Journal of Medicinal Plants Studies
www.PlantsJournal.com

ISSN (E): 2320-3862
ISSN (P): 2394-0530
https://www.plantsjournal.com
JMPS 2024; 12(1): 63-69

Received: 08-12-2023
Accepted: 09-01-2024

Neloy Kundu
Pharmacy Discipline, Khulna University, Khulna, Bangladesh

Brototi Chakrabarty
Pharmacy Discipline, Khulna University, Khulna, Bangladesh

Rabindra Nath Acharyya
Pharmacy Discipline, Khulna University, Khulna, Bangladesh

Md. Moklesur Rahman Sarker
Department of Pharmacy, State University of Bangladesh, 77 Satmasjid Road, Dhanmondi, Dhaka, Bangladesh

Md. Abdul Mazid
Department of Pharmaceutical Chemistry, University of Dhaka, Dhaka, Bangladesh

Md. Mustafizur Rahman
Pharmacy Discipline, Khulna University, Khulna, Bangladesh

Corresponding Author:
Md. Mustafizur Rahman
Pharmacy Discipline, Khulna University, Khulna, Bangladesh

Phytochemical screening, analgesic, anti-hyperglycemic and hepatoprotective potentials of *Codariocalyx motorius* (Houtt.) Leaves Extract

Neloy Kundu, Brototi Chakrabarty, Rabindra Nath Acharyya, Md. Moklesur Rahman Sarker, Md. Abdul Mazid and Md. Mustafizur Rahman

DOI: https://doi.org/10.22271/plants.2024.v12.i1a.1627

Abstract
This study was performed on *Codariocalyx motorius* (Houtt.) leaves commonly known as telegraph plant, is a member of the Fabaceae family, to investigate the phytochemical and various pharmacological activities. Qualitative and quantitative phytochemical screenings confirmed the existence of a variety of phytochemical groups. Total phenolic, flavonoid and tannin contents were 81.24 mg gallic acid equivalence/g, 190.90 mg quercetin equivalence/g and 77.29 mg gallic acid equivalence/g, respectively in the extract. In acetic acid-induced writhing model, the extract inhibited the writhing reflex by 49.4% at a dose of 500 mg/kg body weight. In Oral glucose tolerance test, the extract reduced blood glucose levels in a dose-dependent manner in hyperglycemic mice. The extract treated mice also showed the reduction of bilirubin, SGPT, SGOT, and ALP levels in a dose-dependent manner in paracetamol induced hepatoprotective test, when the doses were given of 250 and 500 mg/kg. The outcomes of these experiments support the applications of this plant in conventional medicine.

Keywords: *Codariocalyx motorius*, OGTT, analgesic, hepatoprotective

Introduction
There is significant worldwide interest in researching the effectiveness of plant-based medicines as replacements for nonsteroidal anti-inflammatory drugs (NSAIDs) due to their lower cost and fewer adverse effects [1]. The increasing global prevalence of diabetes mellitus also requires focused attention on effective management of the disease. Hyperglycemia, which can be caused by autoimmune factors (Type 1 diabetes), insulin resistance (Type 2 diabetes), pregnancy, or other factors such as genetics, infections, and medications, can lead to diabetes. The oral glucose tolerance test (OGTT) can be used to measure pre-diabetes, diabetes, and the body's ability to utilize glucose [2]. In addition to diabetes, liver disease is becoming a major threat to health. In developed countries, excessive alcohol consumption and viral-induced chronic liver infections contribute to liver damage, while in underdeveloped countries, hepatotoxic drugs (such as antibiotics, high doses of paracetamol, and carbon tetrachloride) are among the leading causes of toxicity [3]. Despite the advances in modern medicine, there are few effective medications that can protect the liver from cell destruction and the use of conventional drugs in liver disease treatment can sometimes be inadequate with severe side effects [4]. Therefore, alternative treatments for liver disease are needed.
C. motorius family Fabaceae is known as telegraph plant. Traditionally in different region, the plant was used as an antidote to poison, as antidiabetics, cardiac-tonic, wound healing ointment, strengthening the immune system and to treat rheumatism, cough, malaria, pyrexia, dysentery, hepatitis, hemoptysis, physical nerve damage etc. Along with, it has been applied to assuage symptoms by snake-bite poisons. Moreover, the roots are known to have properties of laxative, antidysentric, emollient and also known to heal cough, asthma, rheumatism and fever. The leaves exhibited stimulant, antispasmodic, and febrifuge properties [5].
Synthetic drugs used in the management of pain, [6] diabetes [7] and liver disease [8] disease often have many side effects, while medicinal plants have the potential to treat these diseases with fewer or no side effects [9-10]. According to the World Health Organization, traditional medicinal plants have been found to be effective with low or no toxicity and are therefore excellent candidates for oral use [2].

