

Cell Membranes and Free Radical Research

Volume 5, Number 1, 2013

ISSN Numbers: 1308-4178 (On-line), 1308-416X

Indexing: Google Scholar, Index Copernicus, Chemical Abstracts, Scopus (Elsevier),
EBSCOhost Research Database

EDITOR

Editor in Chief

Mustafa Nazıroğlu, Department of Biophysics,
Medical Faculty, Suleyman Demirel University, Isparta,
Turkey.
Phone: +90 246 211 37 08. Fax: +90 246 237 11 65
E-mail: mustafanaziroglu@sdu.edu.tr

Managing Editor

A. Cihangir Uğuz, Department of Biophysics, Medical
Faculty, Suleyman Demirel University, Isparta, Turkey.
E-mail: biophysics@sdu.edu.tr

EDITORIAL BOARD

Cell Membranes, Ion Channels and Calcium Signaling

Alexei Tepikin, The Physiological Laboratory, University of
Liverpool, Liverpool, UK

Andreas Lückhoff, Institute of Physiology, Medical Faculty,
RWTH-Aachen University, Germany

Andreas Daiber, 2nd Medical Clinic, Molecular Cardiology,
Medical Center of the Johannes Gutenberg University,
Mainz, Germany

Giorgio Aicardi, Department of Human and General
Physiology, University of Bologna, Italy.

Gemma A. Figtree, North Shore Heart Research Group
Kolling Institute of Medical Research University of Sydney
and Royal North Shore Hospital
Sydney, AUSTRALIA.

Jose Antonio Pariente, Department of Physiology,
University of Extremadura, Badajoz, Spain.

James W. Putney, Jr. Laboratory of Signal Transduction,
NIEHS, NC, USA.

Martyn Mahaut Smith, Department of Cell Physiology and
Pharmacology, University of Leicester, Leicester, UK.

Stephan M. Huber, Department of Radiation Oncology,
Eberhard - Karls University Tübingen, Germany

Enzymatic Antioxidants

Michael Davies, Deputy Director, The Heart Research
Institute, Sydney, Australia.

Süleyman Kaplan, Department of Histology and
Embryology, Medical Faculty, Samsun, Turkey

Xingen G. Lei, Molecular Nutrition, Department of Animal
Science, Cornell University, Ithaca, NY, USA

Ozcan Erel, Department of Biochemistry, Medical Faculty,
Yıldırım Beyazıt University.

Nonenzymatic Antioxidants, Nutrition and Melatonin

Ana B. Rodriguez Moratinos, Department of Physiology,
University of Extremadura, Badajoz, Spain.

Cem Ekmekcioglu, Department of Physiology, Faculty of
Medical University of Vienna, Austria.

Peter J. Butterworth, Nutritional Sciences Division, King's
College London, London, UK

AIM AND SCOPES

Cell Membranes and Free Radical Research is a print and
online journal that publishes original research articles,
reviews and short reviews on the molecular basis of
biophysical, physiological and pharmacological processes
that regulate cellular function, and the control or
alteration of these processes by the action of receptors,
neurotransmitters, second messengers, cation, anions,
drugs or disease.

Areas of particular interest are four topics. They are;

A- Ion Channels (Na⁺ - K⁺ Channels, Cl⁻ channels, Ca²⁺
channels, ADP-Ribose and metabolism of NAD⁺, Patch-
Clamp applications)

B- Oxidative Stress (Antioxidant vitamins, antioxidant
enzymes, metabolism of nitric oxide, oxidative stress,
biophysics, biochemistry and physiology of free oxygen
radicals)

C- Interaction Between Oxidative Stress and Ion Channels

(Effects of the oxidative stress on the activation of the
voltage sensitive cation channels, effect of ADP-Ribose
and NAD⁺ on activation of the cation channels which
are sensitive to voltage, effect of the oxidative stress on
activation of the TRP channels)

D- Gene and Oxidative Stress (Gene abnormalities.
Interaction between gene and free radicals. Gene
anomalies and iron. Role of radiation and cancer on
gene polymorphism)

READERSHIP

Biophysics
Biochemistry
Biology
Biomedical Engineering
Pharmacology
Physiology
Genetics
Cardiology
Neurology
Oncology
Psychiatry
Neuroscience

Keywords

Ion channels, cell biochemistry, biophysics, calcium
signaling, cellular function, cellular physiology,
metabolism, apoptosis, lipid peroxidation, nitric oxide
synthase, ageing, antioxidants, neuropathy.

Musa Sapientum attenuates total antioxidant and lipid profile values in rats with indomethacin-induced gastric ulceration

Abdulazeez Adelaja Akinlolu ¹, Olaide Kamal Ghazali¹, Bamidele Adewale Salau², Jubril Olatoye Otulana³, Mobolaji Omowunmi¹

¹ Department of Anatomy, University of Ilorin, Kwara State, Nigeria.

² Department of Biochemistry, Redeemer's University, Ogun State, Nigeria.

³ Department of Anatomy, Olabisi Onabanjo University, Ogun State, Nigeria.

List of abbreviations

(NaH₂PO₄) Sodium dihydrogen phosphate

(Na₂HPO₄) Disodium hydrogen phosphate

(H₂O₂) Hydrogen peroxide,

(TCA) Trichloroacetic acid

(H₂SO₄) Sulphuric (VI) acid

(HCl) Hydrochloric acid

(TBA) 2-thiobarbituric acid

Corresponding Address

AKINLOLU Abdulazeez Adelaja Department of Anatomy, Faculty of Basic medical Sciences, University of Ilorin, Ilorin, Kwara State, Nigeria.

