



Evaluating the Anti-Pyretic and Anti-Inflammatory Properties of the Ethanolic Extract of *Mangifera Indica* (Mango) Bark in Albino Wistar Rats

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ABSTRACT

This study was undertaken to examine the anti-pyretic and anti-inflammatory properties of the stem-bark ethanolic extract of mangifera indica (mango) using albino Wistar rats. Male and female rats weighing 150-210g were randomly selected into four groups of four rats each after two weeks of acclimatization. Fever was induced in the rats by the subcutaneous injection of 50% baker's yeast suspended in normal saline after taking their basal body temperature. 18 hours later, feverish rats were treated with oral administration of the extract, with group 1 receiving 0.4ml of water and served as the control. Groups two and three received 100mg/kg and 200mg/kg of the extract respectively while group 4 (reference group) was orally administered with 200mg/kg of aspirin (a known antipyretic agent). Temperature changes in the rats were monitored every 30 minutes for two hours using rectal thermometer. To evaluate the anti-inflammatory effect of the extract, the extract was orally administered to the rats following the dosage as described in antipyretic study. 30 minutes later, 2.5% formalin was subcutaneously injected at the right forelimb of each rat and the number of times the rats licked the injected limb was counted for five minutes and taken as inflammatory response. The result of the studies show that the ethanolic extract of the stem bark of Mangifera indica significantly ($p < 0.05$) reduced baker's yeast induced pyrexia in rats faster than aspirin especially at 100mg/kg within one hour. Also, (100-200 mg/kg) oral administration of the extract significantly ($p < 0.05$) inhibited 2.5% formalin induced inflammation. The different chemical component of the plant especially polyphenolics, flavonoids, triterpenoids and mangiferin may be involved in the observed antipyretic and anti-inflammatory effects. Thus, the result of this study lend credence to the suggested folkloric uses of the plants in the management and control of painful arthritis and other inflammatory condition as well as the associated fever (pyrexia). In conclusion that the ethanolic extract of the bark of mango at low doses could be used as a substitute in the management of pyrexia and inflammation.

KeyWords: Pyrexia, Inflammation, *Mangifera indica*, Flavanoids.

INTRODUCTION

The use of plants and its products in the treatment of diseases is unimaginably as old as mankind. Known ancient cultures have written evidences on

the use of plant for the treatment of diseases which is dated as far back as 5000 years^[1]. Egyptian written evidences were mentioned in the Egyptian medical papyri represented in tomb

illustrations or medical jars containing trace units of herbs.^[1] Those of the Greek were documented in Diocus/Carystus in 3rd century B.C and Krateuas in the 1st century B.C. In China, some seeds used as medical remedies have been found in archaeological sites dating from the Shang Dynasty.^[2] while over 100 of the drugs mentioned in the Huangdi Neijing (an early Chinese medical text) were herbs.^[3] Also, the ancient use of herbs for treatment of diseases has been practiced in India and the defunct Roman Empire.^[4] In the recent times, the use of plants and its parts for the treatment of diseases continue to wax strong, with those suffering from chronic diseases like cancer, diabetes asthma and end-stage renal diseases being the most prevalent users^[5]; even though ethnicity, educational and social classes have influence on the prevalence of the usage.^[6] At the moment, about 80% of the population of Asia and Africa use herbal medicine for some aspects of their primary health care,^[7] while about 25% of the drugs used in the United States have been derived from plants.^[8] Also, at least 7000 compounds in modern drugs today are plants derivatives.^[9]

Several of the plants used as herbal remedy contain phytochemicals like alkaloids, flavanoids, terpenoids, saponins, phenolics and others which give them their pharmacological potentials.^[10]

Among the many plants which are in use today for herbal remedy is *Mangifera indica* (mango)