This study was conducted to determine whether the ethanolic extract of *C. motorius* (EECM) has analgesic, anti-hyperglycemic, and hepatoprotective properties.

Materials and Methods

Preparation of ethanolic extract

The leaves of *C. motorius*, collected from Khulna and the surrounding area, were identified by a specialist at the Bangladesh National Herbarium and given voucher specimen number 45456 DACB. After thorough drying, the leaves were ground into coarse powder. A total of 250 g of the powder was placed in clean glass containers and soaked in 1 liter of 96% ethanol. The containers were sealed and the leaves were allowed to macerate for 14 days with occasional stirring [11]. The filtrate was then dried to obtain a sticky substance (yield = 3.24%).

Chemicals and Reagents

HPLC grade solvents were used throughout the experiment. Standard drugs including diclofenac sodium, glibenclamide were obtained from Beximco Pharmaceuticals Ltd., Dhaka, Bangladesh while Silymarin and paracetamol were obtained from Square Pharmaceuticals Ltd., Dhaka, Bangladesh.

Experimental animal

Swiss albino mice (20-35 g) 4-7 weeks old were obtained from Jahangirnagar University in Savar, Bangladesh. The mice were kept in a dedicated animal lab at Khulna University, Bangladesh, under standard environmental conditions. The care and handling of the mice followed the guidelines of the Animal Ethics Committee at Khulna University (approval number: KU/PHARM/AEC/15/06/34).

Qualitative phytochemical screening

The crude extract was tested for the presence of various phytochemicals using established qualitative phytochemical assay methods [12]. The presence of tannins and flavonoids was tested with the ferric chloride test, alkaloids with Mayer's test, glycosides with Liebermann's test, amino acids with the ninhydrin test, terpenoids with Salkowski's test, saponins with distilled water, steroids with sulphuric acid, and xanthoproteins with nitric acid.

Quantitative phytochemical screening

Determination of total flavonoid content: To determine the total amount of flavonoid, aluminium chloride method was followed [13]. In this test, quercetin was taken as reference standard and the result was represented as quercetin equivalence. Here, 1 mL of various concentrations (1, 0.75, 0.5, 0.25, and 0 mg/mL) of standard and sample extract solution was taken separately into different test tubes. Next, distilled water (4 mL) and 5% w/v $NaNO_2$ (0.3 mL) were added to every test tube which was kept for 5 minutes. After that, 0.3 mL of 10% w/v $AlCl_3$ solution and 2 mL of 1M NaOH were also introduced to each test tube followed by volume adjustment to 10 ml with distilled water. Then the tubes were retained for 15 minutes at ambient temperature and the absorbance, at 510 nm, was noted against blank for individual concentration [14].

Determination of total phenolic contents: Total phenolic content determination was performed by Folin-Ciocalteau Colometry method [15]. In this case, the reference standard was Gallic acid and the finding was represented as gallic acid equivalence [16]. Firstly, 0.5 mL of different concentrations (0.15, 0.1, 0.08, 0.06, 0.04, 0.02 mg/mL) of standard and extract solution was weighed separately into various test tubes. After that, 5 mL Folin-Ciocalteu (FC) reagent (1/10) and 7% Na_2CO_3 were introduced to each test tube. Then the test tubes were kept for 30 minutes under 40 ºC temperature. After 30 minutes, the UV absorbance was calculated at 725 nm against blank for each concentration [14].

Determination of total Tannin contents: In determination of total Tannin content, Folin-Ciocalteau Colometry method was used [17] and Gallic acid was reference standard where the result was represented as gallic acid equivalence [16]. Here, 0.1 mL of various concentrations (0.5, 0.4, 0.3, 0.2, 0.1 mg/mL) of standard and extract sample solution was taken separately in different test tube. Next, distilled water (7.5 mL) and FC reagent (0.5 mL) were added to the test tube. Then, adding 35% Na_2CO_3 (1 mL) to the test tube, the solution was diluted to 10 mL with water. After vortex for 15 seconds, all test tubes were stored at ambient temperature for 30 minutes. At last the absorbance, 725 nm, was calculated against blank for each concentration [14].

