



RESEARCH ARTICLE

ISSN: 0975-248X
CODEN (USA): IJPSPP



Peptic Ulcer Healing Activity of Fruit Pulp Extract and Seed Oil of *Adansonia digitata* Linn

Kajal Kamalso Malgave*, Nilofar Shahnwaj Naikwade, Padma Laxmikant Ladda,
Tabbasum S. Shikalgar, Sudhir S. Patil

Department of Pharmacology, Appasaheb Birnale College of Pharmacy, Sangli-416416, Maharashtra, India

Copyright © 2019 Kajal Kamalso Malgave *et al.* This is an open access article distributed under the terms of the Creative Commons Attribution-NonCommercial-ShareAlike 4.0 International License which allows others to remix, tweak, and build upon the work non-commercially, as long as the author is credited and the new creations are licensed under the identical terms.

ABSTRACT

Adansonia digitata (AD) Linn has been used to cure PU in Ayurveda but its efficacy has not been validated. The current study was so carried out to evaluate the antiulcer activity of ethanolic extract of *Adansonia digitata* fruit pulp (ADFP), n hexane extract of *Adansonia digitata* seed oil (ADSO) & their combination (ADFP+ADSO) in rats. The effect of AD on gastric ulcer in pylorus ligation induced and ethanol induced models was studied using doses [ADFO (500 mg/kg), ADSO (300 mg/kg) & combination of ADFP & ADSO] for 10 days. Omeprazole (10 mg/kg) were used as the standard drug. Depending on the model, outcomes measures were gastric volume, pH, free acidity, total acidity, ulcer index, percentage inhibition of ulcer index, protein, pepsin, mucus, antioxidant marker enzyme level (Superoxide dismutase, Catalase, Lipid peroxidation), morphological & Histopathological study. The result obtained with combination was set up near to the standard drug and consequence showed that the combination of ADFP & ADSO was found to be more effective than the individual extract of AD. The outcomes were statistically evaluated with the one-way ANOVA followed by the test of Dennett's't'. The secondary-metabolites such as flavonoids, proteins, saponins, tannins, phenols, terpenoids, alkaloids and Fatty acids (palmitic acid, oleic acid, linoleic acid) are potent as antioxidant, antiulcer and anti-inflammatory. The finding of this reading confirmed that AD has antiulcer activity due to 1 or more of the secondary-metabolites present in it. Therefore, this study validates its antiulcer use in Ayurveda. Future investigation on separation of specific phytochemicals and elucidate MOA are needed.

Keywords: Peptic ulcer, *Adansonia digitata* Linn, Seed oil, Pulp, Antioxidants.

DOI: 10.25004/IJPSDR.2019.110518

Int. J. Pharm. Sci. Drug Res. 2019; 11(5): 263-271

*Corresponding author: Ms. Kajal Kamalso Malgave

Address: Department of Pharmacology, Appasaheb Birnale College of Pharmacy, Sangli-416416, Maharashtra, India

Tel.: +91-9764378107

E-mail ✉: kajalmalgave75@gmail.com

Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Received: 16 August, 2019; **Revised:** 08 September, 2019; **Accepted:** 10 September, 2019; **Published:** 25 September, 2019

INTRODUCTION

In the world peptic ulcer is major gastrointestinal disorders. Annually, peptic ulcer perforation rate was 3.81 - 14 and haemorrhage rate was 19.4 - 57 per 100,000 people. [1] It is a chronic disease which may leads serious and life threatening complications such as

bleeding and/ or perforation that may necessitate surgery and if untreated it can cause death. This disease represents worldwide health problem due to its high morbidity, mortality, economical loss. [2] Over 90% of peptic ulcers are caused by bacterial infection (*H. pylori*) or by use of NSAIDS. [3] Other factor such as

mucosal damaging mechanisms (Mucus, bicarbonate, synthesis of PG & Nitric oxide). [4] In Indian pharmaceutical industry shares 6.21 billion rupees for antiulcer drugs and these drugs occupy 4.31% of the market share. [1] Synthetic drugs produce several side effects. Such as, proton pump inhibitors (Omeprazole, Lansoprazole) may cause nausea, abdominal pain, constipation, diarrhea and H₂ receptor antagonists (Cimetidine) may causes libido loss because of these severe side effects, herbal medicines are considered as safe for use and better alternatives for peptic ulcer therapy. [5] Decrease in gastric acid juice and increase in production of gastric mucosa were the main strategies for the management of peptic ulcer. Therefore demand of synthetic drugs is increased and proposing newer options for prevention/treatment of peptic ulcer. The types of drugs vary from proton pump inhibitor to H₂ antagonist and at the same moment, these synthetic drugs produce some adverse drug reaction like arrhythmias, gynaecomastia, and haemopoetic changes. Because of these ADR there is need to thought of better natural alternative of peptic ulcer. [6] *Adansonia digitata* Linn is a large tree, commonly known as Baobab. It is used in many ways; every part of AD is stated to be useful. [7-8] Chemical constituents of this plant were reported to have flavonoids, steroids, vitamins, fatty acids, terpenoids, besides tannin. [9-10] Antioxidants have protective role in gastric ulcers. [11] *Adansonia digitata* is high in antioxidants [12-13] in recent year, flavonoids, terpenoids, tannins, vitamin C, fatty acids shows antisecretory, antiulcer and ulcer healing activity in many research paper. [14-16] Therefore *Adansonia digitata* Linn is selected for evaluation of antiulcer activity based on its reported phytoconstituents of fruit of *Adansonia digitata* Linn.

