ABOUT IJMMS

The International Journal of Medicine and Medical Sciences is published monthly (one volume per year) by Academic Journals.

The International Journal of Medicine and Medical Sciences (IJMMS) provides rapid publication (monthly) of articles in all areas of Medicine and Medical Sciences such as:

Clinical Medicine: Internal Medicine, Surgery, Clinical Cancer Research, Clinical Pharmacology, Dermatology, Gynaecology, Paediatrics, Neurology, Psychiatry, Otorhinolaryngology, Ophthalmology, Dentistry, Tropical Medicine, Biomedical Engineering, Clinical Cardiovascular Research, Clinical Endocrinology, Clinical Pathophysiology, Clinical Immunology and Immunopathology, Clinical Nutritional Research, Geriatrics and Sport Medicine

Basic Medical Sciences: Biochemistry, Molecular Biology, Cellular Biology, Cytology, Genetics, Embryology, Developmental Biology, Radiobiology, Experimental Microbiology, Biophysics, Structural Research, Neurophysiology and Brain Research, Cardiovascular Research, Endocrinology, Physiology, Medical Microbiology

Experimental Medicine: Experimental Cancer Research, Pathophysiology, Immunology, Immunopathology, Nutritional Research, Vitaminology and Ethiology

Preventive Medicine: Congenital Disorders, Mental Disorders, Psychosomatic Diseases, Addictive Diseases, Accidents, Cancer, Cardiovascular Diseases, Metabolic Disorders, Infectious Diseases, Diseases of Bones and Joints, Oral Preventive Medicine, Respiratory Diseases, Methods of Epidemiology and Other Preventive Medicine

Social Medicine: Group Medicine, Social Paediatrics, Medico-Social Problems of the Youth, Medico-Social Problems of the Elderly, Rehabilitation, Human Ecology, Environmental Toxicology, Dietetics, Occupational Medicine, Pharmacology, Ergonomy, Health Education, Public Health and Health Services and Medical Statistics

The Journal welcomes the submission of manuscripts that meet the general criteria of significance and scientific excellence. Papers will be published approximately one month after acceptance. All articles published in IJMMS are peer-reviewed.

Submission of Manuscript

Submit manuscripts as e-mail attachment to the Editorial Office at: ijmms@academicjournals.org. A manuscript number will be mailed to the corresponding author.

The International Journal of Medicine and Medical Sciences will only accept manuscripts submitted as e-mail attachments.

Please read the Instructions for Authors before submitting your manuscript. The manuscript files should be given the last name of the first author.
Editors

Dr. J. Ibekwe  
Acting Editor-in-chief,  
International Journal of Medicine and Medical Sciences Academic Journals  
E-mail: ijmms.journals@gmail.com  
http://www.academicjournals.org/ijmms

Afrozul Haq  
Editor, Laboratory Medicine  
Department of Laboratory Medicine  
Sheikh Khalifa Medical City  
P.O. Box 51900, ABU DHABI  
United Arab Emirates
Editorial Board

Chandrashekhar T. Sreeramareddy  
Department of Community Medicine,  
P O Box No 155, Deep Heights  
Manipur College of Medical Sciences,  
Pokhara,  
Nepal

Professor Viroj Wiwanitkit  
Wiwanitkit House, Bangkhae,  
Bangkok  
Thailand 10160

Dr. Srinivas Koduru  
Dept of Clinical Sciences  
Collage of Health Sciences  
University of Kentucky  
Lexington USA

Weiping Zhang  
Department of Oral Biology  
Indiana University School of Dentistry  
1121 West Michigan Street, DS 271  
Indianapolis, IN 46202  
USA

Dr. santi M. Mandal  
Internal Medicine  
UTMB, Galveston, TX,  
USA

Konstantinos Tziomalos  
Department of Clinical Biochemistry  
(Vascular Prevention Clinic),  
Royal Free Hospital Campus,  
University College Medical School, University College  
London, London,  
United Kingdom

Lisheng XU  
Ho Sin Hang Engineering Building  
Department of Electronic Engineering  
The Chinese University of Hong Kong  
Shatin, N.T. Hong Kong,  
China

Dr. santi M. Mandal  
Internal Medicine  
UTMB, Galveston, TX,  
USA

Weiping Zhang  
Department of Oral Biology  
Indiana University School of Dentistry  
1121 West Michigan Street, DS 271  
Indianapolis, IN 46202  
USA

Dr. Mustafa Sahin  
Department of Endocrinology and Metabolism  
Baskent University,  
Ankara,  
Turkey

Mojtaba Salouti  
School of Medical and Basic Sciences,  
Islamic Azad University- Zanjan,  
Iran

Dr. Harshdeep Joshi  
Maharishi Markandeshwar  
Institute of Medical Sciences and Research  
Ambala, (Haryana),  
India

Sisira Hemananda Siribaddana  
259, Temple Road, Thalapathpitiya,  
Nugegoda, 10250  
Sri Lanka

Dr. santi M. Mandal  
Internal Medicine  
UTMB, Galveston, TX,  
USA

Cyri Chukwudi Dim  
Department of Obstetrics & Gynaecology  
University of Nigeria Teaching Hospital (UNTH)  
P.M.B. 01129, Enugu. 400001,  
Nigeria

Dr. Mustafa Sahin  
Department of Endocrinology and Metabolism  
Baskent University,  
Ankara,  
Turkey

Mojtaba Salouti  
School of Medical and Basic Sciences,  
Islamic Azad University- Zanjan,  
Iran

Imtiaz Ahmed Wani  
Srinagar Kashmir, 190009,  
India
Instructions for Author

Electronic submission of manuscripts is strongly encouraged, provided that the text, tables, and figures are included in a single Microsoft Word file (preferably in Arial font).