Mango which belongs to the genus *Mangifera* with about thirty other species is a tropical fruiting tree in the flowering plant family *Anacardiaceae*. It is one of the most popular of all tropical plants with varied medicinal properties attributed to the its different parts . The bark is reported to contain protocatechic acid, catechin, mangiferin, alanine, glycine, γ -aminobutyric acid, kinic acid, shikimic acid and the tetracyclic triterpenoids (cycloart-24-en-3 β ,26diol, 3-ketodammar-24 (*E*)-en-20S,26-diol, C-24 epimers of cycloart-25 en 3 β ,24,27-triol and cycloartan-3 β ,24,27-triol.).^[11] Indicoside A and B, manghopanal, mangoleanone, friedelin, cycloartan-3 β -30-diol and derivatives, mangsterol,

manglupenone, mangocoumarin, n-tetacosane, n-heneicosane, n-triacontane and mangiferolic acid methyl ester including Phenolic Antioxidants, Free Sugars and Polyols have been isolated from different parts of the tree.^[12] Mangostin, 29-hydroxy mangiferonic acid, mangiferin together with common flavonoids have been isolated from the stem bark .^[13] The flower contains alkyl gallates such as gallic acid, ethyl gallate, methyl gallate, n-propyl gallate, n-pentyl gallate, n-octyl gallate, 4-phenyl gallate, 6-phenyl-n-hexyl gallate and dihydrogallic acid.^[14] Root of mango contains the chromones, 3-hydroxy-2-(4'-methylbenzoyl)-chromone and 3-methoxy-2-(4'-methyl benzoyl)-chromone. An essential oil containing humulene, elemene, ocimene, linalool, nerol and many others have also been found in the leaves and flowers. The fruit pulp contains vitamins A and C, β -carotene and xanthophylls.^[15] Also, an unusual fatty acid, *cis-9, cis-15-octadecadienoic acid* has been isolated from the pulp lipids of mango.^[16] The most interesting constituents of *Mangifera indica* however are the polyphenolics, flavonoids, triterpenoids. Mangiferin (a xanthone glycoside and major bio-active constituent), isomangiferin, tannins and gallic acid derivatives. These constituent are found in all parts of the plant in various quantities and have been attributed to be responsible for the various pharmacological effects of the plant.

Over the years, Researchers have conducted a lot of studies on the medicinal uses of the various parts of the plant. In a study, 50% ethanolic extract of the leaf of *mangifera indica* produced a significant hypoglycemic effect at a dose of 250 mg/kg, both in normal and streptozotocin-induced diabetic animals by stimulating β -cells to release insulin.^[17] Aqueous extract of the leaves also showed a similar result on blood glucose level in normoglycaemic, glucose - induced hyperglycaemic and streptozotocin (STZ)-induced diabetic rats.^[18] Mangiferin (a xanthone glycoside) predominant in mango extracts and vimang (An extract of mango branch bark) isolated by Cuban scientist have been shown to

possess a very strong antioxidant properties in vitro ^[19] and have been demonstrated to be effective in the fight against *Herpes simplex* viruses type2 and type1 ^[20,21,22,] The extract also possess Anthelmintic and anti-allergenic activities ^[23,24], Antiparasitic activities ^[25], Anti-microbial ^[26], Hepato-protective ^[27] and gastro-protective activities. ^[28] Studies have also reported that the ethanolic (95%) extract of the seed kernel and leaf of *mangifera indica* exhibited significant anti-inflammatory and anti-nociceptive activity in acute, subacute and chronic cases of inflammation. ^[29, 30, 31] However, reports on the antipyretic and anti-inflammatory activities of this plant's stem bark are scanty and that necessitates the present study.

MATERIALS AND METHODS

PREPARATION OF PLANT MATERIALS AND PLANT EXTRACTION

Fresh barks of mango collected from mango trees around Elele town, Rivers State, Nigeria were properly identified and dried under shed. The dried plant barks were grinded into powdery form using manual grinding machine. 300gms of the powdered material was dissolved in 750 ml of ethanol and placed under mechanical shaker for 48 hours. The mixture was then filtered using a white neat handkerchief; the filtrate was collected and concentrated at 50°C in an electric oven using rotatory evaporator. After drying the filtrate, it was stored in a bottle and preserved in the refrigerator at a temperature of 4°C pending usage.

