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Analgesic & Antidiarrheal Activities Investigation on Methanol Extract of Wendlandia Paniculata : In Vivo Approach

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Abstract:- ***Wendlandia paniculata*** **is under Rubiaceae family and well known as"Mimri", "Lodiannol" which is found throughout Hill Tracks areas of Bangladesh. Chakma Tribe within the Hill Tracts of Bangladesh uses its leaves to treat chest pain. Crushed** ***Wendlandia paniculata*** **leaves are rubbed on the chest to reduce pain. Plant leaves of** ***Wendlandia paniculata*** **have been investigated for the evaluation of biological activities. This study has been subjected to scrutinize pharmacological ideas of methanol extract of** ***Wendlandia paniculata*** **like central and peripheral analgesic and antidiarrheal activities. Evaluation of analgesic activity was applied by following two protocols e.g., tail immersion protocol where Morphine was used as a positive control (centrally) and acetic acid-induced pain sensation protocol in mice (peripherally). The methanol fraction exhibits significant analgesic activity both centrally and peripherally at the following doses of 200 mg/kg and 400 mg/kg body weight of mice. The Anti-diarrheal potentiality was evaluated by using the protocol of Ricinoleic acid (castor oil) induced diarrhea in Swiss albino mice and exhibited anti-diarrheal activity with a significant decrease of diarrheal feculences in a contrast to the standard drug loperamide hydrochloride. The bioassay of the methanol extract of the leaves of** ***Wendlandia paniculata*** **justified the analgesic and anti-diarrheal properties of the methanol extract and finally exhorts the test sample as a provenance of analgesic and anti-diarrheal agents as a crude drug source.**

Keywords**:-** *Wendlandia Paniculata, Central And Peripheral Analgesic, Antidiarrheal, Morphine, Methanol Extract.*

I. INTRODUCTION

Any plant that contains substances that have pragmatic therapeutic motives, or which are harbingers for the synthesis of potential drugs are called medicinal plants. Plants have formed the basis of the traditional medicine system for thousands of years around the world. Medicinal plants contain covetable potential components that are the chemical defense against various diseases, which can hold back plenty amount of pathological conditions and might alter physiological compatibility. New drug development involves the identification of new chemical entities (NCEs) and needs to have the required biological potentiality. Nature is a checked wellspring of discovering lead mixes having anticancer, antimicrobial, pain-relieving, against diarrheal, hypoglycemic, and anti-depressant properties [1]. A part of drug science known as Ethnopharmacology manages the investigation of such traditional sources, especially of plants for the separation and distinguishing proof of bioactive metabolites with wanted remedial properties [1]. The World Health Organization (WHO) has assessed that 80% of the world's occupants depend on customary medication for their essential medical care needs and a large portion of these therapies involved the utilization of plant source or their potential compounds [2].

Wendlandia paniculata having a place with Rubiaceae family and notable as "Mimri", "Lodiannol" is distributed all through the Hill Tracts regions of Bangladesh. The Chakma tribe crushed the leaves and rub on the chest [3]. They are local to Northwestern Vietnam, western Africa, the Indian subcontinent, Southeast Asia, just as subtropical countries of East Asia and northern Australia. There are some reported species of genus Wendland such as *Wendlandia glabrata, Wendlandia tinctoria, Wendlandia angustifalia, Wendlandia heynei, Wendlandia Arabica, Wendlandia andamania*. In the discoveries, recognizable proof of anti-diabetic constituent of the delicate shoots of *W. glabrata* was guided through α-glucosidase hindrance and a potential source for postprandial administration of Diabetes mellitus type 2 [4].

FIGURE I: LEAVES OF *wendlandia paniculata*

FIGURE II: ACCESSION NUMBER OF *wendlandia paniculata*

Pain is an upset and terrible inclination related to damage tissue and causes a person to pull out from a harmful circumstance, which ensures the harmed tissues as the recuperating cycle happens [5]. It is a defensive response involving immune cells and blood vessels [6] The N-methyl-D-aspartate (NMDA) receptor inside the spinal cord dorsal

horn is one of the systems that are engaged with central refinement during pain [7]. Prostaglandin endoperoxide synthase (cyclooxygenase [COX]) is a vital catalyst in the creation of prostaglandins and other eicosanoids. As of late, a second isoform of this chemical (prostaglandin-endoperoxide synthase 2 or COX-2) has been recognized, which is inducible by an assortment of cytokines, chemicals, and tumor promoters [7]. Prostaglandin intervenes in tenderness, pain, and expanded internal body temperature level [8]. COX-1 is a physiologic catalyst liable for the creation of prostaglandins, thromboxane, and prostacyclin engaged with pain inauguration, blood coagulating, and armament of the gastric mucosa. COX-2 is an inducible protein communicated during the upregulation of prostaglandins, for example during irritation in cells [9] . Studies based on the medicinal plant that is used by the local population to reduce pain have gotten reasonable and promising to find fresher pain-relieving medicines [10] Chakma tribe in the Hill Tracts regions of Bangladesh use *Wendlandia paniculata* leaves to treat chest pain. They crush the leaves and rub them on their chest to alleviate pain [3].

