

ANTI-ANAEMIC AND HYPOLIPIDAEMIC POTENTIALS OF CORN SILK (*STIGMA MAYDIS*) EXTRACT ON PARACETAMOL-OVERDOSE IN WISTAR RATSNwankpa Promise¹ and Uche Mercylyn Ezinne*²¹Department of Medical Biochemistry, Faculty of Basic Medical Sciences, Imo State University, Owerri, Nigeria.²Department of Biochemistry, Abia State University, Uturu, Nigeria.

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ABSTRACT

Aim: The study assessed the anti-anaemic and hypolipidaemic potentials of corn silk (*Stigma maydis*) extract on paracetamol over-dose in wistar rats. **Methodology:** Thirty-two albino Wistar rats weighing between 120g-130g were divided into four groups and used for the study which lasted for 21 days. Group 1 animals which served as the positive control were given normal rat feed and water daily. Group 2 animals received a single dose of 600mg/kg/day of paracetamol in addition to normal rat chow and water. Group 3 received a single dose of 600mg/kg/day of paracetamol + 100mg/kg of ethanol extract of corn silk daily while group 4 received 600mg/kg/day of paracetamol + 200mg/kg of ethanol extract of corn silk daily. All administrations were done orally. After 21 days of treatment, the animals were fasted for 24 hours, euthanized and haematological and lipid parameters were measured. **Results:** Results showed a significant increase ($p < 0.05$) in the levels of total cholesterol (TC), triglyceride (TG), low density lipoprotein (LDL) and very low-density lipoprotein (VLDL) [369.20±2.44; 187.60±1.01; 276.20±1.46 and 36.20±4.29 respectively] in group 2 animals when compared with group 1 animals which served as a positive control [169.60±7.64; 93.40±5.94; 114.80±8.90; 18.60±1.14 respectively]. There was also a significant decrease ($p < 0.05$) in the level of high-density lipoprotein (HDL) in group 2 rats (23.60±5.17) when compared with group 1 rats (36.60±4.22). The hypolipidaemic effect of corn silk was demonstrated when concomitant administration of the various doses of ethanol extract of corn silk [100mg/kg/day (group 3) and 200mg/kg/day (group 4)] with paracetamol led to a significant decrease ($p < 0.05$) in the levels of TC, TG, LDL, VLDL and a significant increase ($p < 0.05$) in the level of HDL when compared with group 2 animals. Also group 2 animals showed a significant decrease ($p < 0.05$) in packed cell volume (33.00±1.73) when compared with group 1 animals (37.00±1.41). However, the PCV of group 3 and 4 animals increased significantly when compared with group 1 and 2 animals. Also, there was a significant increase in total white blood cell count in group 2 animals when compared with group 1. There was, however, no significant change ($p > 0.05$) in total white blood cell count in group 3 and 4 animals when compared with the positive control. **Conclusion:** In conclusion, *Stigma maydis* has hypolipidaemic effect and enhances haematological parameters as well as ameliorates the toxic effects of paracetamol.

KEYWORDS: paracetamol, stigma maydis, anaemia, lipidaemia and wistar rats.

1.0 INTRODUCTION

The use of medicinal plants in the treatment of various diseases is as old as human existence. Interestingly, these herbs which are employed in the treatment of various ailments form the basis of modern day orthodox medical practice.^[1] Presently, there is a plethora of documented scientific studies on the use of various folklore medicinal plants in the treatment of various illnesses such as cancer, cardiovascular diseases, renal failure, hyperlipidaemia, anaemia and diabetes.^[2] Drug resistance, high cost of drugs, litany of side effects and increasing contraindications associated with synthetic drugs are part of the reasons people rely on herbal

medicines to treat various ailments in recent years.^[3] Over 75% of the world population especially those living in Africa and Asia now rely on the use of plants and plant parts such as corn silk either to supplement orthodox drugs or augment it.^[4] The presence of bioactive compounds called phytochemicals explains the use of these medicinal plants in the treatment of various ailments. Some of these phytochemicals are steroids, saponins, phenols, tannins flavonoids and glycosides.^[5]