PROCUREMENT AND PREPARATION OF EXPERIMENTAL ANIMALS

A total of 16 albino wistar rats (males and females) weighing between 150g and 210g were brought and housed in the animal house of the Department of Human Physiology, Madonna University Elele Campus Rivers State, Nigeria. The rats were allowed to acclimatize (14 days) at an ambient temperature of 12hours light and dark cycle and were fed with pellets growers and normal tap water.

3.4 EXPERIMENTAL DESIGN

Following acclimatization, the rats grouped into four groups in accordance with their body weight, each group having four rats. Group 1(210g) served as control group while groups 2 and 3(150g and 170g respectively) served as the test groups. group 4(190g) was used as the reference group and was administered with 200mg/kg of aspirin (a known antipyretic agent)

ANTIPYRETIC STUDY

Using the established method, three rectal temperatures of the rats in each group were measured using rectal thermometer and the average of the three measurements formed the basal temperature for each rat. 30 minutes after getting the basal temperature, fever was induced rats by the subcutaneous injection of 1ml/100g body weight of 50% dried baker's yeast suspended in normal saline. The rats were however allowed 18 hours to develop fever. Only rats with at least 1°C rise in rectal temperature were selected for the study. Following the establishment of fever as indicated by rise in temperature after 18 hrs, Group 1(control) was orally administered with 0.4ml of distilled water, Group 2(150g) and group3 (170g) (test groups) were orally administered with 100mg/kg and 200mg/kg of mango bark extract respectively while Group 4(190g) which served as the reference group received oral administration 200mg/kg of aspirin (a known antipyretic agent). 30 minutes after extract/drug treatment, the rectal temperatures of the rats were measured and recorded and this was repeated every 30 minutes intervals over duration of 2 hours.

ANTI-INFLAMMATORY TEST

Methanolic extracts of mango bark was orally administered to the rats with group one receiving 0.4 ml of water, group 2 received 100mg/kg of extract, group 3 received 200mg/kg of extract and group 4 received 200mg/kg of aspirin. 30 minutes later, 2.5ml of formalin was dissolved in 97.5ml of distilled water to get 2.5% of formalin solution

and 20µl of the solution was then administered subcutaneously at the right forelimb of each rat, the number of times the rats licked the injected right forelimb were counted for 5 minute, counting was repeated for another 5 minutes after 20 minutes interval. This was done for three more consecutive times and the results were recorded.

STATISTICAL ANALYSIS

At the end of the study, data obtained were analyzed using Statistical Package for Social

Sciences (SPSS Version 18.0). One-Way Analysis of Variance (ANOVA) was used to compare the means and Turkey's Multiple Comparison was used to test for statistically significant differences between control and test groups with a statistically significant difference at $P < 0.05$. The results are presented in tables as mean \pm Standard Error of Mean.

* $P < 0.05$, shows significant different, ** $P < 0.05$ shows very significant different and *** $P < 0.05$ shows highly significant different.

RESULTS AND DISCUSIONS

RESULTS: The tables below represent the results obtained from the study

Table.1: showing the antipyretic effect of *Mangifera indica* bark extract on albino wistar rats.

GROUPS	TEMP. BEFORE INDUCING FEVER(°c) (mean±S.E)	TEMP. 18HRS AFTER FEVER INDUCTION (°c)(mean±S.E)	TREATMENT AND DOSAGE	AVERAGE RECTAL TEMPERATURE FOLLOWING TREATMENT(°c)			
				18½ HRS (mean±S.E)	19 HRS (mean±S.E)	19½ HRS (mean±S.E)	20 HRS (mean±S.E)
GROUP1 (CONTROL)	36.95±0.12	38.70±0.21	DISTILLED WATER 0.4ml	38.83±0.20	38.88±0.18	38.63±0.18	38.53±0.21
GROUP 2	37.08±0.12	38.78±0.12	EXTRACT 100mg/kg	36.65±0.13*	36.70±0.25**	36.88±0.09*	36.93±0.05*
GROUP 3	37.05±0.10	38.78±0.25	EXTRACT 200mg/kg	37.98±0.11*	37.23±0.30*	36.88±0.30*	37.03±0.09*
GROUP4 (REFERENCE GROUP)	37.30±0.10	39.08±0.37	ASPIRIN 200mg/kg	38.48±0.09	38.13±0.63	37.63±0.05*	37.03±0.09*

Values are mean \pm S.E.M * $p < 0.05$, ** $p < 0.05$; significantly and very significantly different from control respectively

Table.2: showing the anti-inflammatory effect of *Mangifera indica* bark extract on albino Wistar rats.