MATERIALS AND METHODS

Preparation of extraction

Fruit of *Adansonia digitata* Linn were collected in month of September and November 2018 from Miraj region (Dist - Sangli) Maharashtra (India) and Authenticated by Prof. M. D. Wadmare, Department of Botany from Smt. Kasturbai Walchand College Sangli. The fruit were dried at room temperature. Then seeds were separated from fruit pulp and seeds were ground to a coarse powder using mixer grinder. The powders were stored in airtight container. Weighed quantity of pulp powder and seed powder with ethanol and n- hexane respectively was subjected to extraction using Soxhlet apparatus at [60-70°C], [60-65°C] boiling point of solvent respectively, then solvent was allowed to evaporate at RT. [17-18] The final extract was weighed and the % of extract was calculated by formula:

Percentage extract obtained = $\frac{\text{Wt. of extract (g)}}{\text{Wt. of sample (g)}} \times 100$

Phytochemical investigation

Tests were carried out to get the presence of constituents viz flavonoids, saponins, glycosides,

alkaloids, carbohydrate, tannins, phenols in fruit pulp and proteins, steroids, calcium, fatty acids in seed oil.

Animals

Adult albino Wistar rats of either sex, weighting 150 - 200 g obtained from animal house of Appasaheb Birnale College of Pharmacy Sangli, were used experiment. Form B protocol were prepared and submitted to Institutional Animal Ethics committee (IAEC). Animals were kept in well- ventilated room at $23 \pm 2^\circ\text{C}$, with humidity of 65-70%, and they were feed with a standard pellet diet with mineral water. Procedures involving laboratory animal were done in accordance with CPCSEA guidelines. Approval for animal use was obtained from IAEC prior to experimental study. The experimental protocol (IAEC/ABCP/4/2018-19) was approved by the IAEC.

Acute Toxicity study in rat (LD₅₀ determination)

According to literature survey the LD₅₀ of ADFP and ADSO was reported previously as 5000 mg/kg and 3000 mg/kg respectively. [19-20]

Dose selection

According to OECD guidelines as the LD₅₀ of ADFP and ADSO was found to be 5000 mg/kg and 3000 mg/kg respectively, then one dose is selected for further evaluation as 1/10th i.e. 500 mg/kg and 300 mg/kg respectively. Combination group was given 250 mg/kg of fruit pulp extract & 150 mg/kg of seed oil.

Experimental Design

Pyloric ligation in rat

Adult albino rats of Wistar strain of either sex weighing 150-200 g were used for the study. All the animals were divided into the seven groups with 6 animals in each group.

Normal: DW 10 ml/kg *p.o.*

Control I: DW 10 ml/kg + Pylorus ligation *p.o.*

Control II: Tween 80 5 ml/kg Pylorus ligation *p.o.*

Standard: Omeprazole 20 mg/kg +Pylorus ligation *p.o.*

Test I: Fruit pulp 500 mg/kg (For 10 days) +Pylorus ligation *p.o.*

Test II: Seed oil 300 mg/kg (For 10 days) +Pylorus ligation *p.o.*

Test III: [Fruit pulp (250 mg/kg) + seed oil (150 mg/kg) (For 10 days)] +Pylorus ligation *p.o.*

Test sample was given for a period of 10 days. On day 10, once the last dose of test sample, the rats were kept for 24 hours fasting. On day 11the animal were anesthetized by using Thiopentone sodium. The stomach had been opened and pylorus was ligated without causing any injury to its blood vessels. The stomach was put and the stomach had been closed with sutures. All animals were sacrificed by CO₂ euthanasia technique 19 hours after ligation. Then stomach was dissected out and the matter of stomach have been gathered into tube and centrifuged. Supernatant was collected for analysis of gastric volume, pH, free acidity, total acidity, protein, pepsin, mucus in gastric juice. Further the stomach was incised along the greater curvature and the inner surface was examined for ulceration by giving score number and severity was

assessed microscopically. The stomach tissue is then homogenate into phosphate buffer using tissue homogenizer; this homogenate was used for estimation of different antioxidant markers. [21-23]

Ethanol induced ulcer in rat model

Adult albino rats of Wistar strain of either sex weighing 150-200 g were used for the study. All the animals were divided into the seven groups with six animals in each group.

Normal: DW 10 ml/kg *p.o.*

Control I: DW 10 ml/kg + Ethanol (5 ml/kg) *p.o.*

Control II: Tween 80 5 ml/kg Ethanol (5 ml/kg) *p.o.*

Standard: Omeprazole 20 mg/kg + Ethanol (5 ml/kg) *p.o.*

Test I: Fruit pulp 500 mg/kg (For 10 days) + Ethanol (5 ml/kg) *p.o.*

Test II: Seed oil 300 mg/kg (For 10 days) + Ethanol (5 ml/kg) *p.o.*

Test III: [Fruit pulp (250 mg/kg) + seed oil (150 mg/kg) (For 10 days)] + Ethanol (5 ml/kg) *p.o.*

Test sample was given for a period of 10 days. On day 10, after the last dose of test extract, the rats were kept for 24 hours fasting. On day 11th ethanol were administered at dose 5 ml/kg and after 1 hr. animals were sacrificed by CO₂ chamber. Then stomach was removed out and contents were taken into clean tubes & centrifuged. Supernatant was collected for measuring parameters. Further stomach of all animals was cut with the greater-curvature and the internal surface was examined for ulceration by giving score number and severity was assessed microscopically. The stomach tissue is then homogenate into phosphate buffer using tissue homogenizer; this homogenate was used for estimation of different antioxidant markers. [21, 24-25]

Parameters was determined

Determination of gastric volume & pH, determination of free, total acidity in gastric juice, determination of gastric protein content, determination of total pepsin, determination of gastric mucus content determination of ulcer index, morphological study of stomach, determination of mucous in gastric content, estimation of antioxidant markers [Superoxide dismutase, Catalase, Lipid peroxidation i.e. Malondialdehyde (MDA)], Morphological study, Histopathological study.

Statistical Analysis

Data was expressed as Mean $\pm$ SEM. The comparisons between the averages of series of values were performed by one way ANOVA test followed by Dunnett's 't' test.

RESULTS

In both models the result on the effect of ADFP, ADSO & combination of ADFP + ADSO reveals that the extract have significantly ($p < 0.001$) reduced the ulcer index. Ulcer index was severe in control group and decreased in test group Maximum inhibition of ulcer index was observed in the animal treated with the combination group. Administration of fruit extract significantly ($p < 0.001$) decreased level of offensive factors such as gastric volume, free acidity, total acidity, ulcer index, protein, pepsin, while increased level of pH, gastric mucus. The level of total protein in the stomach was increased in control group, when compared to test group. Administration of Omeprazole also reduced protein content in the stomach juice. The activities of antioxidant enzymes such as SOD, catalase decreased in control group while lipid peroxidation increased & treatment with fruit extract and Omeprazole increased SOD, catalase level while decreased lipid peroxidation.