Stigma maydis (corn silk) are elongated stigma and style of the female growing *Zea mays* which is made up of a tuft of silky threads or hairs.^[6] Most of the corn silk is a

waste product gotten from the harvest and cultivation of maize and is often discarded since it is present in abundance.^[7] Corn silk is usually green in colour at the initial time but upon maturation may become yellow or reddish-brown. It plays an important role in the transfer of pollen grain from the anthers to the stigma. Corn silk measures about 30cm in length, has a sweet taste and rich in nutrients such as vitamins, carbohydrates, proteins, fats and oil. The presence of phytoactive substances such as Sodium, Potassium, Zinc, Magnesium, Calcium, phenols, steroids, saponin and stigmasterol explains its pharmacological potentials.^[8] An estimation of the proximate composition of *Stigma maydis* by^[9] showed that it contains moisture (3.90%), carbohydrate (29.74%), protein (8.95%), dietary fibre (51.24%), lipid (0.66%) and ash (5.51%). Corn silk has been used in China, Turkey, France and United States because of the following health benefits: anti-diabetic, anti-depressant, anti-fatigue, diuretic and hypoglycaemic effects as demonstrated in various reports.^[10] Also,^[11] reported the efficacy of corn silk in the treatment of oxidative stress-induced inflammatory diseases and pleuritic lung disease. Other therapeutic uses of corn silk are in the management of urinary tract infection, bed wetting, prostate hyperplasia, edema, nephrolithiasis and obesity.^[12] Native Americans use corn silk to restore kidney functions that have been previously deranged by the use of nephrotoxic agents.

Paracetamol (acetaminophen) is an over the counter medication used in the management of fever and pain.^[13] It contains a phenol ring and an acyl group. It is universally accepted that paracetamol-induced hepatotoxicity is one of the commonest causes of liver damage^[14,15] and it is attributed to the formation of a highly reactive intermediate (N-acetyl-p-benzoquinone imine) through the action of cytochrome P-450. Also,^[16] reported that at a dosage of greater than 2000mg/day, paracetamol increases the risk of upper gastrointestinal bleeding. Acetaminophen according to^[17] is lethal to snakes and has been suggested as chemical control program for the brown snake (*Boiga irregularis*) in Guam.

To the best of our knowledge, there is dearth of information on the effect of the concomitant administration of paracetamol and corn silk on the haematological parameters and lipid profile. This formed the basis of this study to evaluate the effects of ethanol extract of *Stigma maydis* on paracetamol overdose and on the haematological parameters and lipid profile in Wistar rats.

MATERIALS AND METHODS

Collection and Identification of Samples

Corn silk was harvested from a farmland in Okporo, Orlu LGA, Imo State, Nigeria in April, 2021. The corn silk was authenticated and deposited at the Department of Plant Science and Biotechnology, Imo State University, Owerri.

Extraction of Plant Material

The ethanol extraction of *Stigma maydis* was carried out according to the method described by^[1] with little modification. Corn silk was air-dried and grinded into a fine powder using electrical blender. Exactly 200g of corn silk was soaked in 1000ml of 95% ethanol for 24 hours. The heterogenous mixture was filtered using Whatman number 1 filter paper and evaporated to dryness afterwards at 40°C in a vacuum using a rotary evaporator. Approximate concentrations of the extract were constituted to the required doses for treatment of the animals using normal saline.

Experimental Animals and Preparation of Paracetamol

Paracetamol tablets (Emzor pharmaceuticals limited) were procured from Salvation Pharmacy, Owerri, Imo State. 3000mg of paracetamol was dissolved in 10mls of distilled water to get a stock concentration of 300mg/ml. The dose used was 600mg/kg which is in accordance with those reported by.^[18]

A total of thirty-two (32) experimental albino Wistar rats weighing between 120-130g were used for this study. The animals were purchased from the animal house of Department of Biochemistry, University of Nigeria Nsukka and kept in the animal house of the Department of Biochemistry, Imo State University, Owerri. They were acclimatized for 14 days under standard environment at a temperature of 22-25°C, 12 hours light and 12 hours dark cycle before the commencement of the experiment. The animals were fed with rat chow and water *ad libitum*. *S. maydis* extract and paracetamol were orally administered to the rats using a gastric tube inserted through their mouth. The extracts and paracetamol were administered for 21 days.