Email: a3akin@gmail.com

Phone: 234 - 8062765308

Acknowledgments

This research has been supported by Gobierno de Extremadura, (Fondos FEDER), PII-Acción VII-UJEX and Centro de Información Cerveza y Salud (CICS).

Abstract

We investigated anti-ulcer properties of *Musa sapientum* sucker in rats. Group I was control. Indomethacin (40mg/kg/bodyweight) was orally administered to rats of Groups II - VI for induction of ulcer. Four hours after administrations of the ulcerogen; 5, 10 and 20mg/kg/bodyweight of *Musa sapientum* sucker extract and 40mg/kg/bodyweight of Omeprazole were administered orally to rats of Groups III - VI respectively. Analyses of lipid profile status, kidney functions tests and activities of glutathione, superoxide dismutase and catalase in the serum and stomach samples of rats of Group II showed statistically significant higher activities ($P \leq 0.05$) of antioxidant enzymes, lipid peroxidation, total cholesterol, triglycerides, creatinine, bilirubin and urea when compared to rats of Groups III - VI. Non-statistically significant lower activities ($P \leq 0.05$) of antioxidant enzymes and lipid profile status were observed in rats of Groups III and IV when compared with rats of Group VI. Evaluations of creatinine, urea and bilirubin levels in sera samples of rats of Groups III - V showed non-statistically significant lower levels ($P \leq 0.05$) of kidney functions tests when compared to rats of Group VI. This study concluded that *Musa sapientum* attenuated total antioxidant and lipid profile values; served reno-protective functions in rats with Indomethacin-induced gastric ulceration.

Keywords

Musa sapientum, Indomethacin, Ulceration.

Introduction

Ulceration refers to any break in the skin or mucus membrane and is classified according to the part of the digestive system in which it occurs (Ramzi *et al.*, 1999; Maity and Chattopadhyay, 2008). Gastric ulcer occurs in the stomach and is characterized by the malfunctioning or non-availability of gastric juice for the breaking down of proteins into smaller polypeptides (Ramzi *et al.*, 1999; Maity and Chattopadhyay, 2008). Peptic ulcer occurs in sections of the gastro intestinal tract exposed to gastric acid and pepsin such as the stomach and duodenum (Ramzi *et al.*, 1999). The etiology is not clearly known. It results probably from an imbalance between aggressive (acid, pepsin and *Helicobacter pylori* infection) and defensive (gastric mucus and bicarbonate secretion, prostaglandins, cyclooxygenases, nitric oxide, innate resistance of the mucosal cells) factors; as well as factors such as genetic, psychosomatic, humoral and vascular derangements (Ramzi *et al.*, 1999).

Reactive oxygen species generated by ulcerogens disrupt the balance between the aggressive and defensive factors, pro- and anti-inflammatory factors, pro- and anti-angiogenic factors resulting in reduction of the viscosity of gastric mucosa thereby affecting the quality of its protection (Maity and Chattopadhyay, 2008). Phospholipids of the gastric mucus gel layer act as primary barriers to acid-induced damage of the stomach by establishing the hydrophobicity of the stomach's epithelial cellular layer (John, 2000). Indomethacin belongs to the group of non-steroidal anti-inflammatory drugs (NSAIDs) which have been established to interact with surface-active phospholipids of the gastric mucosa, thereby reducing its hydrophobicity and the quality of its protection against ulcerogens. (John, 2000).

Musa sapientum belongs to the family musaceae and is a food crop well grown in villages and towns in Nigeria. (Salau *et al.*, 2010). Its various parts have been described to possess different medicinal properties. Its banana pulps (Lewis *et al.*, 1999) and unripe plantain bananas (Prabha *et al.*, 2011) have been reported to have anti-ulcerogenic properties while Hossain *et al.*, 2011 observed that its seeds possess antioxidant, anti-diarrheal and anti-microbial activities. Significant antioxidant properties have similarly been observed in investigations of peel extracts (Ramakrishnan *et al.*, 2011), inflorescence and stalk (Jayamurthy *et al.*, 2013) of *Musa sapientum*.

Although we had earlier investigated the hypoglycaemic properties of *Musa sapientum*'s sucker (Salau *et al.*, 2010), we are not aware of any study that evaluated the anti-ulcer activities of the suckers of *Musa*

sapientum. This study, therefore, tested the hypothesis that *Musa sapientum* improves total antioxidant and lipid profile status of adult wistar rats in indomethacin-induced gastric mucosa ulceration.

Materials and Methods

Collection and Authentication of *Musa sapientum*

10 kg of fresh sucker of *Musa Sapientum* was collected from a farm located on Olabisi Onabanjo University, Remo Campus, Ikenne, Ogun State, Nigeria. The plant was identified and authenticated at the Forest Research Institute of Nigeria, Ibadan, Oyo State, Nigeria. The authenticated plant has the Voucher number FHI 108349 and samples were deposited at the herbarium.