Morphological study in both model showed that the stomach integrity was maintained in normal group and control group showed severe bleeding, perforation, spot ulcer, streaks but standard group and extract treated groups showed extremely fewer ulceration and stomach integrity was maintained. Microscopical change of pylorus ligation induced & ethanol induced model. In Control group, gastric mucosa showed damaged gastric mucosal epithelium, glands with hemorrhages, edema & inflammatory cells infiltration in the submucosal layer. The test groups showed significant regeneration of mucosal layer and significantly prevented the hemorrhages, edema and severity of damage to mucosal epithelium as compared to control. In all over parameters result obtained with combination was set up near to the standard drug and consequence showed that the combination of ADFP & ADSO was found to be more effective than the individual extract of AD.

Table 1: Effect of ADFP and combination of ADFP+ADSO on Gastric volume, pH, Free acidity, Total acidity and Ulcer index in Pylorus ligation induced ulcer.

Treatment & Dose	Gastric volume (ml)	pH	Free Acidity (mEq/L/100 g)	Total Acidity (mEq/L/100 g)	Ulcer Index
Control (10 ml/kg bw)	6.033 $\pm$ 0.09	3.458 $\pm$ 0.011	59 $\pm$ 0.856	73 $\pm$ 0.816	16.17 $\pm$ 0.333
Standard (20 mg/kg)	3.417 $\pm$ 0.04**** (43.36%) $\downarrow$	6.232 $\pm$ 0.004**** (44.51%) $\uparrow$	34 $\pm$ 0.365**** (42.37%) $\downarrow$	52 $\pm$ 0.365**** (28.76%) $\downarrow$	4 $\pm$ 0.182**** (75.26%) $\downarrow$
ADFP (500 mg/kg)	4.633 $\pm$ 0.03**** (23.20%) $\downarrow$	4.885 $\pm$ 0.008**** (29.21%) $\uparrow$	41.50 $\pm$ 0.763**** (29.66%) $\downarrow$	56.33 $\pm$ 0.33**** (22.83%) $\downarrow$	6.83 $\pm$ 0.166**** (75%) $\downarrow$
ADFP + ADSO (250 mg/kg)	4.200 $\pm$ 0.03**** (30.38%) $\downarrow$	5.578 $\pm$ 0.007**** (38%) $\uparrow$	37.67 $\pm$ 0.557**** (36.15%) $\downarrow$	54.50 $\pm$ 0.50**** (25.34%) $\downarrow$	5.33 $\pm$ 0.278**** (67.01%) $\downarrow$
+ 150 mg/kg)					

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean $\pm$ SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test ($n = 6$). **** $p < 0.0001$ Considered Significant when compared with Control.

Table 2: Effect of ADSO and combination of ADFP+ADSO on Gastric volume, pH, Free acidity, Total acidity and Ulcer index in Pylorus ligation induced ulcer.

Treatment & Dose	Gastric volume(ml)	pH	Free Acidity (mEq/L/100 g)	Total Acidity (mEq/L/100 g)	Ulcer Index
Control	6.033 ± 0.09	3.435 ± 0.009	63.83 ± 0.401	72.33 ± 0.802	15.17 ± 0.307
1 ml/kg Tween 80					
Standard	3.417 ± 0.04**** (43.36%) ↓	6.23 ± 0.004**** (44.88%) ↑	34 ± 0.365**** (46.73%) ↓	52 ± 0.365**** (28.10%) ↓	4 ± 0.1826**** (73.63%) ↓
ADSO	4.633 ± 0.03**** (23.20%) ↓	4.71 ± 0.166**** (27.07%) ↑	46 ± 0.365**** (27.93%) ↓	63.17 ± 0.90**** (12.66%) ↓	6.917 ± 0.20**** (54.40%) ↓
ADFP + ADSO	4.200 ± 0.03**** (30.38%) ↓	5.57 ± 0.007**** (38.41%) ↑	37.67 ± 0.557**** (40.98%) ↓	54.50 ± 0.50**** (24.65%) ↓	5.33 ± 0.278**** (64.84%) ↓

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control.

Table 3: Effect of ADFP and combination of ADFP + ADSO on Protein, Pepsin and Mucus in Pylorus ligation induced ulcer.

Treatment & Dose	Protein (µg/ml)	Pepsin (µg/ml)	Mucus (mg/ml)
Control (10 ml/kg bw)	104.2 ± 0.222	79.33 ± 0.421	44.81 ± 0.316
Standard (20 mg/kg)	47.47 ± 0.658**** (54.44%) ↓	48.83 ± 0.401**** (38.44%) ↓	109.6 ± 0.393**** (59.11%) ↑
ADFP (500 mg/kg)	68.05 ± 0.755**** (34.69%) ↓	53 ± 0.365**** (33.19%) ↓	101.1 ± 0.311**** (55.67%) ↑
ADFP + ADSO	52.76 ± 0.684**** (49.36%) ↓	51.17 ± 0.477**** (35.49%) ↓	105.5 ± 0.732**** (57.52%) ↑

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control.

Table 4: Effect of ADSO and combination of ADFP + ADSO on Protein, Pepsin and Mucus in Pylorus ligation induced ulcer.

Treatment & Dose	Protein (µg/ml)	Pepsin (µg/ml)	Mucus (mg/ml)
Control (1 ml/kg Tween 80)	103.3 ± 0.851	76.67 ± 0.494	49.17 ± 0.270
Standard (20 mg/kg)	47.47 ± 0.658**** (54.04%) ↓	48.83 ± 0.401**** (36.31%) ↓	109.6 ± 0.3938**** (55.13%) ↑
ADSO (300 mg/kg)	71.10 ± 0.396**** (31.17%) ↓	59.83 ± 0.542**** (21.96%) ↓	99.04 ± 0.168**** (50.35%) ↑
ADFP + ADSO	52.76 ± 0.684**** (48.92%) ↓	51.17 ± 0.477**** (33.25%) ↓	105.5 ± 0.732**** (53.39%) ↑

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control.