Experimental Design

Thirty-two (32) albino Wistar rats were randomly divided into four (4) groups of eight (8) animals each and were given the following treatments:

Group 1: (positive control) were fed with normal rat chow and water *ad libitum*.

Group 2: (negative control) received a single dose of 2ml (600mg/kg/day) of paracetamol.

Group 3: Received a single dose of 2ml (600mg/kg/day) of paracetamol + 100mg/kg/day of *S. maydis* extract.

Group 4: Received a single dose of 2ml (600mg/kg/day) of paracetamol + 200mg/kg/day of *S. maydis* extract.

Collection of Samples and Analytical Procedures.

The animals were fasted for 24 hours after the administration of the last dose of the extract and then sacrificed under chloroform anesthesia. Blood was collected through cardiac puncture and one portion of it was immediately transferred into EDTA (ethylenediamine-tetraacetic acid) container for determination of haematological parameters which include packed cell volume, haemoglobin level, white blood cell count and differentials (neutrophils,

lymphocytes, eosinophils and monocytes), red blood cells mean corpuscular volume, mean corpuscular haemoglobin and mean corpuscular haemoglobin concentration. The method described by^[19] was used in the measurement of haematological parameters. Another sample was added to plain tubes and serum obtained by centrifuging at 1000rpm for 5 minutes using a Wisperfuge centrifuge (1984). The serum was collected with a Pasteur pipette, transferred to a clean tube and used to determine the levels of total cholesterol (TC), triglyceride (TG) and high-density lipoprotein (HDL) by enzymatic assay method using analytical kits from Randox Laboratories Ltd UK. Very low-density lipoprotein (VLDL) was determined using the formula

$$VLDL = TG/5.$$

Low density lipoprotein was calculated using Friedewald formula:

$$LDL = TC - HDL - VLDL.$$

Statistical Analysis

Data generated from this study was entered in Excel sheets and the means of the values and standard deviation calculated (mean±SD) using Microsoft Excel. The values were subjected to students' T-test using Graphpad Prism statistical software at probability level of 0.05.

RESULTS

The results of the effects of administration of ethanol extract of *Stigma maydis* on paracetamol-overdose and lipid profile and haematological parameters are presented in tables.

Table 1: The effects of administration of ethanol corn silk extract on paracetamol-overdose and lipid profile in albino Wistar rats.

Parameters	Group 1	Group 2	Group 3	Group 4
Total Cholesterol	169.60±7.64	369.20±2.44 ^a	235.25±4.80 ^{ab}	159.40±3.29 ^{bc}
Triglyceride	93.40±5.94	187.60±1.01 ^a	156.20±2.13 ^{ab}	104.10±2.80 ^{bc}
HDL	36.60±4.22	53.60±5.17 ^a	61.20±5.89 ^{ab}	49.80±2.86 ^{ac}
LDL	114.80±8.90	276.20±1.46 ^a	140.20±1.71 ^{ab}	99.20±2.39 ^{ac}
VLDL	18.60±1.14	36.20±4.27 ^a	31.40±4.27 ^a	10.40±1.14 ^{abc}

Legend: HDL- High Density Lipoprotein, LDL- Low Density Lipoprotein, VLDL: Very Low-Density Lipoprotein

a= significantly different ($p < 0.05$) from group 1.

b= significantly different ($p < 0.05$) from group 2.

c= significantly different ($p < 0.05$) from group 3.

Table 1 shows the effect of administration of ethanol extract of *Stigma maydis* on paracetamol-overdose and lipid profile of Wistar rats. There was a statistically significant change ($p < 0.05$) in the levels of TC, TG,

HDL, LDL, and VLDL in group 2 animals which served as a negative control when compared with group 1 animals (positive control). While the level of HDL decreased significantly, there was a statistically significant increase in the levels of TC, TG, LDL and VLDL. Also, animals in groups 3 and 4 showed a dose-dependent significant decrease ($p < 0.05$) in the levels of TC, TG, LDL and VLDL when compared with group 2 animals. There was, however, a statistically significant increase ($p < 0.05$) in HDL in groups 3 and 4 Wistar rats when compared with the negative control group.