Ethical Approval, Care and Feeding of Animals

Thirty adult female wistar rats weighing between 150 - 220g were obtained from the colony bred of the Physiology Department of University of Ibadan, Ibadan, Oyo State, Nigeria. Animals were fed during the experiment with growers feed from Bendel Feed and Flour Mill Limited, Nigeria. The animals were caged under standard condition in the well ventilated animal house of the Faculty of Basic Medical Sciences of University of Ilorin, Ilorin, Nigeria at room temperature of 25 °c. Water was supplied ad libitum to the rats. Ethical approval was sought and received from the ethical committee of the Faculty of Basic Medical Sciences of University of Ilorin, Ilorin, Nigeria on the usage of animals for experimental studies.

Chemicals, Reagents and Laboratory Equipments

Indomethacin (Hovid, Nigeria), Omeprazole (Eprazole, China), Sodium dihydrogen phosphate (NaH_2PO_4), Disodium hydrogen phosphate (Na_2HPO_4), Hydrogen peroxide (H_2O_2) and Trichloroacetic acid (TCA) were products of Aldrich Chemicals; Sulphuric (VI) acid (H_2SO_4) and Hydrochloric acid (HCl) were products of BDH Chemical Limited, Poole, England; Tris buffers, 2-thiobarbituric acid (TBA), Phosphoric acid and Pyrogallol were products of Sigma Chemicals, St. Louis USA and assay kits for Total Cholesterol, High Density Lipoprotein - Cholesterol, Triglycerides, Creatinine, Bilirubin and Urea (Randox Laboratories, United Kingdom). Spectrophotometer (Jenway Model 6405, UV/visible), centrifuge, pH meter (Rex model pHs 25), Norm-jet needles and syringes (Norm-jet Inc. Tuttlinger, Germany) and anticoagulant tubes (Sterling products, England).

Preparation of Plant Extracts

Harvested *Musa sapientum* suckers were rinsed in order to remove any contaminant such as sand. The sucker was chopped into smaller pieces to increase the surface area for easy and fast drying. The pieces were shade dried at room temperature 25°C – 30°C for two weeks in order to prevent direct sunlight which can react with the active ingredients of the plant. This prevented deterioration of the phytochemical constituents of the plant material. Dried pieces of the plant material were pulverized and 200g of the dried sucker extracted with 70% methanol for 48hrs. The extract was filtered, concentrated with rotary evaporator and further dried on a water bath. The yield of the extract was 2.61% (Salau *et al.*, 2010).

Phytochemical Evaluations of *Musa sapientum* Sucker

The *Musa sapientum* extract was evaluated for the presence of different chemical groups using standard methods as earlier described. (Akinlolu *et al.*, 2006; Akinlolu *et al.*, 2008 and Salau *et al.*, 2010).

Evaluations of the Antioxidant Activities of *Musa sapientum* in Indomethacin – induced Gastric Ulceration

Feeding of the animals was terminated 24 hours before the commencement of experimental procedures. The animals were, however, allowed free access to water and were randomly divided into six treatment groups of five rats each. Physiological saline was administered orally to rats of Control Group I. (Day 1). Rats of Experimental Groups II – VI equally received oral administrations of 40 mg/kg bodyweight of Indomethacin with the aid of a 3ml syringe on Day 1 of experimental procedure. Consequently, rats of Group II were sacrificed by cervical dislocation after four hours of drug administration. (Day 1). Their stomach organs were morphologically examined and were all observed to have had gastric ulcerations. The stomach and kidney tissues of the sacrificed rats of Group II were excised and removed for analyses of lipid profile, total antioxidant status and kidney functions tests.

After four hours of the administration of Indomethacin solution, all rats of Experimental Groups III – VI were treated with oral administrations of 5, 10 and 20mg/kg/bodyweight of the methanolic extract of *Musa sapientum*; and 40 mg/kg bodyweight of Omeprazole (the standard control or positive group) respectively on Day 1 and for another six days (Days 2 - 7) of the experimental procedure. Each drug or extract dose was freshly prepared daily before oral administration. At the

end of experimental procedures (on Day 7), all rats of Control Group I and Experimental Groups III – VI were sacrificed by cervical dislocation. The stomach and kidney tissues of the sacrificed rats were excised and removed for analyses of lipid profile, total antioxidant status and kidney functions tests. All assays were carried out within 24 hours of sample collection.

Preparations of the Stomach, Kidney and Sera Samples of Rats for Determinations of Kidney Functions Tests, Total Antioxidant and Lipid Profile Status

Each organ (stomach or kidney) was cut into small pieces, placed in a mortar and 0.1M phosphate buffer (extracting solution) of at least four times the volume of the organ was added. The organ was homogenized into fine solution with the use of mortar and pestle. The homogenate was poured into a test tube and centrifuged at 2000 revolutions per minute for 10minutes. The supernatant was carefully removed and the residue was discarded. The supernatant served as the sample for the estimation of superoxide dismutase, glutathione and catalase values. Similarly blood samples were centrifuged to separate the serum from the red blood cells and the serum was stored away for biochemical analyses of antioxidant and lipid profile status, creatinine, urea and bilirubin assays of kidney function tests.

Evaluations of Superoxide dismutase, Glutathione and Catalase Levels

Standard methods were used to determine the activities of superoxide dismutase (Pelin *et al.*, 2002) and Glutathione (Sedlak and Lindsay, 1968 and Akande *et al.*, 2011) in stomach samples of rats of Groups I – VI; and Catalase (Sinha, 1972 and Olayemi *et al.*, 2012) in sera and stomach samples of rats of Groups I - VI.

Evaluations of Lipid Peroxidation

The thiobarbituric acid assay (TBARS assay) method was used to quantify Malondialdehyde concentrations in sera and stomach samples of rats of Groups I - VI as earlier described by Akinlolu *et al.*, (2012).