Table 5: Effect of ADFP and combination of ADFP + ADSO on SOD, Catalase and MDA in Pylorus ligation induced ulcer.

Treatment & Dose	SOD (U/mg of protein)	Catalase (U/mg of protein)	MDA (nmol/mg of protein)
Normal	53.58 ± 0.525	35.67 ± 0.333	3.915 ± 0.216
Control (10 ml/kg bw)	18.41 ± 0.453####	15.50 ± 0.428####	13.37 ± 0.115####
Standard (20 mg/kg)	52.13 ± 0.322****	31.33 ± 0.760****	7.867 ± 0.098****
ADFP (500 mg/kg)	43.26 ± 0.302****	24.33 ± 0.333****	11.53 ± 0.126****
ADFP + ADSO	46.24 ± 0.371****	27.17 ± 0.600****	9.483 ± 0.104****

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control. # indicates control group compared with normal.

Table 6: Effect of ADSO and combination of ADFP + ADSO on SOD, Catalase and MDA in Pylorus ligation induced ulcer.

Treatment & Dose	SOD (U/mg of protein)	Catalase (U/mg of protein)	MDA (nmol/mg of protein)
Normal	53.58 ± 0.525	35.67 ± 0.333	3.915 ± 0.216
Control (1 ml/kg Tween 80)	18.65 ± 0.2150####	16.67 ± 0.494####	13.45 ± 0.129####
Standard (20 mg/kg)	52.13 ± 0.322****	31.33 ± 0.760****	7.867 ± 0.098****
ADSO (300 mg/kg)	37.74 ± 0.222****	22.50 ± 0.806****	12.04 ± 0.080****
ADFP + ADSO	46.24 ± 0.371****	27.17 ± 0.600****	9.483 ± 0.104****

DISCUSSION

Peptic ulcer is because of imbalance between aggressive-factors and defensive factor. To recover the balance, the use of therapeutic agents prevents gastric juice discharge to enhance the processes of mucosal protection by increasing mucosal output. [26] The current research was conducted to assess the anti-ulcer

effect of oral administration of ADFP extract (500 mg/kg) and ADSO extract (300 mg/kg) and the mixture of ADFP (250 mg/kg) and ADSO (150 mg/kg) of *A. digitata* Linn against gastric mucosal ulceration induced experimentally by Pylorus ligation in rats and ethanol.

Table 7: Effect of ADFP and combination of ADFP + ADSO on Gastric volume, pH, Free acidity, Total acidity and Ulcer index in Ethanol induced gastric ulcer

Treatment & Dose	Gastric volume (ml)	pH	Free Acidity (mEq/L/100 g)	Total Acidity (mEq/L/100 g)	Ulcer Index
Control (10 ml/kg b/w)	5.217 ± 0.1046	3.572 ± 0.025	49.50 ± 0.428	69.50 ± 0.223	15 ± 0.258
Standard (20 mg/kg)	3.150 ± 0.042**** (39.58%) ↓	6.147 ± 0.007**** (41.89%) ↑	36.50 ± 0.428**** (26.26%) ↓	49.17 ± 0.477**** (29.25%) ↓	4.33 ± 0.166**** (71.13%) ↓
ADFP (500 mg/kg)	4.133 ± 0.033**** (20.77%) ↓	5.233 ± 0.144**** (31.74%) ↑	39.17 ± 0.166**** (20.86%) ↓	56.50 ± 0.428**** (18.70%) ↓	6.83 ± 0.166**** (54.44%) ↓
ADFP + ADSO (250 mg/kg + 150 mg/kg)	3.667 ± 0.033**** (29.71%) ↓	5.192 ± 0.026**** (31.20%) ↑	41.17 ± 0.703**** (16.82%) ↓	53.50 ± 0.018**** (23.02%) ↓	5.58 ± 0.153**** (62.80%) ↓

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control.

Table 8: Effect of ADSO and combination of ADFP + ADSO on Gastric volume, pH, Free acidity, Total acidity and Ulcer index in Ethanol induced gastric ulcer.

Treatment & Dose	Gastric volume (ml)	pH	Free Acidity	Total Acidity	Ulcer Index
Control (1 ml/kg Tween 80)	5.467 ± 0.033	3.483 ± 0.009	52.33 ± 0.76	72.17 ± 3.005	15.67 ± 0.27
Standard (20 mg/kg)	3.150 ± 0.042**** (42.38%) ↓	6.147 ± 0.007**** (43.38%) ↑	36.50 ± 0.42**** (30.25%) ↓	49.17 ± 0.47**** (31.86%) ↓	4.333 ± 0.16**** (72.34%) ↓
ADSO (300 mg/kg)	4.367 ± 0.049**** (20.12%) ↓	4.693 ± 0.04**** (25.78%) ↑	45.50 ± 0.42**** (13.05%) ↓	58.33 ± 0.55**** (19.17%) ↓	7.333 ± 0.21**** (53.20%) ↓
ADFP + ADSO (250 mg/kg + 150 mg/kg)	3.667 ± 0.03**** (32.92%) ↓	5.192 ± 0.02**** (32.91%) ↑	41.17 ± 0.70**** (21.32%) ↓	53.50 ± 0.56**** (25.86%) ↓	5.583 ± 0.15**** (64.37%) ↓

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control.

Table 9: Effect of ADFP and combination of ADFP + ADSO on Protein, Pepsin and Mucus in Ethanol induced ulcer.