Diarrhea is a change in ordinary solid discharge and is delineated by a rise in the water substance, volume, or recurrence of stools. Diarrheal sicknesses are a significant issue in Third World nations and are liable for the passing of millions of individuals every year [11] and it mainly affects children and infants [12]. The utilization of plants and plant derivatives in the treatment of diarrhea is a prevailing practice in Africa. Different plants have been utilized traditionally to treat looseness of the bowels, and in fact, developed data shows that few of these plants are viable [11]. However, the adequacy of a significant number of those anti-diarrheal medications has not been scientifically assessed [13]. Some commonly used medicinal plants that are accustomed to treat diarrhea are *Amaranthus caudatus, Balanites rotundifolia, Boscia coriacea, Cissampelos pareira, Plumbago zeylanica, Solanum hastifolium, Berberis crataegina, Rhamnus cathartica, and Teucrium polium* [14]–[16]. The WHO (World Health Organization) has urged studies relating to the therapy and avoidance of diarrheal infections utilizing traditional clinical practices [13]. The significant effect of loose bowels in agricultural nations including constraints of at present accessible antidiarrheal medications and underprivileged medical services may make conventional drugs a great elective remedy for the administration of diarrhea [13]. The investigation aimed to assess the methanol extract of leaves of *Wendlandia paniculata* as anti-diarrheal agents.

Comprehensive writings search demonstrates that there is no logical proof on the investigations of the plant to affirm its restorative properties. Thusly, in this exploration, the plant *Wendlandia paniculata* was altogether researched for assessment of pharmacological possibilities by In vivo central and peripheral pain relieving, and against loose bowel movements in an appropriate exploratory model.

II. MATERIALS

A. Collection and extraction of plant material

Plant leaves of *Wendlandia paniculata* were gathered from the Hills of Sylhet, Bangladesh in February 2019. The specialists from Bangladesh National Herbarium, Mirpur, Dhaka, taxonomically distinguished the plant and provided the Accession No-48248. The appropriately cleaned leaves became dry through contact with unheated air and ground to an indelicate powder. 500 grams of the pounded powdered material was taken in a spotless, amber-colored bottle (3 liters) and absorbed 2 liters of methanol solvent. The containers with their contents were saved for a time of 15 days joined by successive shaking and mixing. The entire mixture was then sifted through a fresh cotton plug and lastly with a Whatman No.1 filter paper. Using a Buchii Rotavapour, the volume of the filtrate was then decreased at low temperature (400c) by observing pressure. The heaviness of the unrefined concentrate was 30 grams.

B. Drugs and reagents

Reagents and synthetics that had been utilized in this research were of scientific evaluation grade. Some drug materials were purchased from Square Pharmaceuticals Limited (Pabna, Dhaka) and Beximco Pharmaceuticals Limited (Tongi, Dhaka) e.g., ascorbic acid, streptokinase, aspirin, morphine, diclofenac sodium, glibenclamide, fluoxetine, thiopental sodium, castor oil, and loperamide.

C. Experimental Animals

Swiss-albino mice of one or the other sex, matured about 4-5 weeks (weighing around 25–30 grams), got from the Animal Resource Branch of the International Center for Diarrheal Diseases and Research, Bangladesh (ICDDR, B) were used for the experiments. The animals got a standard research facility diet and water (ad libitum) and followed adequate ventilation (Temp 24 ± 2 °C; RH 60–70%) within the space following the traditional (12hour) day-night cycle. 12 hours before and during the experiment, food was withdrawn. All the experiments took place in a separate and quiet state. The animals were adjusted to the exploration research center environment, for 10-12 days before the test. The analyses were accomplished by following the guidelines for the consideration and utilization of laboratory animals. The conventions for leading the tests on the animals were endorsed by the institutional moral panel [17].

III. METHODS (IN VIVO STUDIES)

A. Central analgesic activity

Twelve experimental animals were arbitrarily chosen and separated into four bunches indicated as Group I, Group II, Group III (A-B) comprising of three mice in each bunch. The principal bunch that filled in as negative control getting 1% Tween 80 in ordinary saline (10 ml/kg body weight) .The second bunch that filled in was positive control and was given standard medications for the particular examination. Group III (A) and Group III (B) got oral administration of MESF (Methanol Soluble Fraction) 200 and 400 (mg per kg, body weight) respectively. Group III (A-B) and Group (I-II) got oral administration at zero hours

by feeding to the mice. At the following hour, a few (1-2) cm of the tail of mice was drenched in warm water kept steady at 55^0C. The response time is the time needed by the mice to ricochet their tails. The primary count is disposed of and therefore the response time is documented as a mean of subsequent three perusing. An inertness phase of 20 seconds was characterized as complete pain relive and so the estimation was halted to stay away from injury to mice. The idle time of tail-flick reaction was observed previously and 0, 30, 60, and 90 (an hour and a half) minutes after the dispensation of the medications orally. The time interval of half an hour (30 minutes) was given to guarantee appropriate dispensation of the controlled substances. At that point, morphine solution was dispensed subcutaneously to the mice. Following 30 minutes, an hour, and an hour and a half, the tail submersion time was estimated.