Acute toxicity assay: The mice model was used to perform acute oral toxicity assay and Organization for Economic Co-operation and Development (OECD) guidelines-425 was followed with little changes [18]. Mice were separated into five groups (6 mice/group) named as control, test groups-I, II, III and IV. The test groups received the EECM at doses 0.5, 1, 2 and 3 g/kg b. wt. Prior to experiment, weight of individual mouse was recorded carefully. Mice were noticed separately at first 30 minutes after treatment and then at every 24 hr. for 14 days to find out any behavioral changes or mortality from toxicity.

Acetic acid induced analgesic test: Using the acetic acid induced writhing model with slight modification, analgesic activity of the EECM leaves were tested as described previously [1]. Experimental mice were separated into four groups (6 mice/group) denoting group-I, II, III and IV. A particular therapy was given to each group i.e. control (2% tween solution) (I), positive control (25 mg/kg diclofenac sodium) (II) and the group III and IV were given extract at the dose of (250 and 500 mg/kg b. wt.). The test consisted through injection of 0.7% acetic acid. An assessment of writhing was done among positive control, control and test samples.

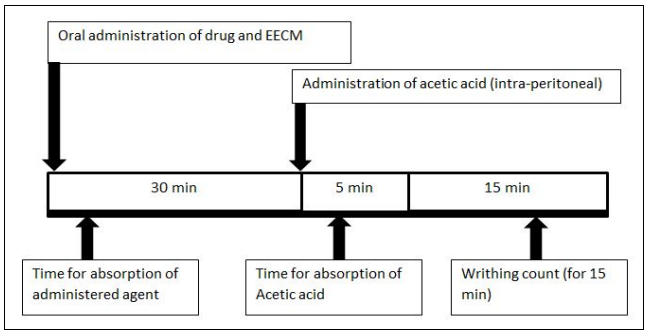

Fig 1: Represented the methodology of analgesic test.

Oral glucose tolerance test (OGTT): OGTT was conducted according to established procedure with minor changes [19]. Twelve hours fasted mice were designated arbitrary and isolated into four groups (6 mice/group). Group-I was given 2% tween solution (0.5 mL/mice); group-II: glibenclamide 10

mg/kg as standard drug and group-III and IV extract at 250 and 500 mg/kg b. wt. correspondingly. Glucose solution was given after 30 minutes, in all groups. The blood glucose content was noted through glucometer and represented in mmol/L. The blood sample was gathered by penetrating the tail and calculated glucose level at 0, 30, 90 and 150 minutes after applying glucose.

Assessment of hepatoprotective activity: Hepatoprotective assessment was conducted according to previously stated method [20] with necessary modifications. Here, we used Swiss albino mice for this investigation. Paracetamol 250 mg/kg was applied to exacerbate the hepatotoxicity and 50 mg/kg b. wt. silymarin was considered as standard hepatoprotective drug. The mice were arbitrarily chosen and isolated into five groups (6 mice /group) namely Group-I, II, III, IV and V. Acute hepatotoxicity was induced by paracetamol treatment through Group-II to V except Group-I. Each group received specific treatment(s) as follows:

- Group I - as control group, which received 2% Tween 80 solution as a daily dose for nine days.
- Group II - as positive control group, which received Paracetamol suspension (250 mg/kg) single dose on 8th day.
- Group III - as Standard Hepatoprotective group, which received Silymarin (50 mg/kg) for nine days + Paracetamol suspension (250 mg/kg) single dose on 8th day.
- Group IV - as Test group 1, which received EECM (250 mg/kg) for nine days + Paracetamol (250 mg/kg) single dose on 8th day.
- Group V -as Test group 2, which received EECM (500 mg/kg) for nine days + Paracetamol (250 mg/kg) single dose on 8th day.

As mentioned above, paracetamol was treated on 8th day as the same process in groups II, III, IV and V. Food was removed 12h before paracetamol application to expedite the acute liver toxicity. Mice were sacrificed on 9th day using minor chloroform anaesthesia and blood samples were taken from cervical vein [21] and centrifuged at 500 rpm for 10 minutes to collect serum which was taken to check the marker enzymes such as SGOT, SGPT and ALP. After dissection the liver and kidney, blood was blotted off and cleaned with saline water and also kept in 10% formalin buffer for histopathological examination. However, we could not perform histopathological examination because of COVID-19 pandemic crisis.

Statistical analysis: Statistical evaluation was done through Student's t-test Calculator and results were presented as Mean ± SEM. P Value < 0.05 was treated as analytically significant.

Results

Phytochemical screening: The outcomes of qualitative phytochemical test of the EECM are represented in Table 1.