Evaluations of Lipid Profile status

The Total Cholesterol, High Density Lipoprotein – Cholesterol and Triglycerides levels were determined in sera samples of rats of Groups I - VI based on the protocols described in assay kits of Randox Laboratories, United Kingdom. Low Density Lipoprotein-Cholesterol was calculated from measured mean values of Total

Cholesterol, HDL-cholesterol and Triglycerides according to the relationship below:

$$[\text{LDL-cholesterol}] = [\text{total cholesterol}] - [\text{HDL-cholesterol}] - [\text{Triglycerides}]/5$$

Evaluations of Kidney Functions Tests

Creatinine, bilirubin and urea concentrations were determined in sera samples of rats of Groups I - VI based on the protocols described in assay kits of Randox Laboratories, United Kingdom.

Statistical Analyses

The Mean ± S.E.M (S.E.M. = Standard Error of Mean) value of each of the measured parameters of kidney functions tests, antioxidants and lipid profile assays in rats of Control Group II (which received Indomethacin only) were compared with rats of Groups I and III - VI (which received physiological saline, Indomethacin plus extract doses of *Musa sapientum* sucker or Indomethacin plus Omeprazole) for any significant difference using the Student's t-test for unpaired samples. P values of 0.05 (or less) were taken as statistically significant.

Results

Phytochemical Analyses:

Phytochemical screenings of *Musa sapientum* sucker showed the presence of Saponins, Saponin glycosides, Tannins, Alkaloids and Indole Alkaloids. (Salau *et al.*, 2010).

Evaluations of Glutathione Activities:

Analyses of the activities of glutathione in the stomach samples of rats of Groups III, IV and VI showed

statistically non-significant higher levels ($P \leq 0.05$) of glutathione when compared with glutathione activities in rats of Group II. (Table 1). However, statistically significant higher levels ($P \leq 0.05$) of glutathione were observed in rats of Group V when compared to glutathione activities in rats of Group II. (Table 1).

Evaluations of Superoxide dismutase

Activities:

Analyses of the superoxide dismutase activities in the stomach samples of rats of Groups III - VI showed statistically significant lower levels ($P \leq 0.05$) of superoxide dismutase when compared with superoxide dismutase activities in rats of Group II. (Table 1).

Evaluations of Catalase Activities:

Analyses of catalase activities in the sera and stomach samples of rats of Groups III - VI showed statistically significant lower levels ($P \leq 0.05$) of catalase when compared with catalase activities in rats of Group II. (Table 1).

Evaluations of Malondialdehyde

Concentrations:

Analyses of lipid peroxidation in sera samples showed comparatively similar concentrations of Malondialdehyde ($P \leq 0.05$) in rats of Groups II - VI. (Table 2). However, statistically significant lower concentrations of Malondialdehyde ($P \leq 0.05$) were observed in stomach samples of rats of Groups III - VI when compared with rats of Group II. (Table 2).

Table 1. Results of the statistical analyses of the activities of Glutathione and Superoxide dismutase (in the stomach); and Catalase in the Sera or Stomach Samples of Rats of Groups I - VI. ($P \leq 0.05$).

Groups of rats	Doses of drugs/extract	Glutathione levels mg/100g (Mean ± S.E.M.)	SOD levels U/L (Mean±S.E.M.)	Catalase activity (U/L) (Mean ± S.E.M.)	
				Stomach	Serum
I	Physiological saline	6.30±0.58	149.75±13.05	3.48±0.18	4.35±0.12
II	40mg/kg bw Indomethacin	5.58±0.14	320.06±68.84	6.23±0.12	5.10±0.82
III	5mg/kg bw <i>Musa sapientum</i>	7.68±0.29	98.44±0.94	3.71±0.47	2.85±0.38
IV	10mg/kg bw <i>Musa sapientum</i>	7.78±0.30	17.93±1.04	2.02±0.24	1.97±0.50
V	20mg/kg bw <i>Musa sapientum</i>	13.85±0.37	10.50±0.81	1.45±0.92	1.06±0.56
VI	40mg/kg bw Omeprazole	7.65±0.27	146.25±26.36	2.42±0.34	2.18±0.75

bw = bodyweight, S.E.M. = Standard Error of Mean, SOD = Superoxide dismutase

Lipid Profile Analyses:

Statistically significant lower levels ($P \leq 0.05$) of total cholesterol, low density lipoprotein-cholesterol and triglycerides but higher levels ($P \leq 0.05$) of high density lipoprotein-cholesterol were observed in the sera and stomach samples of rats of Group III - VI when compared with lipid profile levels in rats of Group II. (Table 3).

Evaluations of Kidney Functions Tests

Evaluations of Creatinine, Urea and Bilirubin Concentrations in the sera samples of rats of Groups I - VI showed statistically significant dose-dependent lower concentrations ($P \leq 0.05$) of Creatinine, Urea and Bilirubin in rats of Groups III - V when compared with observed

results of kidney function tests in rats of Group II. (Table 4). Statistically non-significant dose-dependent lower concentrations ($P \leq 0.05$) of Creatinine, Urea and Bilirubin were observed in rats of Groups III - V when compared with observed results of kidney functions tests in rats of Group VI. (Table 4). The extract doses of *Musa sapientum* sucker better attenuated values of kidney function tests when compared with the standard drug (Omeprazole). (Table 4).