% Time elongation = [(Average estimated time of tail flicking of test Groups - Average estimated time of tail immersing of Group I) / Average estimated time of tail flicking of Group I] X 100

B. Peripheral Analgesic Activity

The peripheral Analgesic response was assessed by the acetic acid-prone writhing technique *[18]*. In this following strategy, acidic acid is dispensed intra-peritoneally to the trial animals to make pain sensation. Thus, the animals spent on the licking and gnawing responses of the infused paws at routine stretches out of pain. However long the animals feel pain, they keep on giving licking and biting responses. Each licking and the biting reaction is considered and taken as a sign of pain sensation. Any substance that has pain-relieving activity should diminish the quantity of licking and biting reactions animals inside a given period and concerning the Control group (Group I). The licking and biting reaction hindrance of positive control (Group II) was taken as standard and contrasted with Group III (A-B) and Control Group I. As a Standard group, acetylsalicylic acid was used as a positive control *[19]*. Twelve exploratory animals were inconstantly chosen and isolated into four bunches meant as Group I, Group II, and Group III (A-B) comprising of three mice in each bunch. Each bunch got a specific treatment. At zero hour test samples, control (1% Polysorbate 80 solution in saline) and Acetylsalicylic acid sodium were directed orally by methods for a long needle with a ball-molded end. Group III (A) and Group III (B) got an oral administration with MESF 200 and MESF 400 (mg/kg, b.w; p.o) separately 30 minutes before intraperitoneal infusion of 1% acidic acid solution at a portion of 10 mL/kg body weight. Following 5 minutes spans, each bunch of mice was noticed for 30 minutes to tally the number of writhing reactions.

Inhibition (%) = [(Control licking response – Test licking response)/ Control licking response] x 100

ll

– Test paw volume

C. In VIVO studies on Anti-diarrheal Activity

The anti-diarrheal effect of the methanol extract of leaves of *Wendlandia paniculata* was assessed utilizing the technique for Ricinoleic acid (castor oil)-actuated motility in mice. [21]. As per this strategy, each mouse was treated orally with 1ml of the exceptionally unadulterated analytical grade of Ricinoleic acid (castor oil) which would instigate loose bowels. The numbers of fecal stools were recorded for singular mice. The trial aftereffects of the experimental groups were contrasted against that of the control with assessing the anti-diarrheal activity of the samples. Twelve test animals were haphazardly chosen, for example, control, positive control, and test groups (two) containing three mice (M-1, M-2, M-3) in each bunch. Each bunch got a specific treatment before any treatment, each mouse was weighed appropriately, and therefore, the doses of the test specimens and control materials were changed as needs are. To administer at dosages of 200 and 400 mg/kg body weight of mice, the precisely measured extracts were estimated individually and ground up in a solitary directional manner by adding some polysorbate-80 (a suspending agent). After appropriate blending of extract and suspending agent, ordinary saline was gradually added. The suspension volume was settled up to 3.0 ml. To settle the suspension, vortex combinator had blended it well. The Control bunch (Group I) administered vehicle (1% Polysorbate 80 in ordinary saline) at the portion of 10 ml per kg orally. The positive control group got loperamide at the portion of 50 mg/kg orally. Every animal was kept in an individual confine; the floor lining was changed each hour. During a perception time of 5 hours, the number of diarrheal appearances discharged by the animals was recorded.

IV. RESULTS

A. Assessment of the central analgesic activity of MESF(Methanolic Soluble Fraction) of leaves of Wendlandia paniculata:

The duration for tail dipping of each mouse was recorded and the normal drenching season of each group was determined. The Percentage (%) of time elongation of tail submerging was determined regarding the control. The higher the elongation level(%) of the group was viewed as the more noteworthy for the analgesic activity of the group. The central pain-relieving potentiality of the test samples was compared in respect to Morphine.