Table 1: Qualitative test of ethanolic extract of *C. motorius* leaves

SI. No.	Phytochemical groups	Results
1.	Tannins	+
2.	Flavonoids	+
3.	Alkaloids	+
4.	Glycosides	+
5.	Amino acids	+
6.	Terpenoids	+
7.	Saponins	_
8.	Steroids	_
9.	Xanthoproteins	_

Presence +, Absence -

Total flavonoid content in ethanolic extract of *C. motorius* leaves: The flavonoid content of the *C. motorius* found from calibration curve ($y=0.5144x-0.0002$, $R^2=0.9964$). A calibration curve was driven from various concentrations of quercetin. The content of flavonoids was evolved by QE in mg/g dry weight of EECM and is represented in Table 2. The amount of flavonoids was 190.90±1.59 mg QE/g.

Total phenolic content in ethanolic extract of *C. motorius* leaves: The phenolic content of the *C. motorius* found from calibration curve ($y=8.0935x-0.0315$, $R^2=0.995$). A calibration curve was driven from various concentrations of gallic acid. The phenolic content (81.24±0.18 mg GAE/g) was evolved by GAE in mg/g dry weight of EECM and is represented in Table 2.

Total tannin content in ethanolic extract of *C. motorius* leaves: The tannin content of the *C. motorius* found from calibration curve ($y= 0.502x+0.0732$, $R^2= 0.9626$). A calibration curve was driven from various concentrations of gallic acid. The tannin content (77.29±1.62 mg GAE/g) was evolved by GAE in mg/g dry weight of EECM and is represented in Table 2.

Table 2: Total amount of flavonoid, phenol, and tannin content in ethanolic extract of *C. motorius* leaves

Sample	Total flavonoid content (mg QE/g)	Total phenolic content (mg GAE/g)	Total tannin content (mg GAE/g)
EECM	190.90±1.59	81.24±0.18	77.29±1.62

Acute toxicity effect: No mortality along with any toxic reactions (Table 3) was observed throughout the test at doses up to 3 g/kg, indicating the LD_{50} of the EECM is above 3 g/kg. This finding suggested the assurance of this plant for medicinal purpose.

Table 3: Effects of ethanolic extract of *C. motorius* on behavioural characteristic

Treatment groups	Behavioral characteristic			Mortality	Body weight	
	Food intake	Urination	Diarrhea		Average initial b. wt. (g±SEM)	Average b. wt. after 14 days (g±SEM)
Control (2% tween solution)	Normal	Normal	Absent	0	25.33±0.33	28.17±0.31
Test I (0.5 g/kg b.wt) EECM	Normal	Normal	Absent	0	24.53±0.43	27.17±0.48
Test II (1 g/kg b.wt) EECM	Normal	Normal	Absent	0	25.50±0.43	28.17±0.54
Test III (2 g/kg b.wt) EECM	Normal	Normal	Absent	0	25.83±0.60	28.83±0.54
Test IV (3 g/kg b.wt) EECM	Normal	Normal	Absent	0	26.33±0.42	29.17±0.31

Values were stated as Mean±SEM (n=6). *p < 0.05 comparing with control.

Analgesic activity: The EECM was assayed for analgesic effect against acetic acid induced visceral pain. The outcome of acetic acid prompted writhing reactions in mice proves the analgesic action of extracts represented in (Table 4). It was found that EECM at 250 and 500 mg/kg b. wt. exhibited considerable retardation of writhing reflex by 28.65% and 49.40%, respectively. However, diclofenac sodium was more powerful (65.86% inhibition) than EECM.

Table 4: Effects of *C. motorius* on acetic acid-induced writhing response

Group	Mean of Writhing ± SEM	% Writhing	% Inhibition of writhing
Control (2% tween solution)	27.33±3.25	100.00	0
Positive control (Diclofenac Sodium 25 mg/kg)	9.33±1.54**	34.14	65.86
EECM (250 mg/kg)	19.5±1.18	71.35	28.65
EECM (500 mg/kg)	13.83±2.41*	50.60	49.40

Values were stated as Mean ± SEM (N=6). ** indicates p<0.001 comparing with control, * indicates p<0.05 comparing with control.

Anti-hyperglycemic activity: In OGTT, the EECM at the dose of 250 mg/kg and 500 mg/kg b. wt. showed considerable (p<0.05) drop in blood glucose content comparing to control group mice in dose dependent manner (Table 5).

Table 5: Hypoglycemic effect of extract of *C. motorius* leaves on blood glucose level.

