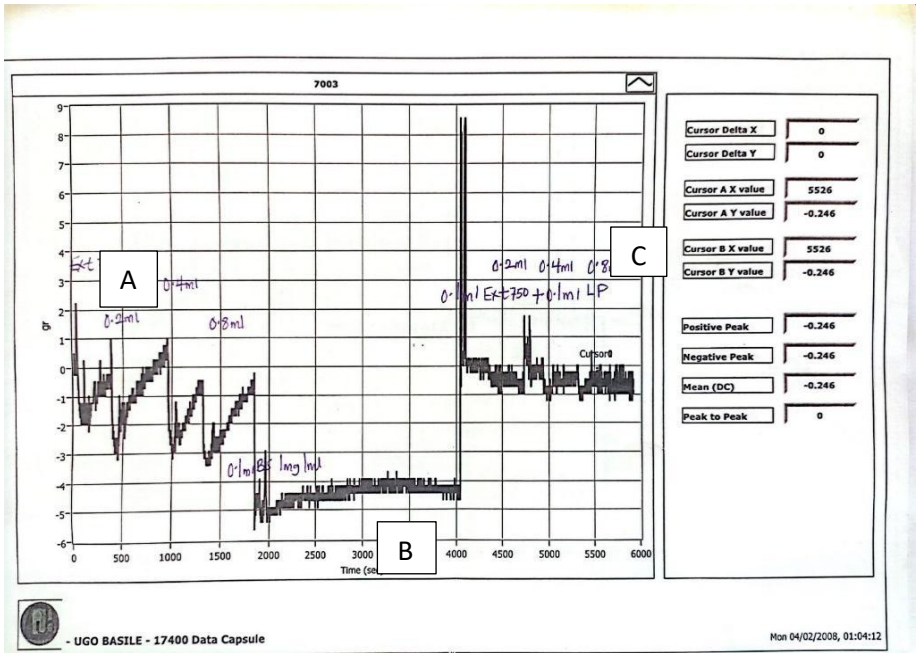

Figure 7a, b, and c: a. Smooth muscle relaxation of the colon at an extract concentration of 750 mg/ml. b. Smooth muscle relaxation of the colon at a Bisacodyl concentration of 1 mg/ml. c. *Senna siamea* interaction with Loperamide at concentrations of 750 mg/ml and 3 mg/ml, respectively.

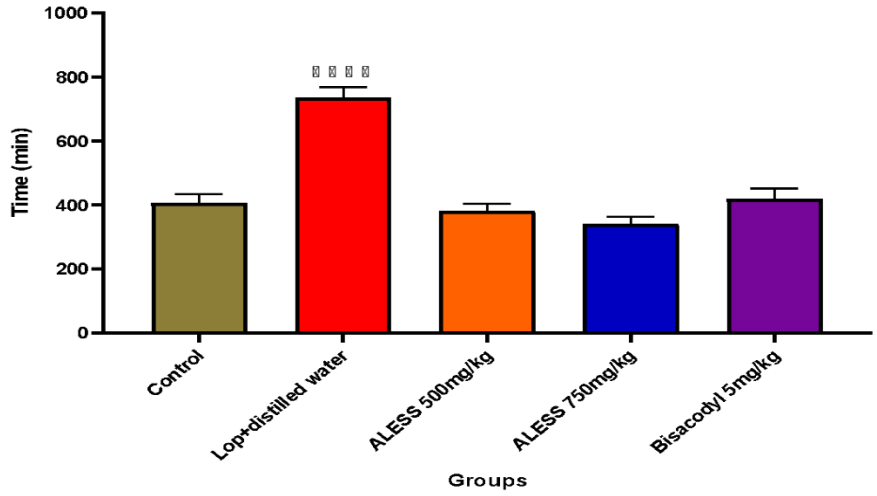

Figure 8: Gastrointestinal transit time across groups. Data analysis (n=6) using a two-way ANOVA with Tukey's post-hoc test revealed that the loperamide + distilled water group differed significantly (****P<0.0001) from the control, bisacodyl (5 mg/kg), and both ALESS (500 and 750 mg/kg) groups.

DISCUSSION

Throughout the acute toxicity testing, administering the aqueous *Senna siamea* leaf extract orally did not produce any mortality or toxic symptoms in the rats, establishing the LD_{50} value as greater than 5000 mg/kg. This is in line with the LD50 of *Senna siamea* as earlier reported [33, 34, 9,] indicating its safety for use without any toxicological effects.