Discussions

Indomethacin is an established ulcerogen, especially in an empty stomach (Akinlolu *et al.*, 2006 and Akinlolu *et al.*, 2008). Indomethacin induced ulceration occurs mostly

Table 2. Lipid peroxidation (Malondialdehyde) levels in the Sera and Stomach of Rats of Groups I - VI. ($P \leq 0.05$).

Groups of rats	Doses of drugs/extract	Malondialdehyde levels	
		Serum ($\mu\text{mol/ml}$) \pm S.E.M.	Stomach ($\mu\text{mol/ml}$) \pm S.E.M.
I	Physiological saline	2.97 \pm 0.87	1.81 \pm 0.30
II	40mg/kg bw Indomethacin	3.31 \pm 0.94	8.63 \pm 0.46
III	5mg/kg bw <i>Musa sapientum</i>	4.10 \pm 0.34	3.43 \pm 0.20
IV	10mg/kg bw <i>Musa sapientum</i>	3.08 \pm 0.11	2.83 \pm 0.44
V	20mg/kg bw <i>Musa sapientum</i>	1.78 \pm 0.48	2.45 \pm 0.18
VI	40mg/kg bw Omeprazole	3.08 \pm 0.96	3.38 \pm 0.28

bw = bodyweight, S.E.M. = Standard Error of Mean

Table 3. Results of the statistical analyses of Total Cholesterol, HDL-Cholesterol, Triglycerides and LDL-Cholesterol Levels in the Sera of Rats of Groups I - VI. ($P \leq 0.05$).

Groups of rats	Doses of drugs/extract	Total cholesterol (mg/dl) \pm S.E.M.	HDL-cholesterol (mg/dl) \pm S.E.M.	Triglycerides (mg/dl) \pm S.E.M.	LDL-cholesterol (mg/dl)
I	Physiological saline	112.93 \pm 3.63	93.25 \pm 2.21	40.75 \pm 0.95	11.53
II	40mg/kg bw Indomethacin	256.00 \pm 12.70	62.00 \pm 5.19	59.75 \pm 7.16	182
III	5mg/kg bw <i>Musa sapientum</i>	118.50 \pm 2.63	100.25 \pm 0.85	52.80 \pm 6.17	7.69
IV	10mg/kg bw <i>Musa sapientum</i>	112.25 \pm 0.25	101.25 \pm 2.17	36.28 \pm 1.43	3.74
V	20mg/kg bw <i>Musa sapientum</i>	117.75 \pm 1.89	105.58 \pm 4.55	32.63 \pm 1.68	5.64
VI	40mg/kg bw Omeprazole	124.50 \pm 6.99	91.00 \pm 3.42	69.00 \pm 4.38	19.7

bw = bodyweight, S.E.M. = Standard Error of Mean

Table 4. Results of the statistical analyses of Creatinine, Urea and Billirubin Concentrations in Sera Samples of Rats of Groups I - VI. ($P \leq 0.05$).

Groups of rats	Doses of drugs/extract	Creatinine ($\mu\text{mol/L}$) \pm S.E.M.	Urea (mmol/L) \pm S.E.M.	Bilirubin ($\mu\text{mol/L}$) \pm S.E.M.
I	Physiological saline	0.86 \pm 0.15	74.00 \pm 2.45	1.79 \pm 0.15
II	40mg/kg bw Indomethacin	1.16 \pm 0.14	141.80 \pm 13.6	4.01 \pm 0.86
III	5mg/kg bw <i>Musa sapientum</i>	0.14 \pm 0.04	78.4 \pm 4.13	1.50 \pm 0.12
IV	10mg/kg bw <i>Musa sapientum</i>	0.11 \pm 0.02	75.25 \pm 1.67	1.23 \pm 0.13
V	20mg/kg bw <i>Musa sapientum</i>	0.51 \pm 0.02	67.18 \pm 1.06	0.93 \pm 0.10
VI	40mg/kg bw Omeprazole	0.81 \pm 0.11	81.92 \pm 0.74	2.94 \pm 0.33

bw = bodyweight, S.E.M. = Standard Error of Mean

in the glandular (mucosal) part of the stomach. (Ramzi *et al.*, 1999 and John, 2000). Although, the mechanisms underlying the ulcerogenicity of Indomethacin are not completely understood; it has been known that Indomethacin induces gastric mucosa ulceration through the inhibition of the release of protective factors like cyclooxygenases, prostaglandin E2 (PGE2), bicarbonate, mucus and antioxidants; while aiding vasoconstriction and the increase of aggressive factors such as acid and oxidants. (John, 2000, Maity and Chattopadhyay, 2008 and Suleyman *et al.*, 2010).

Reactive oxygen species such as super oxide anion (O_2^-), singlet oxygen (O^{\cdot}), hydroxyl radical (OH^{\cdot}) and hydrogen peroxide (H_2O_2) generated by ulcerogens disrupt the balance between defensive and aggressive factors maintaining the integrity of gastric epithelial layers (Maity and Chattopadhyay, 2008) resulting in reduction of mucous viscosity thereby affecting the quality of its protection. (John, 2000 and Sen *et al.*, 2009). Reactive oxygen species are, therefore, involved in the aetiopathogenesis of inflammatory and ulcerative lesions of the gastrointestinal tract. (John, 2000 and Maity and Chattopadhyay, 2008).