TABLE I: MATERIALS UTILIZED IN THE ASSESSMENT OF CENTRAL ANALGESIC ACTIVITY OF MESF OF LEAVES OF *Wendlandia paniculata.*

Identifier	Test specimens	Group	Identification	Dose (mg/kg) *	Route of administration
CTRL	1% Polysorbate-80 in ordinary saline	I	Control	30 ml	0ral
STND	Morphin	II	Standard	2 mg	Sub-

	e				cutaneous
MESF 1	Methanol Fraction	III A	Trial models	200 mg/kg	0ral
MESF 2	Methanolic fraction	IIIB	Trial models	400 mg/kg	0ral

TABLE II: OBSERVATION OF DATA OBTAINED AFTER 30 MINUTES OF THE DISPENSATION

Animal Group	Immersion time count (seconds)			Average time of immersion	Standard deviation	Standard error	% Elongation
	M-2	*M-3*	*M-1*				
CTRL	2.32	2.12	2.33	2.26	0.1185	0.0684	-
STND	3.93	4.0	4.56	4.16	0.3453	0.1994	84.49
MESF 200	3.67	3.46	3.95	3.69	0.2458	0.1419	63.66
MESF 400	3.86	3.62	4.12	3.87	0.2501	0.1444	71.34

As per the information in the above table, the MESF (Methanol Soluble Fraction) of leaves of *Wendlandia paniculata* at the quantity of 400mg/kg displayed 71.34% (t=10.078,P=0.0005) of restraint and at quantity of 200mg/kg additionally indicated 63.66% (t=9.119,P=0.0008) of hindrance contrasted with the standard medication morphine that shows 84.49% (t=9.046,P=0.0008) of inhibition.

TABLE III: ANALYSIS OF THE DATA OBTAINED AFTER 60 MINUTES

Animal Group	Immersion time count(seconds)			Aver-age time of immersion	Standard deviation (SD)	Standard error (SEM)	% Elongation
	M-1	*M-2*	*M-3*				
CTRL	2.45	2.06	2.6	2.37	0.2787	0.1609	-
STND	7.56	9.38	9.01	8.65	0.9619	0.5554	264.98
MESF 200	4.98	5.01	5.12	5.04	0.0737	0.0426	112.52
MESF 400	5.3	4.75	5.11	5.05	0.2793	0.1613	113.22

In the above table are three data in above table showed that after 60 minutes the central analgesic action of the Methanol soluble fraction of leaves of *Wendlandia paniculata* at a dose of 400 mg/kg and 200 mg/kg became stronger. They showed a percent elongation of 112.52% (t=16.0192, P=0.0001) and 113.22% (t=11.7773, P=0.0003) contrasted with the standard medication morphine that shows 264.98% (t=10.861, P=0.0004) of inhibition.

TABLE IV: OBSERVATION OF DATA OBTAINED AFTER 90 MINUTES OF THE DISPENSATION.

Animal Group	Immersion time count (seconds)			Average time of immersion	Standard deviation	Standard error	% Elongation
	M-1	*M-2*	*M-3*				
CTRL	1.76	2.30	2.48	2.18	0.3747	0.2163	-
STND	14.06	14.25	12.85	13.72	0.7594	0.4384	529.36
MESF 200	6.92	8.02	7.56	7.50	0.5524	0.3190	244.04
MESF 400	6.82	6.36	5.93	6.37	0.4451	0.2570	192.20

The information in the above table the MESF (methanol soluble fraction) of leaves of *Wendlandia paniculata* at the portion of 400mg/kg displayed 192.20% (t=12.4737,P=0.0002) of inhibition and at dosages of 200mg/kg additionally demonstrated 244.04% (t=13.8038, P=0.0002) of hindrance contrasted with the standard medication morphine that show 529.36% (t=24.8713, P<0.0001) of hindrance.

GRAPHICAL REPRESENTATION

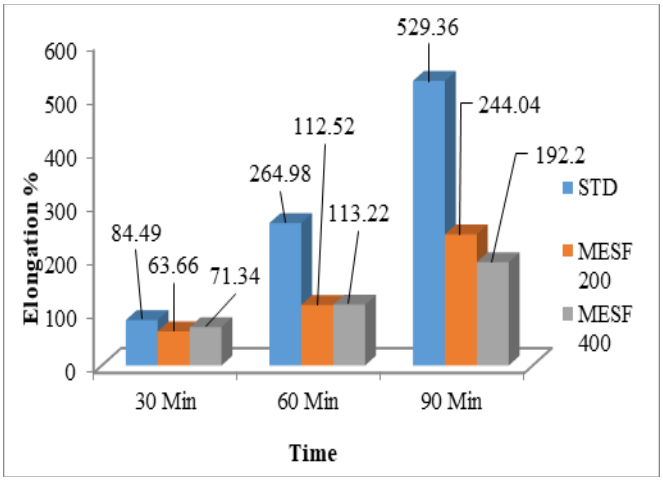

FIGURE III: EXAMINATION OF PERCENT TIME ELONGATION OF TAIL IMMERSION OF DIFFERENT SAMPLES.

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Factual assessment of the information prompts the accompanying end that the MESF (methanol soluble fraction) of *Wendlandia paniculata* showed significant pain-relieving action centrally at the two dosages of 200 mg/kg and 400 mg/kg body weight following 30, 60, and 90 minutes (an hour and a half) of dispensation. Therefore, they can be further investigated for the development of the central analgesic drugs.