Group	Fasting stage (mMol/L)	30minutes (mMol/L)	90 minutes (mMol/L)	150 minutes (mMol/L)
Group-I (Control)	5.08±0.30	14.44±0.27	9.88±0.21	6.16±0.31
Standard (10 mg)	5.14±0.31	6.9±0.44**	4.92±0.33**	3.62±0.24**
EECM (250 mg/kg)	5.9±0.32	12.58±.73*	7.44±0.43**	5.32±0.29
EECM (500 mg/kg)	5.66±0.14	12.86±0.26*	6.86±0.33**	5.12±0.30*

Values were stated as Mean±SEM (n=6). ** indicates p < 0.001 comparing with control, * indicates p < 0.05 comparing with control.

Hepatoprotective activity

The content of bilirubin, SGPT, SGOT and ALP showed significant (p<0.05) increase in paracetamol treated positive control group (Group-II), comparing with vehicle only treated control group (Group-I). But the EECM treated mice showed considerable (p < 0.05) decrease in the content of bilirubin, SGPT, SGOT, and ALP after comparison with paracetamol only treated group. The maximum reduction was noticed with the higher dose (500 mg/kg b. wt.) which was practically analogous to sylmarin, standard hepatoprotective drug (Table 6).

Table 6: Effect of the ethanolic extract of *C. motorius* leaves on serum Bilirubin, SGPT, SGOT and ALP levels in paracetamol induced hepatotoxicity

Liver function markers	Control (Group-I)	Positive control (Group-II)	Standard (sylmarin) (Group-III)	EECM (250 mg/kg), (Group-IV)	EECM (500 mg/kg), (Group-V)
Bilirubin	0.82±.0293	5.2±0.1317#	1.1±0.086 ***	3.05±0.212 **	1.88±0.1 ***
SGPT	45±6.31	287.83±7.89#	84.33±6.72 ***	216.17±7.96 ***	124±7.73 ***
SGOT	29±0.97	169.5±5.9 #	59±8.87***	155.83±5.08	111.67±5.9 **
ALP	91±7.73	329±14.19#	81.2±9.24 ***	242±7.64 **	162.67±7.67***

Values are stated as mean ± standard error of mean.(n=6) # indicates P <0.0001 comparing with control, *** indicates P< 0.0001 when compared with positive control, ** indicates P< 0.001 when compared with positive control.

Discussion

The presence of secondary metabolites in *C. motorius* extracts, such as tannins, flavonoids, alkaloids, glycosides, amino acids, terpenoids, and phenolic compounds, suggests their potential for biological activity and supports the use of plant-based medicines. Qualitative phytochemical screening in this study identified the presence of tannins, flavonoids, alkaloids, glycosides, amino acids, and terpenoids. In addition, quantitative phytochemical analysis estimated the levels of flavonoids, phenolic compounds, and tannins. Phytochemicals such as tannins, flavonoids, and phenolic

compounds have been shown to have biological activities including analgesic [22], antihyperglycemic [23] and hepatoprotective [24] effects. Based on the traditional uses of *C. motorius* for wound healing, diabetes, and as an antidote to poison, and the presence of phytochemical compounds, we aimed to investigate the analgesic, anti-hyperglycemic, and hepatoprotective activities of the ethanolic extract of *C. motorius* (EECM) leaves through *in vivo* studies. In the analgesic test, we used acetic acid to induce pain. Acetic acid administration stimulates the release of endogenous substances that stimulate nerve endings and cause pain [25-26]. The symptoms of acetic acid-induced pain (intraperitoneal injection) include abdominal muscle contractions and the widening of hind limbs with the extension of the body [27]. Acetic acid has also been shown to increase the production of prostaglandins, particularly the lipoxygenase products PGE2, PGI2, and PGF2α [28-31]. Substances that are able to reduce the number of acetic acid-induced contractions are thought to have analgesic activity [32]. Terpenoids, gums, flavonoids, and tannins are known to have analgesic activity [14]. The qualitative and quantitative analysis of EECM showed the presence of terpenoids, flavonoids, and tannins, which may be responsible for this analgesic activity.

The EECM leaves significantly reduced the number of writhings in a dose-dependent manner. In addition, previous studies on EECM (aerial parts) showed anti-inflammatory action through the inhibition of nitric oxide and prostaglandin E2 release [33]. Flavonoids and tannins extracted from medicinal plants have been known for their significant antinociceptive and/or anti-inflammatory actions (22). Therefore, the analgesic property found with these extracts in the current study may be attributed to its flavonoids and tannins components. In order to investigate a new anti-hyperglycemic drug, our extract was tested for its ability to reduce elevated glucose levels.