Indomethacin has equally been established to uncouple mitochondrial respiration resulting in depletion of Adenosine Triphosphate and a reduced potential of gastric epithelial cells to coordinate normal cellular functions. (John, 2000). Furthermore, Indomethacin accumulates within gastric epithelial cells resulting in osmotic movement of water into the cells, swelling and lyses of epithelial cells. (John, 2000). Hence, it reduces the quality and the amount of mucus secretion and changes in ionic permeability characteristics of gastric mucosa (Ramzi *et al.*, 1999, John, 2000 and Maity and Chattopadhyay, 2008).

Antioxidant enzymes and molecules (such as glutathione, superoxide dismutase and catalase) scavenge free radicals thereby reversing the adverse effects produced by their presence. (John, 2000 and Rahman, 2007). Glutathione acts as a co-factor for many detoxifying enzymes, encourages amino acid transport across plasma membranes, directly scavenges singlet oxygen and hydroxyl radicals and helps in the conversion of non-enzymatic antioxidants (vitamins C and E) to their potent forms. (Rahman, 2007). Superoxide dismutase converts superoxide anions to dioxygen and hydrogen peroxide thereby neutralizing their adverse effects on gastric epithelial layers. (Rahman, 2007). Catalase has a very high turnover enzymatic rate and is able to convert about six million molecules of hydrogen peroxide to water and molecular oxygen per minute (Rahman, 2007).

This study observed statistically significant lower activities ($P \leq 0.05$) of superoxide dismutase and catalase in the sera and stomach samples of rats of Groups III – VI when compared to rats of Group II. (Table 1). This implied sustained gastric ulceration through the continuous release of reactive oxygen species in rats of Group II despite the increased antioxidant activities of superoxide dismutase and catalase. *Musa sapientum* and the standard drug (Omeprazole) were probably able to counteract the adverse effects of reactive oxygen species generated by the ulcerogen and were equally able to balance the disrupted equilibrium between aggressive and defensive factors maintaining the integrity of gastric mucosal epithelial layers.

Lipid peroxidation results from oxidative degradation of lipids and involves the stealing of electrons from the lipid cell membrane by free radicals. (Sen *et al.*, 2009 and Akinlolu *et al.*, 2012). It involves a free radical chain reaction mechanism terminated either by the counter-effects of antioxidants such as phospholipid hydroperoxide glutathione peroxidase or production of reactive aldehydes such as malondialdehyde. (Akinlolu *et al.*, 2012). Lipid peroxidation leads to increased oxidative stress, compromised cell membranes and cellular damage; and is increased in inflammatory conditions such as gastric ulceration. (Akinlolu *et al.*, 2012).

Statistically significant lower concentrations ($P \leq 0.05$) of malondialdehyde were observed in the sera and stomach samples rats of Groups III – VI when compared to rats of Group II. (Table 2). This implied increased oxidative stress and cellular damage in rats of Group II. The lowest non-statistically significant ($P \leq 0.05$) malondialdehyde concentration resulted from treatment with 20mg/kg bodyweight of *Musa sapientum* sucker extract (Group V) implying increased antioxidant performance when compared to rats of Group VI treated with the standard drug (Omeprazole) (Table 2). *Musa sapientum* and the standard drug (Omeprazole) were, therefore, possibly able to increase the production of glutathione peroxidase and attenuated the increased malondialdehyde levels due to the effects of the administered ulcerogen.

Phospholipids of the gastric mucus gel layer act as primary barriers to acid-induced damage of the stomach by establishing the hydrophobicity of the stomach's epithelial cellular layer. (John, 2000). Indomethacin interacts adversely with surface-active phospholipids of the gastric mucosa, thereby reducing its hydrophobicity and the quality of its protection against ulcerogens. (John, 2000). Statistically significant lower levels ($P \leq 0.05$) of total cholesterol, low density lipoprotein-cholesterol and triglycerides, but higher levels ($P \leq 0.05$) of high density

lipoprotein-cholesterol were observed in the sera and stomach samples of rats of Group III - VI when compared with rats of Group II (Table 3). The administrations of 10 and 20mg/kg bodyweight of *Musa sapientum* sucker extract (Groups IV and V) were, however, able to attenuate lipid profile status better than the standard drug (Omeprazole) (Table 3).

The observed lipid profile results of rats of Groups III - VI implied that *Musa sapientum* and the standard drug (Omeprazole) attenuated lipid profile status of rats in Indomethacin - induced gastric ulceration. *Musa sapientum* and the standard drug (Omeprazole) were probably able to inhibit the negative interactions of Indomethacin with surface-active phospholipids of the gastric mucosa, thereby increasing its hydrophobicity and the quality of its protection against the ulcerogen.

Creatinine concentration determines the glomerular filtration rate and is the direct measurement of kidney function (Nwagwa, 2012). Similarly, urea and bilirubin are used to determine the functional state of the kidneys. Elevated levels of creatinine, urea and bilirubin are indicative of kidney cellular damage or anomalies of kidney functions (Nwagwa, 2012). Evaluations of kidney function tests showed statistically significant higher concentrations ($P \leq 0.05$) of creatinine, urea and bilirubin in sera samples of rats of Group II when compared to rats of Groups III - VI. (Table 4). Administrations of extract doses of *Musa sapientum* sucker showed attenuated concentrations of creatinine, urea and bilirubin in rats of Groups III - V better than those observed in rats of Group VI. (Table 4). This implied that administrations of doses of *Musa sapientum* sucker extract in wistar rats was well tolerated by body organs and could serve renal protective functions in gastric ulceration.