B. Assessment of the central analgesic activity of MESF(Methanolic Soluble Fraction) of leaves of Wendlandia paniculata:

The diverse test samples were exposed to screening for pain-relieving action by formalin test. The test was performed by taking samples at portions of 200 mg/kg and 400mg/kg body weight. Formalin 1% was administered at a portion of 0.1ml/10 gram of body weight to each mouse [21]. The outcome was measurably assessed and the t-Test and p values were resolved. Both the test materials displayed exceptionally huge peripheral pain-relieving action while the concentrate at 400mg/kg portion showed the greatest hindrance of writhing 65.57%.

TABLE V: TEST MATERIALS UTILIZED IN THE ASSESSMENT OF PAIN-RELIEVING ACTIVITY OF MESF (METHANOL SOLUBLE FRACTION) OF LEAVES OF *Wendlandia paniculata*

Identifier	Test specimens	Group	Identification	Dose (mg/kg) *
CTRL	1% Polysorbate -80 in ordinary saline	I	Control (Negative Control)	0.1 ml/10 g of body weight
STND	Diclofenac	II	Standard (Positive Control)	50
MESF 1	Methanol soluble fraction	III A	Trial model	400
MESF 2	Methanol soluble fraction	IIIB	Trial model	200

TABLE VI: SCREENING OF ANALGESIC ACTIVITY BY SUBCUTANEOUS ADMINISTRATION OF 1% FORMALIN

Animal Group	licking and biting responses (sec)			Average (sec)	Standard Error	Inhibition (%)
	M-1	*M-2*	*M-3*			
CTL	20	22	19	20.33		-
STD	3	4	3	3.33	0.94	83.61
MESF 200	8	7	9	8.00	1.05	60.66
MESF 400	7	6	8	7.00	1.05	65.57

The above information in the table the MESF (Methanol Soluble Fraction) of leaves of *Wendlandia paniculata* at a portion of 400mg/kg displayed 65.57% (t=12.6491, P=0.0002) of restraint and at dosages of 200mg/kg likewise demonstrated 60.66% (t=11.7004, P =0.0003) of hindrance contrasted with the standard medication Diclofenac that show 83.61% (t=18.0312, P<0.0001) of the inhibition.

GRAPHICAL REPRESENTATION

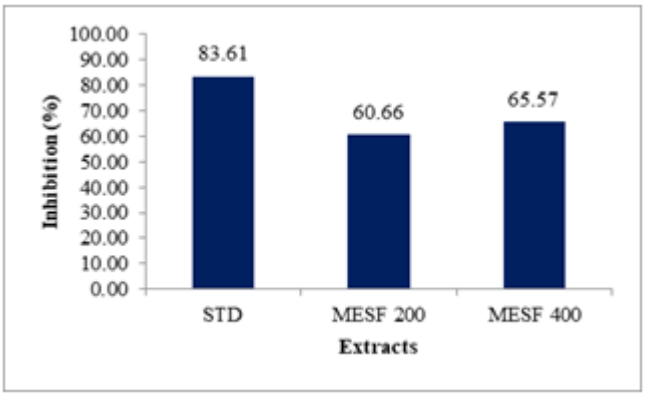

FIGURE 4: CORRELATION OF % INHIBITION OF WRITHING REACTIONS

The diverse test specimens were exposed to screening for pain-relieving activity by acidic acid initiated writhing inhibition strategy. The test was performed by taking samples at portions 400 mg/kg and 200 mg/kg body weight. The outcome was measurably assessed and the t-Test and p esteems were resolved. The test materials display critical peripheral pain-relieving potentiality at both the two portions while the MESF (Methanol Soluble Fraction) at 400 mg/kg portion showed the greatest reduction of writhing at 65.57%.

C. Evaluation of anti-diarrheal potentiality of MESF (Methanol Soluble Fraction) of leaves of Wendlandia paniculata

The MESF (Methanol Soluble Fraction) of leaves of *Wendlandia paniculata* was exposed to castor oil instigated anti-diarrheal test and the accompanying information are gathered.