Treatment & Dose	Protein (µg/ml)	Pepsin (µg/ml)	Mucus (mg/ml)
Control (10 ml/kg bw)	107.0 ± 1.06	79.82 ± 0.275	55.50 ± 0.562
Standard (20 mg/kg)	48.54 ± 0.230**** (54.63%) ↓	48.71 ± 0.244**** (38.97%) ↓	141 ± 0.516**** (60.63%) ↑
ADFP (500 mg/kg)	64.78 ± 1.168**** (39.45%) ↓	53.34 ± 0.285**** (33.17%) ↓	123.3 ± 0.421**** (54.98%) ↑
ADFP + ADSO (250 mg/kg + 150 mg/kg)	53.17 ± 0.591**** (50.30%) ↓	51.70 ± 0.326**** (35.22%) ↓	138.3 ± 0.421**** (59.86%) ↑

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control.

Table 10: Effect of ADSO and combination of ADFP + ADSO on Protein, Pepsin and Mucus in Ethanol induced ulcer.

Treatment & Dose	Protein (µg/ml)	Pepsin (µg/ml)	Mucus (mg/ml)
Control (1 ml/kg Tween 80)	104.9 ± 0.239	78.08 ± 0.405	51.67 ± 0.421
Standard (20 mg/kg)	48.54 ± 0.230**** (53.72%) ↓	48.71 ± 0.244**** (37.62%) ↓	141.0 ± 0.516**** (63.35%) ↑
ADSO (300 mg/kg)	67.99 ± 0.584**** (35.18%) ↓	56.61 ± 0.299**** (27.49%) ↓	130.0 ± 0.632**** (60.25%) ↑
ADFP + ADSO (250 mg/kg + 150 mg/kg)	53.12 ± 0.591**** (49.36%) ↓	51.70 ± 0.326**** (33.78%) ↓	138.3 ± 0.421**** (62.63%) ↑

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control.

Table 11: Effect of ADFP and combination of ADFP + ADSO on SOD, Catalase and MDA in Ethanol induced ulcer.

Treatment & Dose	SOD (U/mg of protein)	Catalase (U/mg of protein)	MDA (nmol/mg of protein)
Normal	53.58 ± 0.525	35.67 ± 0.333	3.915 ± 0.216
Control (10 ml/kg Dw)	18.14 ± 0.520####	17 ± 0.3651####	13.17 ± 0.084####
Standard (20 mg/kg)	48.47 ± 0.355****	32.67 ± 0.333****	8.100 ± 0.148****
ADFP (500 mg/kg)	40.90 ± 0.366****	25.17 ± 0.4014****	11.26 ± 0.065****
ADFP + ADSO (250 mg/kg + 150 mg/kg)	43.48 ± 0.588****	27.83 ± 0.3073****	9.683 ± 0.060****

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control. # indicates control group compared with normal.

Yield of ethanolic extract ADFP was 12.5% and n hexane extract of ADSO was 22.5% thus supporting that ethanol & n-hexane solvent possesses a good extracting potential. In the preliminary phytochemical

screening, the fruit pulp was positive for flavonoids, steroids, proteins, amino acids, tannin, vitamins, terpenoids. Seed oil having protein, lipids, vitamin B₁, Fatty acids (palmitic acid, oleic acid, linoleic acid). [9]

Table 12: Effect of ADSO and combination of ADFP + ADSO on SOD, Catalase and MDA in Ethanol induced ulcer.

Treatment & Dose	SOD (U/mg of protein)	Catalase (U/mg of protein)	MDA (nmol/mg of protein)
Normal	53.58 ± 0.525	35.67 ± 0.333	3.915 ± 0.216
Control (1 ml/Tween 80)	16.94 ± 0.233####	17 ± 0.3651####	12.70 ± 0.051####
Standard (20 mg/kg)	48.47 ± 0.355****	32.67 ± 0.333****	8.100 ± 0.148****
ADSO (300 mg/kg)	35.54 ± 0.742****	25.17 ± 0.4014****	19.95 ± 0.117****
ADFP + ADSO (250 mg/kg + 150 mg/kg)	43.48 ± 0.588****	27.83 ± 0.3073****	9.683 ± 0.060****

ADFP: *Adansonia digitata* fruit pulp, ADSO: *Adansonia digitata* seed oil; All the values were expressed as a Mean ± SEM. The results were analyzed statistically by the one-way analysis of variance (ANOVA) followed by Dunnett's test (n = 6). ****p<0.0001 Considered Significant when compared with Control. # indicates control group compared with normal.

In pylorus ligated model – Ulcer is caused by ligation of the pyloric end at the beginning of small intestine that can cause gastric acid to accumulate in the stomach. This is due to the gastric mucosa auto digestion leading to a breakdown of the gastro mucosal barrier. Therefore, an increase in accumulation of acid-pepsin due to pylorus blockade may trigger later digestion of the mucosa. [24] In pylorus ligation model the result indicated that the Omeprazole treated group showed 73.63% protection of ulceration and average percentage decrease in gastric volume, free acidity, total acidity, pepsin, protein, 43.36%, 46.73%, 28.10%, 36.31%, 54.04% respectively and average percentage increased in the pH, gastric mucous content 44.88%, 55.13% respectively in pylorus ligation model. The ethanolic extract of ADFP (500 mg/kg) were administered for 10 days which showed 59.25% protection of ulceration and average percentage decrease in gastric volume, free acidity, total acidity, pepsin, protein, 19.77%, 29.66%, 22.83%, 33.19%, 34.69% respectively and average percentage increased in the pH, gastric mucous content 29.21%, 55.67% respectively in pylorus ligation model. 10 days oral pretreatment with n-hexane extract of ADSO (300 mg/kg) showed 54.40% protection of ulceration and decrease in level of gastric volume, free acidity, total acidity, pepsin, protein, 23.20%, 27.93%, 12.66%, 21.96%, 31.17%, respectively and increased in the level of pH, gastric mucous content 27.07%, 50.35% respectively in pylorus ligation model. Combination group [ethanolic extract of ADFP (250 mg/kg) + n-hexane extract of ADSO (150 mg/kg)] result was set up near to the standard drug & has got better and more effective result than individual fruit extract of *Adansonia digitata* Linn. It means when combination extract was administered which gives maximum protection of ulceration & significantly decreased the level of ulcer index (64.84%), gastric volume (30.38%), free acidity (40.98%), total acidity (24.65%), pepsin (33.25%), protein (48.92%) & increases level of pH (38.41%), gastric mucous content (53.39%). It means the result obtained with combination was set up near to the standard drug and consequence showed that the combination of ethanolic extract of ADFP & n-hexane extract of ADSO was found to be more effective than the individual extract of *Adansonia digitata* Linn. The concentration of flavonoids and vitamin C is rich in fruit pulp, vitamin C is Natural antioxidant. [7] Reduced catalase and SOD are considered as antioxidant enzymes which are responsible for the antioxidant

activities (scavenging and disposal of free radicals from the tissues).