The precise mechanism of action of *Musa sapientum* sucker is not clear, however, its anti-ulcer properties might be due to its phytochemical components such as tannins, saponins and alkaloids. Tannins being an astringent with vaso-constricting effects (Sabiha *et al.*, 2011) might have precipitated microproteins on the sites of ulcers thereby forming a non-penetrable protective pellicle over the lining to prevent absorption of toxic substances and resisted the attack of proteolytic enzymes. (John and Onabanjo, 1990; Sabiha *et al.*, 2011). Tannins as phenolic compounds are known to enhance the status of oxidative stress biomarkers with the ability to scavenge free radicals, which could also have enhanced its anti-ulcer activity. (Sabiha *et al.*, 2011). *Musa sapientum* was probably able to mobilize mucosa endogenous prostaglandins to attenuate the effects of Indomethacin - induced gastric

ulceration (Borrelli and Izzo, 2000 and Irma *et al.*, 2010).

Saponins possess immunomodulatory, anti-inflammatory and vasoprotective effects which could have aided the observed anti-ulcer activity of *Musa sapientum*. (Borrelli and Izzo, 2000 and Irma *et al.*, 2010). Alkaloids have equally been reported to achieve dose-dependent prevention of ulcer formation, decreased acid secretion, total inhibition of gastric and duodenal ulcers, and even healed chronic acetic acid-induced ulcers (Maity and Chattopadhyay, 2008).

This study observed that administrations of 5, 10 and 20mg/kg bodyweight of *Musa sapientum* sucker extract attenuated the values of kidney functions tests, total antioxidant and lipid profile status of female wistar rats in Indomethacin - induced gastric ulceration. The observed anti-ulcer properties of *Musa sapientum* sucker could be due to its saponins, tannins and alkaloid components.

References

- Akande I.S., Akande B.E. and Gbenle G.O. (2011). Toxicological and antioxidant effects of ethanolic extract of *Baphia nitida* on diazepam induced oxidative stress in rats. *Af. J. of Biochem. Res.* Vol. 5(8): 255 - 263.
- Akinlolu A.A., Sadiq M., Ayoola M.D., Otulana J.O., Abimbola O., Ejiwunmi A.B. (2006). Morphological Gastroprotective Effects of *Heliotropium indicum* on Gastric Ulcerated Mucosa. *Pak. J. of Path.* 17, No 2: 60 - 64.
- Akinlolu A. A., Ayoola M. D., Otulana J. O., Akinola O. B., Abimbola Olayiwola and A. B. Ejiwunmi. (2008). Evaluation of the Histo- Gastroprotective and Antimicrobial Activities of *Heliotropium Indicum*. *Mal. J. of Med. Sci.* Vol. 15, No. 3: 22-30.
- Akinlolu A.A., Salau B.A. and Akingbola T. (2012). Lipid Peroxidation in Nigerians Affected With Hematological Malignancies. *The Af. J. of Med. and Med. Sci. (AJMMS)*. Vol. 41 Suppl: 145 -148.
- Borrelli F. and Izzo A.A. (2000). The Plant Kingdom as a Source of Anti-ulcer Remedies. *Review Article. Phytother. Res.* 14: 581-591.
- Hossain S.M., Alam B.M., Assadujjaman M., Zahan R., Islam M.M., Mazumder M.E.H. and Haque Md.E. (2011). Antidiarrheal, Antioxidant and Antimicrobial Activities of the *Musa sapientum* Seed. *Avicenna J Med Biotechnol.* 3(2): 95-105.
- Irma P., Aghieszka G., Danuta S. (2010). Saponins as cytotoxic agents: a review. *Phytchem. Rev.* 9(3): 425 - 474.
- Jayamurthy P., Aparna B., Gayathri G. and Nisha P. (2013). Evaluation of Antioxidant Potential of Inflorescence and Stalk of Plantain (*Musa sapientum*). *J. of Food Biochem.* 37 (1): 2 - 7.
- John A.T., Onabanjo A.O. (1990). Gastroprotective Effects of an Aqueous Extract of *Entandro Pragma utile* Bark in Experimental Ethanol-Induced Peptic Ulceration in Mice and Rats. *J. Ethnopharm.* 29:87-93.
- John L.W. (2000). How do NSAID's cause ulcer disease? *Bailliere Clin. Gastr.* 14 (1): 147 -159.