TABLE VII: TEST MATERIALS UTILIZED IN THE ASSESSMENT OF ANTI-DIARRHEAL ACTION OF MESF (METHANOL SOLUBLE FRACTION) OF LEAVES OF *Wendlandia paniculata*

Identifier	Test specimens	Group	Identification	Dose (mg/kg) *
CTRL	1% Polysorbate-80 in ordinary saline	I	Control	0.1 ml/10 g of body weight
STND	Loperamide	II	Standard	50
MESF 200	Methanol soluble fraction	III A	Trial model	200
MESF 400	Methanol soluble fraction	III B	Trial model	400

TABLE VIII: INFORMATION ADDRESSING THE SUM OF NO. OF DIARRHEAL FECES STOOL GIVEN BY EACH MOUSE

Identifier	Mouse no	No. of diarrheal feces				Sum of diarrheal feces	Avrg.	% Devaluation of diarrhea
		1st hour	2nd hour	3rd hour	4th hour			
CTRL	1	1	3	1	3	8	8.00	
	2	2	2	2	2	8		
	3	3	2	1	2	8		
STND	1	0	0	1	2	3	2.33	70.83
	2	0	0	1	1	2		
	3	0	1	1	0	2		
MESF 200	1	1	1	1	1	4	4.67	41.67
	2	0	2	1	2	5		
	3	0	2	1	3	5		
MESF 400	1	0	2	2	0	4	4.33	45.83
	2	0	2	2	1	5		
	3	0	1	2	1	4		

TABLE 9: EFFECT OF METHANOL EXTRACT OF LEAVES of *Wendlandia paniculate*

Treatment	Dose (b.w.)	%Devaluation of	Standard Error	t-test value	P value	Level of Significance
Control (Saline)	10 ml/kg					
Standard (Loperamide)	5 mg/kg	70.83	0.33	17	<0.0001	Extremely statistically significant
Crude Extract	200 mg/kg	41.67	0.33	10	0.0006	Statistically significant
Crude Extract	400 mg/kg	45.83	0.33	11	0.0004	Very statistically significant

GRAPHICAL REPRESENTATION

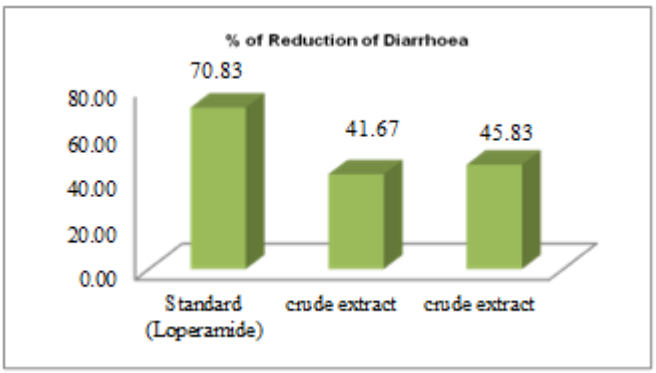

FIGURE 5: ANTI-DIARRHEAL IMPACT OF METHANOL SOLUBLE FRACTION OF LEAVES OF *Wendlandia paniculata* ON CASTOR OIL (1ML/MICE) INITIATED LOOSE BOWELS IN MICE.

The methanol soluble fraction of leaves of *Wendlandia paniculata* showed genuinely critical enemy of diarrheal movement with a 41.67% and 45.83% decrease of diarrhea at a portion of 200mg/kg and 400mg/kg contrasted with the standard loperamide 70.83%.

V. DISCUSSION

In this investigation, the pain relieving and against diarrheal properties of the methanol soluble fraction of leaves of *Wendlandia paniculata* were resolved and the aftereffects of our examination demonstrate huge pain-relieving and anti-diarrheal properties.

In this examination, the Pain-relieving action of *Wendlandia paniculata* was assessed by two tests that were the tail-drenching test for peripheral pain-relieving movement and acidic acid-initiated writhing for central pain-relieving action in the mice model. Standard pharmacological models for the evaluation of analgesia by traditional medicine are both tail immersion and acidic acid incited writhing test [22]. Expanding the limit for pain and modifying the physiological reaction to pain is the activity of centrally acting analgesics. Be that as it may, peripherally acting medications inhibit the production of pain sensation at the chemoreceptor level [23].

This In Vivo study states, the methanol soluble fraction of leaves of *Wendlandia paniculata* at portions of 200 and 400 mg/kg body weight indicated an exceptionally significant ($P < 0.001$) deduction of pain in Swiss albino mice. The acidic acid instigated writhing is suggested for assessing the peripheral pain-relieving action of traditional medicinal plants. Intra-peritoneal dispensation of acidic acid (1%) discloses the inflammation reaction through the creation of prostaglandins particularly PGE2 and PGF2α and histamine in the peritoneal liquid of the trial models [1] In the peripheral analgesic study, the methanol fraction at portions of 200 and 400 mg/kg body weight altogether restrained the licking and biting reaction actuated by the infusion of the acidic acid connotative to the pain-relieving action of *Wendlandia paniculata* by the restraint of pain mediators. As the methanol, soluble fraction of *Wendlandia paniculata* is repressing pain started both centrally and peripherally, it tends to be proposed that they may be acting through both central and peripheral mechanisms. Further phytochemical examinations are warranted to find out the mindful segments.