GROUPS	TREATMENT	DOSAGE	LIMB LICKING RESPONSE FOLLOWING TREATMENT.			
			0-5 MINS (mean±S.E)	25-30 MINS (mean±S.E)	50-55 MINS (mean±S.E)	75-80 MINS (mean±S.E)
GROUP1 (CONTROL)	DISTILED WATER	0.4ml	105.25±2.29	105.50±0.87	105.00±1.47	88.25±4.03
GROUP 2	EXTRACT	100mg/kg	125.00±2.55	82.50±2.78*	37.75±0.63**	18.75±1.49**
GROUP 3	EXTRACT	200mg/kg	106.00±7.12	74.25±2.39*	35.00±2.35*	17.50±1.00**
GROUP4 (REFERENCE)	ASPIRIN	200mg/kg	95.75±7.05	26.25±2.78*	13.00±2.04**	7.00±0.58***

Values are mean \pm S.E.M * significant at $p < 0.05$, **more significant at $P < 0.05$; different from control

DISCUSIONS

Fever (pyrexia) has been argued to be beneficial in the fight against infections; as it helps the immune system to increase mobility of leucocytes

and its phagocytic activity^[32], decrease endotoxin effects, increase proliferation of T-cells^[33] and enhance ability of interferons activity^[33] in order to destroy pathogens or detoxify their pyrogens.

These pyrogens are known to cause the synthesis and release of cytokines that activate the arachnidonic acid pathway, which in turn leads to the synthesis and release of PGE₂ that act on the temperature regulating areas of the hypothalamus and reset it to a higher point; hence, stimulating the body to generate heat through its output to the sympathetic nervous system, pituitary gland and endocrine organs to meet up with this new high set point^[34]. However, pyrexia could be dangerous as it could lead to abnormal functions of the organs and systems of the body and most times fatal especially in children. In this case, treatment becomes inevitable. The result of the study shows that the ethanolic extract of the bark of mango significantly ($p < 0.05$) decreased the core body temperature of the rats in the test groups especially at 100mg/kg. The result also shows that the ethanolic extract of the bark of mango at 100m/kg acts faster than aspirin at 200mg/kg in reducing the core body temperature in yeast induced pyretic rats. This effect could however be as a result the phytochemical "bioflavanoid" which is known to inhibit the cyclooxygenase reaction, which as well inhibit PGE₂ biosynthesis and hence reduce core body temperature.

The result of the anti-inflammatory study shows that the ethanolic extract of mango bark significantly ($P < 0.05$) decrease inflammation in the test groups compared to the control. For instance, at 200mg/kg of extract, the number of times the rats licked the injected limb in five minutes, reduced from an average of 88 times in the control to an average of 17 times in group3. However, when compared to aspirin at 200mg/kg, aspirin appeared to be more effective in reducing inflammation within the same time (table 2). The observed anti-inflammatory effect could also be linked to the presence of bioflavanoid especially quercertin which has inhibitory effect on the synthesis and release of prostaglandins and other proinflammatory mediators like histamine, serotonin and bradykinins.

CONCLUSION

Fever and inflammation are not actually diseases but the response of the body system to disease conditions. Due to the devastating nature of these responses to the body system, the need for treatment may be considered. This study successfully investigated the use of the ethanolic extract of the bark of mango to treat these responses. The results however, showed that the extract significantly ($p < 0.05$) reduced core body temperature in yeast induced pyretic rats and also reduced inflammation. We therefore conclude that the ethanolic extract of the stem bark of mango at a low dose, could serve as a substitute to the conventional drugs used for these purposes.

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