Oral hypoglycemic agents should effectively lower blood glucose levels [34]. Flavonoids derived from food have the ability to enhance glucose metabolism [35]. Tannins found in various plants are known to increase glucose consumption through the modulation of insulin signaling pathways, such as pI3K and p38 MAPK, and GLUT-4 translocation. In addition, tannins also exhibit anti-hyperglycemic activity [23]. Alkaloids extracted from medicinal plants have been studied for their potential antidiabetic action in *in vivo* models, which exert their antidiabetic effects through various mechanisms [36]. The EECM (root extract) has also been shown to have anti-hyperglycemic effects in streptozotocin-induced diabetic rats [37].

The EECM leaves showed significant anti-hyperglycemic activity. The extract contains significant amounts of flavonoids, tannins, and alkaloids. The anti-hyperglycemic action of the extract may be due to the presence of these phytochemical components. This result encourages further research to investigate its anti-diabetic action.

Liver damage caused by paracetamol can be used to test the hepatoprotective activity of compounds. In a previous study, ethanolic extracts of *Piper retrofractum* showed hepatoprotective effects against paracetamol-induced acute liver damage in rats [38]. In the current study, the leaves of *C. motorius* were tested for hepatoprotective activity in an *in vivo* model and were found to have a significant hepatoprotective effect in paracetamol-intoxicated models. This effect may be due to the presence of tannins, flavonoids, and phenolic compounds in the extract. Pre-treatment with the extract was able to decrease elevated levels of biochemical factors such as SGPT, SGOT, and ALP. A decrease in the levels of SGOT and SGPT to normal levels is a sign of the restoration of the plasma membrane and the repair of liver tissue damage caused by paracetamol [24]. A previous study of *C. motorius* roots found that they contain highly active antioxidant substances which can be used to treat oxidative stress-related diseases and there is a strong correlation between total phenolic content and the scavenging ability of various reactive oxygen species [39]. Thus, the EECM leaves showed a significant hepatoprotective effect in a dose-dependent manner by reducing high levels of biochemical enzymes when treated with paracetamol. This study supports the traditional use of this plant in the treatment of hepatitis and further investigation is needed to identify the specific mechanisms of action and the phytochemicals responsible for this pharmacological response.

Conclusion

Our study confirms the potential analgesic, oral glucose tolerance, and hepatoprotective effects of the leaves of *C. motorius.* Additionally, this study provides pharmacological evidence for the traditional use of this plant in the treatment of hepatitis through the positive result in the hepatoprotection test and the reduction in oral glucose. It also suggests its potential effects on diabetes. The phytochemical classes present in the leaves, such as flavonoids, alkaloids, glycosides, and amino acids, may be responsible for these effects. Further research is needed to isolate these active compounds and to more fully explore their activities using *in vivo* experimental models in order to validate the findings of this study. This information could be useful in future efforts to develop nutraceuticals and pharmaceuticals using *C. motorius* leaves.

Funding

No funding was available to conduct the research.

Consent of publication

Authors have consent for publication.

Author's contributions

The project was designed by Md. Mustafizur Rahman and Neloy Kundu. Neloy Kundu and Brototi Chakrabarty performed the comprehensive literature research and extraction. Neloy Kundu carried out phytochemical screening, acute toxicity tests, analgesic activity test, OGTT, and hepatoprotective activity assay. Moklesur Rahman Sarker, Rabindra Nath Accaryya and Md. Abdul Maziz conducted data analysis including statistical analysis. Neloy Kundu and Brototi Chakrabarty drafted the article which was first checked by Md. Moklesur Rahman Sarker. Critical revision was made by Md. Mustafizur Rahman and Md. Abdul Mazid.

Ethical approval

The standard guidelines were followed for handling and caring of mice (reference code: KU/ PHARM/ AEC/ 15/06/34).

Acknowledgement

The authors are thankful to the administration of Jahangirnagar University, for supplying the mice. The authors would like to show cordial thanks and respects to Phytochemistry and Pharmacology Research Laboratory, Khulna University, Bangladesh.