Table 2: Effects of administration of ethanol extract of *Stigma maydis* on paracetamol-overdose and haematological parameters of albino Wistar rats.

Parameters	Group 1	Group 2	Group 3	Group 4
PCV	37.00±1.41	33.00±1.73 ^{ab}	39.80±1.10 ^b	41.60±1.67 ^{ac}
HB(g/dl)	11.56±0.79	6.82±1.20 ^{ab}	13.22±0.37 ^{bc}	13.76±0.55 ^{ab}
HB (%)	76.80±5.54	58.40±8.17 ^{ab}	87.60±2.19 ^{abc}	91.20±3.83
T. WBC	4200.00±122.47	6500.00±500.00 ^a	4900.00±1341.64 ^b	4420.00±576.19 ^{ab}
RBC	8.08±0.43	5.56±0.52 ^a	4.50±2.12 ^a	6.90±0.29
MCV	43.16±3.42	64.44±8.46 ^a	72.98±8.30 ^{ab}	60.37±12.72 ^{abc}
MCH	14.34±1.15	21.40±2.85 ^a	24.24±2.77 ^{ab}	19.98±1.49 ^{abc}
MCHC	33.20±0.10	33.20±0.10	33.26±0.05	33.10±0.39
N	48.20±1.30	48.80±0.84	48.40±0.89	48.40±0.55
L	49.20±0.84	49.00±0.71	48.40±0.89	49.00±0.00
E	1.80±0.84	1.00±0.00	1.40±0.55	1.80±0.45
M	1.00±0.00	1.00±0.00	1.00±0.00	1.00±0.00

a= significantly different ($p < 0.05$) from group 1.

b= significantly different ($p < 0.05$) from group 2.

c= significantly different ($p < 0.05$) from group 3.

Values presented are mean \pm SD of three determinations. n=6.

Values bearing different superscripts (a-d) are significantly different ($p < 0.05$).

Legend: PCV- Packed Cell Volume, HB- Haemoglobin, T. WBC- Total White Blood Cells, RBC- Red Blood Cells, MCV- Mean Cell Volume, MCH- Mean Cell Volume, MCHC- Mean Corpuscular Haemoglobin Concentration, N- Neutrophils, L- Lymphocytes, E- Eosinophils, M- Monocytes.

Table 2 shows the effects of administration of ethanol extract of corn silk on paracetamol-overdose and haematological parameters in albino Wistar rats. The results showed a statistically significant decrease ($p < 0.05$) in the levels of PCV, RBC, in group 2 animals when compared with the group 1 animals. MCHC, neutrophils, lymphocytes, eosinophils and monocytes showed no significant change ($p > 0.05$) in their levels when group 2 Wistar rats are compared with group 1 rats though WBC level increased significantly ($p < 0.05$). Administration of paracetamol with 100mg/kg/day and 200mg/mg/kg/day in group 3 and 4 animals respectively led to a significant change ($p < 0.05$) in the levels of PCV, Hb, WBC, RBC in group 3 and 4 animals when compared with group 2 animals.

DISCUSSION

Lipid profile is a panel of diagnostic blood serum tests.^[20] which is employed as a screening tool for abnormalities in lipids such as total cholesterol, triacylglycerol, low density lipoprotein, high density lipoprotein and very low-density lipoprotein.^[19,21] The result of our lipid profile assessment is an essential tool in the diagnosis of cardiovascular diseases.^[22] and to some extent certain forms of cancer.^[23] and diabetes mellitus/ pancreatitis.^[23] due to hyperlipidaemia. Hyperlipidaemia is characterized by elevated levels of cholesterol, triglycerides, phospholipids and changes in lipoprotein.^[24] and one of the ways of managing the increasing cases of hyperlipidaemia and its attendant complications such as hypertension and atherosclerosis is through the consumption of plants and plant products.^[23] These plants contain phytochemicals such as saponins, flavonoids, and steroids which possess some lipid lowering potentials.^[25]