Activities of reduced catalase and SOD enzymes was reduced in the current research while LPO level high in ulcerated animal, whereas the activities of SOD (52.13 U/mg of protein), CAT (31.33 U/mg of protein), enzymes were increased & LPO (12.70 nmol/mg of protein) level decreased in the animals which were treated with standard drug. Then administered ethanolic extract of ADFP showed increase in SOD (24.33 U/mg of protein), CAT (43.26 U/mg of protein) while LPO (9.267 nmol/mg of protein) level decreases as compared to control group. While n-hexane extract of ADSO contains Vitamin E which having antioxidant property & it showed increase in antioxidant enzymes i.e. SOD (22.50 U/mg of protein) & CAT (37.74 U/mg of protein) while LPO (9.667 nmol/mg of protein) level decreases as compared to control group but the results of individual extract of ADFP & ADSO are lesser than combination of ADFP & ADSO. Antioxidant enzyme level in combination group was increased i.e., SOD (27.17 U/mg of protein), CAT (46.24 U/mg of protein) while LPO (11.95 nmol/mg of protein) level decreases as compared to control group. This observation clearly indicated that the fruit extract of *Adansonia digitata* Linn are more capable of enhancing the antioxidant-activities. This act may be due to the presence of antioxidant phytochemicals in the extract of *Adansonia digitata* Linn. Morphological study in both model indicated that the normal group, stomach integrity was maintained and appeared normal. In experimental control group, perforation, several bleeding, spot ulcer, streaks were observed. The gastric mucosa was appeared intact in the stomach of Omeprazole treated animals. The gastric mucosa was seen to be slightly damaged with red coloration, spot ulcers in the stomach of ethanolic extract of ADFP, n-hexane extract of ADSO & in Combination of both extract treated groups. Scoring of ulcer was made as follows;

Normal coloured stomach (0)

Red coloration (0.5)

Spot ulcer (1)

Hemorrhagic ulcer (1.5)

Deep ulcer (2)

Perforation (2.5)

Microscopical change of pylorus ligation induced & ethanol induced model were showed in below, Histopathological changes in experimental control group showed damaged mucosal epithelium,

hemorrhage, edematous appearance of the gastric tissue whereas ethanolic extract of ADFP, n hexane extract of Seed oil and combination of both had less mucosal damage to epithelium observed.

Normal

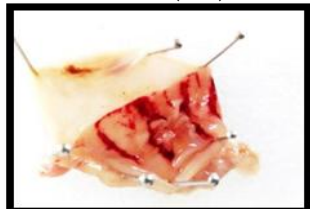


Pylorus ligation induced gastric ulcer Control (DW)

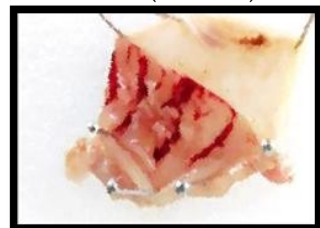


Control (Tween 80)

Ethanol induced gastric ulcer Control (DW)



Control (Tween 80)



Standard



Standard



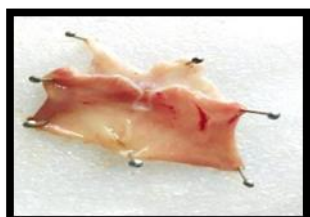
ADFP



ADFP



ADSO



ADSO



ADFP + ADSO



ADFP + ADSO

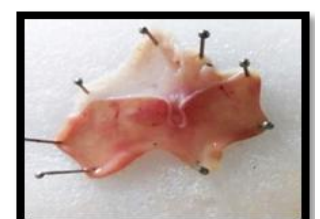
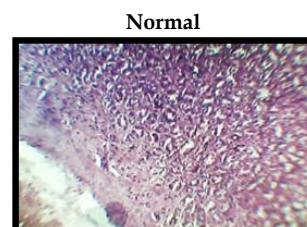
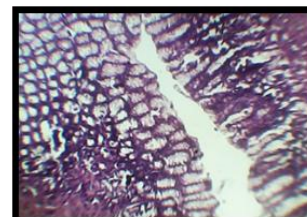


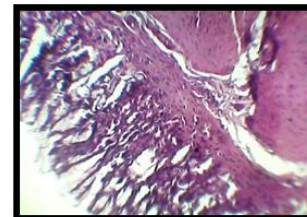
Fig. 1: Morphological study of stomach in Pylorus ligation induced ulcer and Ethanol induced ulcer.