References

1. Das BK, Al-Amin MM, Russel SM, Kabir S, Bhattacherjee R, Hannan JMA. Phytochemical screening and evaluation of analgesic activity of Oroxylum indicum. Indian J Pharm Sci. 2014;76(6):571-5.
2. Chakrabarty B, Acharyya RN, Kundu N, Mahmud I, Hossain H, Biswas NN, *et al.* Evaluation of anti-diabetic and anti-allergic activities of *Brownlowia tersa* (L.) kosterm leaves extract and determination of its phenolic compounds by HPLC-DAD. Trop J Nat Prod Res. 2020;4(8):326-33.
3. Saleem TM, Chetty CM, Ramkanth SV, Rajan VS, Kumar KM, Gauthaman K. Hepatoprotective herbs-a review. Int J Res Pharm Sci. 2010 Dec;1(1):1-5.
4. Singh N, Kamath V, Narasimhamurthy K, Rajini PS. Protective effect of potato peel extract against carbon tetrachloride-induced liver injury in rats. Environ Toxicol Pharmacol. 2008;26(2):241-6.
5. De Silva WS, KN Wijesekara. Preliminary screening of telegraph *Codariocal motorius* plant extract for skin whitening property and cytotoxicity activity. 2009;4(7):33-42.
6. Acharyya RN, Mitra K, Risha MA, Hasan MF, Barman AK, Karmakar UK, *et al.* Evaluation of toxicological, anti-allergic, and neuropharmacological activities of *Bridelia stipularis*: *In vivo* and in-silico studies. J. Medicinal Plants. 2022;10(4):113-23.
7. Omodanisi EI, Aboua YG, Chegou NN, Oguntibeju OO. Hepatoprotective, antihyperlipidemic and anti-inflammatory activity of *Moringa oleifera* in diabetic-induced damage in male wistar rats. Pharmacognosy Res. 2017;9(2):182-7.
8. Nayak SS, Jain R, Sahoo AK. Hepatoprotective activity of Glycosmis pentaphylla against paracetamol-induced hepatotoxicity in Swiss albino mice. Pharm Biol. 2011;49(2):111-7.
9. Pant DR, Pant ND, Saru DB, Yadav UN, Khanal DP. Phytochemical screening and study of antioxidant, antimicrobial, antidiabetic, anti-inflammatory, and analgesic activities of extracts from stem wood of Pterocarpus marsupium roxburgh. J Intercult Ethnopharmacol. 2017;6(2):170-6.
10. Luper S. A review of plants used in the treatment of liver disease: Part two. Altern Med Rev. 1999;4(3):178-89.
11. Acharyya RN, Mithila S, Rani S, Islam MA, Golder M, Ahmed KS, *et al.* Anti-allergic and Anti-hyperglycemic Potentials of Lumnitzera racemose Leaves: *In vivo* and In silico Studies. Proc Natl Acad Sci India Sect B - Biol Sci. 2023 Mar;93(1):147-58.
12. Sofowora A. Research on medicinal plants and traditional medicine in Africa. J Altern Complement Med. 1996;2(3):365-72.
13. Zhishen J, Mengcheng T, Jianming W. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. Food Chem. 1999 M;64(4):555-9.
14. Karmakar UK, Akter S, Sultana S. Investigation of antioxidant, analgesic, antimicrobial, and anthelmintic activity of the aerial parts of Paederia foetida (Family: Rubiaceaea). Jordan J Pharm Sci. 2020;13(2):131-47.
15. Tasnim N, Chakrabarty B, Biswas B, Golder M, Kundu P. Exploration of analgesic, laxative, and immunomodulatory effects of leaves and twigs of Euphorbia tirucalli along with in silico analysis. 2024;12(1):1-10.
16. Hasperué JH, Rodoni LM, Guardianelli LM, Chaves AR, Martínez GA. Use of LED light for Brussels sprouts postharvest conservation. Sci Hortic (Amsterdam). 2016;213(1974):281-6.
17. Wolfe K, Wu X, Liu RH. Antioxidant activity of apple peels. J Agric Food Chem. 2003;51(3):609-14.
18. Dev S, Acharyya RN, Akter S, Al Bari MA, Asma K, Hossain H, *et al.* Toxicological screening and evaluation of anti-allergic and anti-hyperglycemic potential of *Sonneratia caseolaris* (L.) Engl. fruits. Clin Phytoscience. 2021;7:1-3.
19. Chakrabarty B, Kundu N, Acharyya RN, Khan A, Islam MA, Dev S, *et al.* Investigation of Anti-hyperglycemic, Anti-allergic activities of ethanolic extract from *Brownlowia tersa* (L.) stem. J Med Plants Stud. 2022;10(1):43-6.
20. Baheti JR, Goyal RK, Shah GB. Hepatoprotective activity of Hemidesmus indicus R. Br. in rats. Indian J Exp Biol. 2006;44(5):399-402.
21. Mahajan SG, Mali RG, Mehta AA. Effect of *Moringa oleifera* Lam. seed extract on toluene diisocyanate-induced immune-mediated inflammatory responses in rats. J Immunotoxicol. 2007;4(2):85-96.
22. Burn JH. Essential pharmacology. Annual Review of Pharmacology. 1969;(1):1-21.
23. Mazumder A, Acharyya RN, Kundu MK, Kundu P, Islam MA, Rahman MM. Phytochemical screening and evaluation of the pharmacological potential of Baliospermum solanifolium leaf extract. Clinical Complementary Medicine and Pharmacology. 2023;100123.
24. Kumar R, Kumar S, Patra A, Jayalakshmi S. Hepatoprotective activity of aerial parts of Plumbago zeylanica linn against carbon tetrachloride-induced hepatotoxicity in rats. Int J Pharm Pharm Sci. 2009;1(SUPPL. 1):171-5.
25. Akhter M, Arefin S, Acharyya RN, Akter H, Jahan S, Rajbangshi JC. Evaluation of Analgesic, CNS Depressant and Antidiarrhoeal Activities of *Psidium guineense* Leaf Extract. Trop J Nat Prod Res. 2021 M;5(3):460-4.
26. Pavao-De-Souza GF, Zarpelon AC, Tedeschi GC, Mizokami SS, Sanson JS, Cunha TM, *et al.* Acetic acid- and phenyl-p-benzoquinone-induced overt pain-like behavior depends on spinal activation of MAP kinases, PI 3K and microglia in mice. Pharmacol Biochem Behav. 2012;101(3):320-8.
27. Bentley GA, Newton SH, Starr J. Studies on the antinociceptive action of α-agonist drugs and their interactions with opioid mechanisms. Br J Pharmacol. 1983;79(1):125-34.
28. Deraedt R, Jouquey S, Delevallée F, Flahaut M. Release of prostaglandins E and F in an algogenic reaction and its inhibition. Eur J Pharmacol. 1980;61(1):17-24.
29. Islam MA, Atanu MS, Siraj MA, Acharyya RN, Ahmed KS, Dev S, *et al.* Supplementation of syringic acid-rich Phrynium pubinerve leaves imparts protection against allergic inflammatory responses by downregulating iNOS, COX-2, and NF-κB expressions. Heliyon. 2023 F;9(2):13343.
30. Le Bel M, Brunet A, Gosselin J. Leukotriene B4, an endogenous stimulator of the innate immune response against pathogens. J Innate Immun. 2014;6(2):159-68.
31. Ribeiro RA, Vale ML, Thomazzi SM, Paschoalato ABP, Poole S, Ferreira SH, *et al.* Involvement of resident macrophages and mast cells in the writhing nociceptive