Evaluating the laxative efficacy of aqueous leaf extract of *Senna siamea* in loperamide-induced constipated rats revealed marked improvements in intestinal motility. Treatment led to elevated fecal output, higher stool moisture, and increased stool mass by the eighth day of administration. Maximum defecation frequency over a 12-hour observation window was achieved at the highest extract dose. These findings indicate that *Senna siamea* extract has a laxative effect, successfully reversing loperamide-induced constipation. Similarly, the standard medication, bisacodyl, significantly increased the water content of rat droppings, the weight of rat droppings, and the number of rat droppings. Comparable research on *Mareya micrantha* and *Aloe ferox* leaf extracts also demonstrated increased number of rat droppings, weight, and water content in constipated rats [25, 35]. Compared to the control group, administering an aqueous extract of *L. platyphylla* and gallotannin-enriched G. rhois increased stool output in rats at a dosage of 1000 mg/kg and 250 to 1000 mg/kg, respectively [37, 38]. In the loperamide-induced constipation model, both *Senna siamea* doses produced similar effects on stool parameters, despite slight variations in extract concentrations. Results obtained at the *in vivo* phase of this study demonstrated that oral administration of *Senna siamea* has a lesser or no significant effect on the body weight of experimental animals. *Senna siamea* exerted its laxative effect on the colon as observed in the nature and texture of animal stool. It was observed that the stool size of *Senna-treated* animals appeared normal as compared to stool samples collected from the control. The stool was well-formed and softened, which is attributable to the high moisture content and crude fiber contained in the plant as documented by Ali [9].

During the gastrointestinal motility test, the plant extract accelerated transit of substances inside the gut, demonstrating a significant increase in intestinal movement. Aqueous leaf extract of *Senna siamea* exhibited a significantly lower gastrointestinal transit time at different doses, with a recorded reduced time taken by the carmine red dye to travel through the length of the gut when it is compared to constipated rats. This result is similar to the report of Widjanarko *et al*. [38] on Konac flour (*Amorphophallus muelleri* Blume), another herbal laxative with the same mechanism of action as *Senna siamea*. The laxative effect of *Senna siamea* is mediated by the inhibition of water, Na+, Cl-, and bicarbonate absorption in the transverse colon. Consequently, retained electrolytes and fluids accelerate the movement of stool toward the rectum. By promoting intestinal motility, *Senna siamea* reduces gut transit time. This limits the reabsorption of water and electrolytes in the small intestine, likely by triggering chloride secretion and blocking sodium absorption[39]. Consequently, the moisture of rat droppings increases, further stimulating bowel movements and enhancing the plant's laxative properties. Studies conducted by researchers[40, 41, 42] also suggested that *Senna* may influence intestinal transit time. This further buttresses the fact that *Senna siamea* eased the evacuation of stool in opioid induced constipation, as seen in this study. However, animals treated with loperamide at 3 mg/kg orally craved more water, and their stool was observed to be very dry and reduced (evidence that the animals were constipated), while some animals in the same group did not pass stool at all. The effect of bisacodyl (a stimulant laxative) on the colon was observed to be similar to the effect exerted by *Senna siamea* on the colon (though *Senna siamea* proved to be a better stimulant as demonstrated in this study). This is due to their peristaltic-inducing potential (their potential to induce peristalsis within the colon) and their ability to retain water in the gut. Research by Wintola[25] demonstrated that loperamide significantly slowed intestinal transit time in constipated rats. Conversely, administration of *Aloe ferox Mill.* enhanced gastrointestinal motility in a dose-dependent manner, yielding laxative effects comparable to the standard medication, Senokot.

In the *in vitro* organ bath assay using Tyrode's solution, *Senna siamea* extract induced concentration-dependent relaxation of colonic contractility, while loperamide interacted with *Senna siamea* to reduce the relaxation induced by *Senna siamea*. This result demonstrates that the extract is an opiate (loperamide) inhibitor or opioid blocker and causes decreased contractions. It is likely that the extract exerts its action by specific interference at the receptor level and may not be due to the direct action on colon smooth muscles.

CONCLUSION

Experimental findings indicate that *Senna siamea* mitigates loperamide-induced constipation by promoting intestinal fluid accumulation in the transverse colon, resulting in increased fecal frequency, mass, and water content in rat models.

Conflict of interests: The authors have no conflict of interests to declare.

Author's contribution: SSA, SAM, and SBO conceptualized and designed the study. PAO and AAO collected data, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. SSA, SAM, and SBO revised and edited the drafted manuscript. All authors read and approved the manuscript.

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