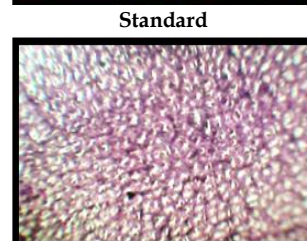
Normal



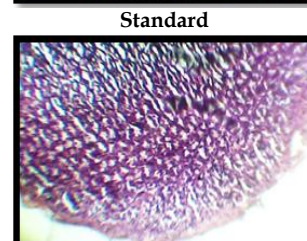
Control



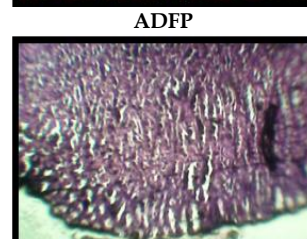
Control



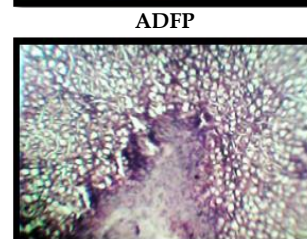
Standard



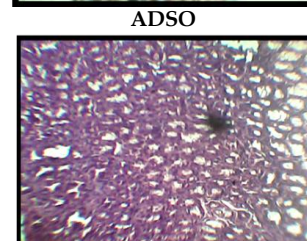
Standard



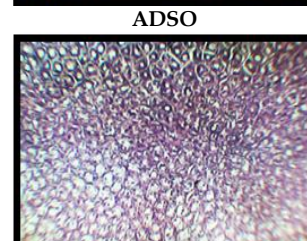
ADFP



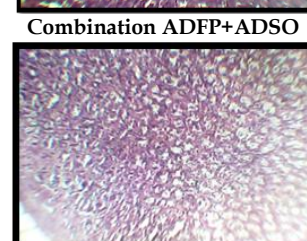
ADFP



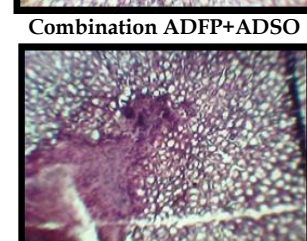
ADSO



ADSO



Combination ADFP+ADSO



Combination ADFP+ADSO

Fig 2: Histology of stomach in pylorus ligation induced ulcer and Ethanol induced gastric-ulcer.

Omeprazole treated groups showed regeneration of mucosal layer and substantial prevention of the formation of hemorrhage and edema. Ulcer due to ethanol is the result of reactive oxygen species generation, micro vascular injury and release of inflammatory mediators, which leads to increased vascular permeability, edema formation resulting in necrotic lesions in the gastric mucosa. Ethanol causes ulceration by creation of reactive oxygen species and gives lesion in mucosa.

During ulcerogenesis the total protein level in the stomach were greater than before, because of stress in

the gastric blood flow which gave to the broadening of hemorrhage and tissue damage due to necrosis. Because of tissue damage, the total protein count was rise in stomach fluid of ulcerated animals. [6] control group rises level of offensive factors such as ulcer index, gastric volume(ml), free and total acidity, protein, pepsin and decreases the level of pH, gastric mucus [6] When ethanolic extract of ADFP (500 mg/kg) was administered to animal result showed significantly decreased the level of offensive factors such as ulcer index (54.44%), gastric volume (20.77%), free acidity (20.86%), total acidity (18.70%), pepsin (33.17%), protein (41.98%) increases level of pH (31.74%) gastric mucous content (54.98%). Similarly, when n-hexane extract of ADSO (300 mg/kg) was administered which were showed significantly decreased the level of ulcer index (53.20%), gastric volume (20.12%), free acidity (13.05%), total acidity (19.17%), pepsin (27.49%), protein (35.18%) & increases level of pH (25.78%) gastric mucous content (60.25%). In combination treated group [ethanolic extract of ADFP (250 mg/kg) + n-hexane extract of ADSO (150 mg/kg)] result was set up near to the standard drug & has got better and more effective result than individual fruit extract of *Adansonia digitata* Linn. Which showed significantly decreased the level of ulcer index (64.37%), gastric volume (32.92%), free acidity (21.32%), total acidity (25.86%), pepsin (33.78%), protein (49.36%) & increases level of pH (32.91%) gastric mucous content (62.63%). The secondary metabolites such as flavonoids, proteins, saponins, tannins, phenols, terpenoids, alkaloids and Fatty acids (palmitic acid, oleic acid, linoleic acid) are act as antioxidant, anti-ulcer. Flavonoids are increasing mucosal PG content, reduction in histamine secretion. Saponins may stimulate mucous membrane protective factors, and tannins render the outermost layer of the mucosa less permeable, for instance to chemical irritation. In addition, terpenoids and alkaloid compounds are also reported to have potent activity against gastric ulcers. [27] In both the models, extracts provided a significant protection against ulcer as indicated by a decrease in the ulcer index, gastric volume, free acidity, total acidity, protein, pepsin and increase in mucous, pH of gastric content. The pathogenesis of pylorus ligation and ethanol-induced ulcer also involves the generation of reactive oxygen species (ROS). *Adansonia digitata* Linn has been previously reported of its strong free radical scavenging properties against superoxide anion, hydroxyl radicals, NO radical, and peroxynitrite anion. In peptic ulcer the level of antioxidant enzymes like SOD, CAT is decreased while LPO is increased. Ascorbic acid acts as an antioxidant molecules The present study reports that standard drug having increase in SOD (32.67 U/mg of protein), CAT (48.47 U/mg of protein) while decrease in LPO(12.70 nmol/mg of protein) 0 Then in test group there was individual ethanolic extract of ADFP showed

significant ($p<0.001$) increase in the activity of SOD (25.17 U/mg of protein), CAT (40.90 U/mg of protein) & decrease in LPO (9.683 nmol/mg of protein) & in n-hexane extract of ADSO showed increase in antioxidant enzymes i.e. SOD (23.17 U/mg of protein) & CAT (35.54 U/mg of protein) while LPO (11.95 nmol/mg of protein) level decreases as compared to control group but the results are lesser than combination of ADFP & ADSO. Antioxidant enzyme level in the combination group was increased i.e., SOD (27.83 U/mg of protein), CAT (43.48 U/mg of protein) while LPO (12.13 nmol/mg of protein). The combination suggests its potent antiulcerogenic and healing effect also reduction in gastric lesion reflects the gastric mucoprotective effect. Hence *Adansonia digitata* Linn showed anti-ulcer activity.

ACKNOWLEDGEMENT

I am thankful to Principal of Appasaheb Birnale College of Pharmacy, Sangli-416416, Maharashtra, India and HOD of Department of Pharmacology for their guidance.