The methanol soluble fraction of leaf extract significantly ($P < 0.001$) hindered castor oil-incited loose bowels in mice. The castor oil-actuated loose bowels exhibit secretory diarrhea, since an unsaturated fatty acid named Ricinoleic acid, the active ingredient of castor oil, initiates diarrhea by a hyper-secretory reaction [24]. Ricinoleic acid also causes aggravation and irritation of the gut mucosa which prompts improved peristalsis and decreased reabsorption of Na^+ particle, Cl^- ions, and water from the gut which at last incites diarrhea [25]. Since the Methanol concentrate of *Wendlandia paniculata* effectively hindered the castor oil-prompted loose bowels, it tends to be accepted that the antidiarrheal activity was exerted by the anti-secretory component. An investigation report indicates the decrease of the absolute number of wet feces in the trail models in this analysis. Phyto-constituents like alkaloids, terpenes, glycosides, tannins, and flavonoids might present in *Wendlandia paniculata* that enhances the anti-diarrheal potentiality of the plant samples.

VI. CONCLUSION

Our findings indicate that the use of *Wendlandia paniculata* in traditional medicine to treat chest pain is justified. The Methanol Soluble Fraction (MESF) showed the significant antidiarrheal effect on diarrheal condition in trail models. Given these discoveries, it tends to be accepted that *Wendlandia paniculata* could be a likely hotspot for novel 'lead' disclosure for antidiarrheal and pain-relieving drug improvement. Further researches are expected to identify the compound or compounds liable for the noticed impacts. Besides, the research works on it are very limited but have multiple potential effects that can demonstrate its applicable use for better treatment windows in near future.

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REFERENCES

[1] M. Islam, T. Jannat, M. R. Kuddus, M. A. Rashid, and M. R. Haque, “In vitro and in vivo evaluation of pharmacological potentials of Campsis radicans L,” *Clin. Phytoscience*, vol. 5, no. 1, 2019, doi: 10.1186/s40816-019-0144-9.

[2] S. Dhankhar, R. Kaur, S. Ruhil, M. Balhara, S. Dhankhar, and a K. Chhillar, “A review on Justicia adhatoda: A potential source of natural medicine,” *African J. Plant Sci.*, vol. 5, no. 11, pp. 620–627, 2011, [Online]. Available: http://www.academicjournals.org/AJPS.

[3] M. Rahman, S. Uddin, and C. Wilcock, “Medicinal plants used by Chakma tribe in Hill Tracts districts of Bangladesh,” *Indian J. Tradit. Knowl.*, vol. 06, no. 3, pp. 508–517, 2007.

[4] Y. Sheikh *et al.*, “Procyanidin A2, an anti-diabetic condensed tannin extracted from: Wendlandia glabrata, reduces elevated G-6-Pase and mRNA levels in diabetic mice and increases glucose uptake in CC1 hepatocytes and C1C12 myoblast cells,” *RSC Adv.*, vol. 9, no. 30, pp. 17211–17219, 2019, doi: 10.1039/c9ra02397f.

[5] P. Swieboda, R. Filip, A. Prystupa, and M. Drozd, “Assessment of pain: types, mechanism and treatment,” *Ann. Agric. Environ. Med.*, vol. . 1, no. July 2014, pp. 2–7, 2013.

[6] S. Rudra *et al.*, “Biological investigations of the methanol extract of tetrastigma leucostaphylum (Dennst.) alston ex mabb. (vitaceae): In vivo and in vitro approach,” *J. Adv. Biotechnol. Exp. Ther.*, vol. 3, no. 3, pp. 216–224, 2020, doi: 10.5455/jabet.2020.d127.

[7] H. T. Cheng *et al.*, “Inflammatory pain-induced signaling events following a conditional deletion of the N-methyl-d-aspartate receptor in spinal cord dorsal horn,” *Neuroscience*, vol. 155, no. 3, pp. 948–958, 2008, doi: 10.1016/j.neuroscience.2008.06.024.

[8] Y. Oishi, K. Yoshida, T. E. Scammell, Y. Urade, M. Lazarus, and C. B. Saper, “The roles of prostaglandin E2 and D2 in lipopolysaccharide-mediated changes in sleep,” *Brain. Behav. Immun.*, vol. 47, no. December, pp. 172–177, 2015, doi: 10.1016/j.bbi.2014.11.019.

[9] R. N. DuBois *et al.*, “Cyclooxygenase in biology and disease,” *FASEB J.*, vol. 12, no. 12, pp. 1063–1073, 1998, doi: 10.1096/fasebj.12.12.1063.

[10] O. C. Stanley, O. U. Kelechi, and C. S. Chieme, “Preliminary phytochemical screening and gas chromatographic FID evaluation of Garcinia kola seed extracts,” *J. Pharmacogn. Phytochem. JPP*, vol. 115, no. 26, pp. 115–119, 2014, [Online]. Available: http://www.phytojournal.com/vol2Issue6/Issue_feb_2014/25.1.pdf.