- Lewis D.A., Fields W.N., Shaw G.P. (1999). A natural flavonoid present in unripe plantain banana pulp (*Musa sapientum* L. var. *paradisiaca*) protects the gastric mucosa from aspirin-induced erosions. *J. Ethnopharmacol.* 65(3):283-288.
- Maity B. and Chattopadhyay S. (2008). Natural Antiulcerogenic Agents: An Overview. *Curr. Bio. Comp.*, 4: 225-244.
- Nwangwa E. K. The Reno-Protective Effects of Coconut Water on the Kidneys of Diabetic Wistar Rats. *J. of H. Sci.* 2012, 2(1): 1-4 DOI: 10.5923/j.health.20120201.01
- Olayemi S.O., Arikawe A.P., Akinyede A., Oreagba A.I. and Awodele O. (2012). Effects of Malarial Treatments on Biochemical Parameters and Plasma pH of Mice Infected with *Plasmodium berghei*. *Int. J. of Pharm.*, 8:549 – 554.
- Pelin A. K., Zeynep S., Guzin O. K., Yildiz D. and Gulen T. (2002). Zinc treatment affects superoxide dismutase in growth retardation. *Biol. T. E. Res.* Vol. 90, Issue 1 – 3:39 – 46.
- Prabha P., Karpagam T., Varalakshmi B. and Packiavathy A.S.C. (2011). Indigenous anti-ulcer activity of *Musa sapientum* on peptic ulcer. *Pharm. Res.* 3(4): 232 – 238.
- Rahman K. (2007). Studies on free radicals, antioxidants and co-factors. *Clin Interv Aging.* 2(2): 219 – 236.
- Ramakrishnan B., Selvaraj S., Babu S., Radhakrishnan S., Radhakrishnan N. and Palanisamy P. (2011). Antioxidant Potential of Peel Extracts of Banana Varieties (*Musa sapientum*). *Food and Nutrition Sciences.* Vol. 2 Issue 10:1128.
- Ramzi S.C., Vinay K., Turcker C. (1999). *Robbins' Pathological Basis of Disease*, 6th edition, Saunders's Company, pages 795 – 796.
- Sabiha S., Mohd. A.A., Asif M., Aktar M. (2011). Roles of phenolic compounds in peptic ulcer: An overview. *J. Pharm Bioallied Sci.* 3(3): 361-367.
- Salau B. A, Ajani E. O., Akinlolu A. A., Ekor M. N. and Soladoye M. O. (2010). Methanolic Extract of *Musa Sapientum* Sucker Moderates Fasting Blood Glucose and Body Weight of Alloxan Induced Diabetic Rats. *Asian J. of Exp. Biol. Sci.* Number (1) (1): 30 – 35.
- Sedlak J. and Lindsay R.H.L. (1968). Estimation of total and non protein sulfhydryl groups in tissue with Ellman's reagent. *Anal. Biochem.* 25:192-205.
- Sen S., Chakraborty R., De B. and Mazumder J. (2009). Plants and phytochemicals for peptic ulcer: An overview. *Pharm. Rev.* 3(6): 270 – 279
- Sinha A.K. (1972). Colorimetric assay of catalase. *Anal. Biochem.* 47: 389 – 394.
- Suleyman H., Albayrak A., Bilici M., Cadirci E. and Halici Z. (2010). Different mechanisms in formation and prevention of indomethacin-induced gastric ulcers. *Inflammation.* 33(4):224-34.

Cell Membranes and Free Radical Research

SUBSCRIPTION ORDER FORM

I would like to subscribe to **Cell Membranes and Free Radical Research**
(ISSN Numbers : 1308-4178 (online) 1308-416X). All prices in Turkish Liras
2012, Vols. (4 issues)

Intitutional Rates:

Print Only: 30 TL per issue + postage 100 TL per year

Credit Card _____ Expiration Date _____

Print Name _____ Signature _____

Receiving Address

Name _____

Department/Title _____

Street _____

City _____ State/Province _____ Post/Zip Code _____ Country _____

Tel _____ Fax _____ E-mail _____

Billing Address Check here if same as above

Name _____

Department/Title _____

Street _____

Cell Membranes and Free Radical Research

LIBRARY ROUTING CARD

To (Librarian) : _____ Date _____

From: _____ Department _____

I have reviewed **Cell Membranes and Free Radical Research**

(ISSN Numbers : 1308-4178 (online) 1308-416X) and suggest that you subscription with:

2012, Vols. (4 issues) All prices in Turkish Liras.

Intitutional Rates:

Print Only: 30 TL + postage 100 TL per year

Süleyman Demirel Üniversitesi Tıp Fakültesi, Isparta / TURKEY

Tel:+ 90 246 211 37 08 Fax:+ 90 246 237 11 65 EMAIL: mustafanaziroglu@sdu.edu.tr

COPYRIGHT FORM

Cell Membranes and Free Radical Research

Date _____ Contributor Name _____

Contributer Address _____

Manuscript Number (if Know) _____

Re: Manuscript entitled _____

"For publication in Cell Membranes and Free Radical Research published by Society of Cell Membranes and Free Oxygen Radicals "

Submission of a manuscript implies:

- that the work described has not been published before (except in the form of an abstract or as part of a published lecture, review or thesis);
- that it is not under consideration for publication elsewhere;
- that its publication has been approved by all co-authors, if any, as well as by the responsible authorities at the institute where the work has been carried out;
- that, if and when the manuscript is accepted for publication, the authors agree to automatic transfer of the copyright to this publisher;
- that the manuscript will not be published elsewhere in any language without the consent of the copyright holders;
- that written permission of the copyright holder is obtained by the authors for material used from other copyrighted sources, and that any costs associated with obtaining this permission are the authors' responsibility. Copyright notice: The contributor and the company/employer agree that all copies the final published version of the contribution or any part thereof distributed or posted by them in print or electronic format as permitted herein will include the notice of copyright as stipulated in the journal and a full citation to the journal as published by Cell Membranes and Free radical Society, Isparta, Turkey.

CHECK ONE BOX

Contributor owned work: Contributor's signature _____ Date _____
Type or print name and title _____
Co-Contributor's signature _____ Date _____
Type or print name and title _____

Company-Institution owned work _____
Company-Institution (Employer for here) _____ Date _____
Authorized signature _____ Date _____

U.S. Government work _____

U.K. Government work _____

Other Government work _____