response induced by zymosan and acetic acid in mice. Eur J Pharmacol. 2000;387(1):111-8.
32. Subedi NK, Rahman SMA, Akbar MA. Analgesic and Antipyretic Activities of Methanol Extract and Its Fraction from the Root of Schoenoplectus grossus. Evidence-based Complement Altern Med. 2016;2016.
33. Kim E, Yoon KD, Lee WS, Yang WS, Kim SH, Sung NY, *et al*. Syk/Src-targeted anti-inflammatory activity of *Codariocalyx motorius* ethanolic extract. J Ethnopharmacol. 2014;155(1):185-93.
34. Zimmet PZ, Alberti G. The Metabolic Syndrome : Perhaps an Etiologic Mystery but Far From a Myth -- Where Does the International Diabetes Federation Stand ? Medscape Diabetes Endocrinol. 2005;7(2).
35. Vinayagam R, Xu B. Antidiabetic properties of dietary flavonoids: A cellular mechanism review. Nutr Metab. 2015;12(1):1-20.
36. Barman AK, Ahmed T, Das H, Biswas B, Ali M, Acharyya R, *et al*. Evaluation of antidiabetic potential of extract of *Sonneratia caseolaris* (L.) Engl. leaves against alloxan-induced diabetes in mice. Trop J Nat Prod Res. 2021;5(1):77-83.
37. Uma C, Suganya N, Vanitha P, Bhakkiyalakshmi E, Suriyanarayanan S, Maria John KM, *et al*. Antihyperglycemic effect of *Codariocalyx motorius* modulated carbohydrate metabolic enzyme activities in streptozotocin-induced diabetic rats. J Funct Foods. 2014;11(C):517-27.
38. Mahaldar K, Hossain A, Islam F, Islam S, Islam MA, Shahriar M, *et al.* Antioxidant and hepatoprotective activity of *Piper retrofractum* against Paracetamol-induced hepatotoxicity in Sprague-Dawley rat. Nat Prod Res. 2020;34(22):3219-25.
39. Chidambaram U, Pachamuthu V, Natarajan S, Elango B, Suriyanarayanan, Ramkumar KM. *In vitro* evaluation of free radical scavenging activity of *Codariocalyx motorius* root extract. Asian Pac J Trop Med. 2013;6(3):188-94.