REFERENCES

1. Vimala G, Gricilda Shoba F. A review on antiulcer activity of few Indian medicinal plants. International journal of microbiology 2014; Article ID 519590: 14 pages.
2. Sayed MA. Peptic Ulcer: Mini Review. Peptic Ulcer Disease 2017; July: 2-41.
3. Katzung BG, Masters SB, Trevor AJ. Basic and Clinical pharmacology. Edn. 12, McGraw Hill Education, Mumbai, 2012, pp. 1081-1091.
4. Rang HP, Ritter JM, Flower RJ, Henderson G. Pharmacology. Edn. 5, Elsevier Ltd, Delhi, 2003, pp. 376-379.
5. Chauhan AK, Swamy BM, Kotagiri S, Fariyaz S, Dey T. Indian Medicinal Plants with Antiulcer and Related Beneficial Effects: A Review. Research Journal of Pharmaceutical Biological and Chemical Sciences. 2015;6(3):22-30.
6. Gopinathan S, Rameela N. Anti-ulcer activity of *Aloe vera* juice and *Aloe vera* and *Amla* fruit combined juice in ethanol induced ulcerated rats. Int J Pharm Pharm Sci. 2014;6(6):190-7.
7. Gebauer J, El-Siddig K, Ebert G. Baobab (*Adansonia digitata* L.): a review on a multipurpose tree with promising future in the Sudan. Gartenbauwissenschaft. 2002 Jul 1;67(4):155-60.
8. Kirtikar KR, Basu BD. Indian medicinal plants. Edn. 2, Vol. II, Lalit Mohan Publication, Allahabad, 1996, pp. 354-357.
9. Sundarambal M, Muthusamy P, Radha R. A review on *Adansonia digitata* Linn. Journal of Pharmacognosy and Phytochemistry. 2015 Nov 1;4(4):12.
10. Shahat AA. Procyanidins from *Adansonia digitata*. Pharmaceutical Biology. 2006 Jan 1;44(6):445-50.
11. Tandon R, Khanna RD, Dorababu M, Goel RK. Oxidative stress and antioxidants status in peptic ulcer and gastric carcinoma. Indian Journal of Physiology and Pharmacology. 2004 Jan;48(1):115-8.
12. *Adansonia digitata* Linn. [Internet] 2018. Available from: http://en.m.wikipedia.org/wiki/adansonia_digitata.
13. Zahra'u B, Mohammed AS, Ghazali HM, Karim R. Baobab tree (*Adansonia digitata* L) parts: nutrition, applications in food and uses in ethno-medicine-a review. Ann Nutr Disord & Ther. 2014;1(3):1011.
14. Mota KS, Dias GE, Pinto ME, Luiz-Ferreira A, Souza-Brito AR, Hiruma-Lima CA, Barbosa-Filho JM, Batista LM. Flavonoids with gastroprotective activity. Molecules. 2009 Mar 3;14(3):979-1012.

15. Dalu D. Anti Ulcer Activity of *Leea indica* in Wistar Albino Rats. Archives of Nano medicine: Open Access Journal; 2018 1(1): 1-6.
16. Rajkapoor B, Anandan R, Jayakar B. Anti-ulcer effect of *Nigella sativa* Linn. against gastric ulcers in rats. Current Science. 2002 Jan 25:177-9.
17. Saravanaraj M. Anti-Diabetic Effect of the Ethanolic Extract of Dried Fruits of *Adansonia digitata* Linn. World Journal of Pharmacy and Pharmaceutical Sciences 2017; 6(5): 1597-1605.
18. Dass PM, Danbature WL, Karu E, Ibrahim A, Baba A. Extraction and Biodegradation of Baobab (*Adansonia digitata*) Seeds Oil By Fungi (*Aspergillus niger*). Journal of Natural Sciences Research. 2013;3(5): 128-136.
19. Muhammad IU, Jarumi IK, Alhassan AJ, Wudil AM, Dangambo MA. Acute Toxicity and Hypoglycemic Activity of Aqueous Fruit Pulp Extract of *Adansonia digitata* L. (Afpead) on Alloxan Induced Diabetic Rats. Journal of Advances in Medical and Pharmaceutical Sciences. 2016;6(3):1-6.
20. Ibrahim HM, Ogbadoyi EO, Adamu Ky. Evaluation Of Antitrypanosomal activity of ethyl acetate extract of *Adansonia digitata* Seed Extract In Tb Brucei Infected Albino Mice. International Journal of Drug Research and Technology. 2017 Apr 22;2(7):7.
21. Vogel HG (Ed.) In: Chapter E, Vogel WH, Scholkens BA, Muller J, Voegel WF (eds.) Drug Discovery and Evaluation. Pharmacological Assays Edn.2, Shinger-Verlag, New york, 2002. pp. 867-872.
22. Kulkarni SK. Handbook of Experimental Pharmacology. Vallabh Prakashan, Mumbai, 2011 pp. 148-150.
23. Tamboli FA, More HN. Evaluation of antiulcer and antioxidant activity of *Barleria gibsoni* Dalz. leaves. Pharmacognosy research. 2016 Oct;8(4):226.
24. Adinortey MB, Ansah C, Galyuon I, Nyarko A. In vivo models used for evaluation of potential antigastroduodenal ulcer agents. Ulcer. 2013; Article ID 796405: 12 pages.
25. Balogun ME, Besong EE, Obimma JN, Djobissie SF, Mbamalu OS. Gastroprotective Effect of Ethanolic Extract of *Vigna subterranea* in Ethanol-induced Gastric Mucosal Ulceration in Rats. Indian J Physiol Pharmacol. 2018;62(3):347-58.
26. Kumar NR, Kumar NS, Vangoori Y. Evaluation of antiulcer activity of ethanolic extract of *Momor dicadioica* on pylorus ligation induced and ibuprofen induced ulcer in albino rats. International Journal of Medical Research & Health Sciences. 2013;2(4):762-7.
27. Kaur M, Singh A, Kumar B. Comparative antidiarrheal and antiulcer effect of the aqueous and ethanolic stem bark extracts of *Tinospora cordifolia* in rats. Journal of advanced pharmaceutical technology & research. 2014 Jul;5(3):122.

HOW TO CITE THIS ARTICLE: Malgave KK, Naikwade NS, Ladda PL, Shikalgar TS, Patil SS. Peptic Ulcer Healing Activity of Fruit Pulp Extract and Seed Oil of *Adansonia digitata* Linn. Int. J. Pharm. Sci. Drug Res. 2019; 11(5): 263-271. DOI: 10.25004/IJPSDR.2019.110518