The results of this study clearly indicate that ethanol extract of corn silk produced hypolipidaemic effect on experimental animals after paracetamol induced hyperlipidaemia with elevated levels of cholesterol, triglyceride, low density lipoprotein, very low-density lipoprotein and the decreased level of the "good cholesterol": high density lipoprotein. Previous studies

have shown that *Stigma maydis* is rich in bioactive compounds such as flavonoids (e.g. maysin and glycosylflavones), glycosides, saponins, steroids and phenols which have contributed to its pharmacological potentials.^[26] Inhibition of lipid absorption by saponins may be the underlying mechanism through which corn silk exerts its hypolipidaemic effect.^[27] According to the findings of.^[28] inhibition of cholesterol esterase, activation of fatty acid synthase, acetyl CoA carboxylase and production of triglyceride precursors such as acetyl CoA and glycerol phosphate may explain the lipid lowering effect of corn silk. Formation of complexes with compounds such as glycosides and saponins inhibits the absorption of cholesterol from the intestine and may be one of the means through which corn silk extract acts.^[29] Reports by.^[30,31] showed the ability of flavonoids to inhibit HMG-CoA and hence decrease levels of triacylglycerol and cholesterol in experimental animals. Corn silk is rich in flavonoids.

The above findings show that oral administration of corn silk extract enhances metabolism of lipids and hence can be antihyperlipidaemic. It is obvious from this study that treatment of Wistar rats with corn silk extract reduced atherogenic index (TG/HDL) in a dose dependent manner. Also.^[32] reported such a reduction in atherogenic indices in plants containing similar phytoconstituents as corn silk in alloxan induced diabetic rats. Increase in HDL (which is responsible for the transport of cholesterol from the peripheral tissues to the liver) is an enhancement of atherogenicity. HDL exerts its atherogenic effect by inhibiting the peroxidation of LDL by transition metal ions as well as countering LDL oxidation.^[33]

Assessment of blood indices in experimental animals remains a useful tool in ascertaining the effect of xenobiotics on such an organism. Measurement of parameters such as packed cell volume, white blood cell count and related indices can be employed in the diagnosis of diseases such as cancer, cardiovascular diseases, bleeding disorders and infective processes as well as unravelling their pathogenesis.^[34] This is important considering the fact that the blood composition changes directly or indirectly in relation to the pathophysiologic state of such organisms. For optimal functioning of an organism's body, blood and its constituents must remain in their natural state.^[35] Treatment of rats with 600mg/kg/day of paracetamol in our study led to a significant decrease ($p < 0.05$) in PCV levels while the white blood cell counts significantly increased ($p < 0.05$). The decreased PCV could mean that there was either a decrease/inhibition of erythropoiesis/erythropoietin production or release from the kidneys. This is similar to the findings of.^[36, 37] who treated rats with ethanol extract of *D. tripetala* and *Treulia africana*. Leukocytosis seen in this study is a reflection of the toxicity associated with paracetamol intake. Administration of ethanol extract of corn silk to our experimental animals brought about stabilization of

haematological parameters disrupted by the administration of paracetamol. The significant increase in PCV, Hb, MCV and MCH seen in group 3 and 4 animals when compared with the control gives an indication that the extract may contain some bioactive compounds that may trigger and enhance erythropoiesis in the stem cells of such animals. Increased erythropoiesis increases haemoglobin level^[38] and this may be attributable to the presence of antioxidants such as vitamin C, Zinc and flavonoids which reduce lipid peroxidation and hence haemolysis of red blood cells.^[38] These antioxidants present in corn silk also possess the ability to inhibit production of polyunsaturated fatty acids in the cells.^[39] The reduction in total WBC level seen in animals treated with various doses of the extract when compared with animals that received only paracetamol lends credence to the use of corn silk in the treatment of various forms of infections.

CONCLUSION

This study investigated the ameliorative effects of corn silk ethanol extract on paracetamol induced toxicity on Wistar rats. The results demonstrated that the extract has a strong hypolipidaemic and haematopoietic effect and can hence reduce the risk of cardiovascular disease and protect the body against infections.

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