[11] S. Meite, J. D. N’Guessan, C. Bahi, H. F. Yapi, A. J. Djaman, and F. Guede Guina, “Antidiarrheal activity of the ethyl acetate extract of Morinda morindoides in rats,” *Trop. J. Pharm. Res.*, vol. 8, no. 3, pp. 201–207, 2009, doi: 10.4314/tjpr.v8i3.44533.

[12] T. T. Tadesse, E. E. Hailu, E. E. Gurmu, and F. F. Mechesso, “Experimental assessment of antidiarrheal and antisecretory activity of 80% methanolic leaf extract of Zehneria scabra in mice,” *BMC Complement. Altern. Med.*, vol. 14, no. 1, pp. 1–8, 2014, doi: 10.1186/1472-6882-14-460.

[13] M. K. Rahman *et al.*, “Studies on the anti-diarrheal properties of leaf extract of Desmodium puchellum,” *Asian Pac. J. Trop. Biomed.*, vol. 3, no. 8, pp. 639–643, 2013, doi: 10.1016/S2221-1691(13)60129-X.

[14] E. Altundag and M. Ozturk, “Ethnomedicinal studies on the plant resources of east Anatolia, Turkey,” *Procedia - Soc. Behav. Sci.*, vol. 19, no. December 2011, pp. 756–777, 2011, doi: 10.1016/j.sbspro.2011.05.195.

[15] B. Abera, “Medicinal plants used in traditional medicine by Oromo people, Ghimbi District, Southwest Ethiopia,” *J. Ethnobiol. Ethnomed.*, vol. 10, no. 1, 2014, doi: 10.1186/1746-4269-10-40.

[16] T. Teklehaymanot and M. Giday, “Quantitative ethnobotany of medicinal plants used by Kara and Kwego semi-pastoralist people in lower Omo River Valley, Debub Omo Zone, Southern Nations, Nationalities and Peoples Regional State, Ethiopia,” *J. Ethnopharmacol.*, vol. 130, no. 1, pp. 76–84, 2010, doi: 10.1016/j.jep.2010.04.013.

[17] M. Zimmermann, "Ethical guidelines for investigations of experimental pain in conscious animals," *Pain*, vol. 16, no. 2, pp. 109–110, 1983, doi: 10.1016/0304-3959(83)90201-4.
[18] Y. N. Dey, S. De, and A. K. Ghosh, "Evaluation of analgesic activity of methanolic extract of Amorphophallus paeoniifolius tuber by tail flick and acetic acid-induced writhing response method," *Int. J. Pharma Bio Sci.*, vol. 1, no. 4, 2010.
[19] M. M. U. Mazumder, A. Sukul, and S. K. Saha, "Analgesic activities of synthesized divalent metal complexes of tolfenamic acid," *Dhaka Univ. J. Pharm. Sci.*, vol. 15, no. 1, pp. 89–96, 2016, doi: 10.3329/dujps.v15i1.29202.
[20] F. G. Shoba and M. Thomas, "Study of antidiarrhoeal activity of four medicinal plants in castor-oil induced diarrhoea," *J. Ethnopharmacol.*, vol. 76, no. 1, pp. 73–76, 2001, doi: 10.1016/S0378-8741(00)00379-2.
[21] A. R. Biswas, S. Ramaswamy, and J. S. Bapna, "Short communication ANALGESIC EFFECT OF," vol. 31, no. 1, pp. 115–118, 1991.
[22] C. A. Hiruma-Lima, J. S. Gracioso, E. J. B. Bighetti, L. Germonsén Robineou, and A. R. M. Souza Brito, "The juice of fresh leaves of Boerhaavia diffusa L. (Nyctaginaceae) markedly reduces pain in mice," *J. Ethnopharmacol.*, vol. 71, no. 1–2, pp. 267–274, 2000, doi: 10.1016/S0378-8741(00)00178-1.
[23] C. Stein *et al.*, "Peripheral mechanisms of pain and analgesia," *Brain Res. Rev.*, vol. 60, no. 1, pp. 90–113, 2009, doi: 10.1016/j.brainresrev.2008.12.017.
[24] S. J. Uddin, J. A. Shilpi, S. M. S. Alam, M. Alamgir, M. T. Rahman, and S. D. Sarker, "Antidiarrhoeal activity of the methanol extract of the barks of Xylocarpus moluccensis in castor oil- and magnesium sulphate-induced diarrhoea models in mice," *J. Ethnopharmacol.*, vol. 101, no. 1–3, pp. 139–143, 2005, doi: 10.1016/j.jep.2005.04.006.
[25] S. Pérez Gutiérrez, D. Zavala Mendoza, C. Soto Peredo, O. Sánchez Sánchez, and M. A. Zavala Sánchez, "Evaluation of the anti-diarrheal activity of Salvia connivens," *Pharm. Biol.*, vol. 52, no. 11, pp. 1467–1470, 2014, doi: 10.3109/13880209.2014.898076.