The cover letter should include the corresponding author’s full address and telephone/fax numbers and should be in an e-mail message sent to the Editor, with the file, whose name should begin with the first author’s surname, as an attachment.

Article Types
Three types of manuscripts may be submitted:

Regular articles: These should describe new and carefully confirmed findings, and experimental procedures should be given in sufficient detail for others to verify the work. The length of a full paper should be the minimum required to describe and interpret the work clearly.

Short Communications: A Short Communication is suitable for recording the results of complete small investigations or giving details of new models or hypotheses, innovative methods, techniques or apparatus. The style of main sections need not conform to that of full-length papers. Short communications are 2 to 4 printed pages (about 6 to 12 manuscript pages) in length.

Reviews: Submissions of reviews and perspectives covering topics of current interest are welcome and encouraged. Reviews should be concise and no longer than 4-6 printed pages (about 12 to 18 manuscript pages). Reviews are also peer-reviewed.

Review Process
All manuscripts are reviewed by an editor and members of the Editorial Board or qualified outside reviewers. Authors cannot nominate reviewers. Only reviewers randomly selected from our database with specialization in the subject area will be contacted to evaluate the manuscripts. The process will be blind review. Decisions will be made as rapidly as possible, and the journal strives to return reviewers’ comments to authors as fast as possible. The editorial board will re-review manuscripts that are accepted pending revision. It is the goal of the IJMMS to publish manuscripts within 8 weeks after submission.

Regular articles
All portions of the manuscript must be typed double-spaced and all pages numbered starting from the title page.

The Title should be a brief phrase describing the contents of the paper. The Title Page should include the authors’ full names and affiliations, the name of the corresponding author along with phone, fax and E-mail information. Present addresses of authors should appear as a footnote.

The Abstract should be informative and completely self-explanatory, briefly present the topic, state the scope of the experiments, indicate significant data, and point out major findings and conclusions. The Abstract should be 100 to 200 words in length. Complete sentences, active verbs, and the third person should be used, and the abstract should be written in the past tense. Standard nomenclature should be used and abbreviations should be avoided. No literature should be cited.

A list of non-standard Abbreviations should be added. In general, non-standard abbreviations should be used only when the full term is very long and used often. Each abbreviation should be spelled out and introduced in parentheses the first time it is used in the text. Only recommended SI units should be used. Authors should use the solidus presentation (mg/ml). Standard abbreviations (such as ATP and DNA) need not be defined.

The Introduction should provide a clear statement of the problem, the relevant literature on the subject, and the proposed approach or solution. It should be understandable to colleagues from a broad range of scientific disciplines.

Materials and methods should be complete enough to allow experiments to be reproduced. However, only truly new procedures should be described in detail; previously published procedures should be cited, and important modifications of published procedures should be mentioned briefly. Capitalize trade names and include the manufacturer’s name and address. Subheadings should be used. Methods in general use need not be described in detail.
Results should be presented with clarity and precision. The results should be written in the past tense when describing findings in the authors’ experiments. Previously published findings should be written in the present tense. Results should be explained, but largely without referring to the literature. Discussion, speculation and detailed interpretation of data should not be included in the Results but should be put into the Discussion section.

The Discussion should interpret the findings in view of the results obtained in this and in past studies on this topic. State the conclusions in a few sentences at the end of the paper. The Results and Discussion sections can include subheadings, and when appropriate, both sections can be combined.

The Acknowledgments of people, grants, funds, etc should be brief.

Tables should be kept to a minimum and be designed to be as simple as possible. Tables are to be typed double-spaced throughout, including headings and footnotes. Each table should be on a separate page, numbered consecutively in Arabic numerals and supplied with a heading and a legend. Tables should be self-explanatory without reference to the text. The details of the methods used in the experiments should preferably be described in the legend instead of in the text. The same data should not be presented in both table and graph form or repeated in the text.

Figure legends should be typed in numerical order on a separate sheet. Graphics should be prepared using applications capable of generating high resolution GIF, TIFF, JPEG or Powerpoint before pasting in the Microsoft Word manuscript file. Tables should be prepared in Microsoft Word. Use Arabic numerals to designate figures and upper case letters for their parts (Figure 1). Begin each legend with a title and include sufficient description so that the figure is understandable without reading the text of the manuscript. Information given in legends should not be repeated in the text.

References: In the text, a reference identified by means of an author’s name should be followed by the date of the reference in parentheses. When there are more than two authors, only the first author’s name should be mentioned, followed by ‘et al’. In the event that an author cited has had two or more works published during the same year, the reference, both in the text and in the reference list, should be identified by a lower case letter like ‘a’ and ‘b’ after the date to distinguish the works.

Examples:

Nishimura (2000), Agindotan et al. (2003), (Kelebeni, 1983), (Usman and Smith, 2001), (Chege, 1998; Stein, 1987a,b; Tijani, 1993,1995), (Kumasi et al., 2001)

References should be listed at the end of the paper in alphabetical order. Articles in preparation or articles submitted for publication, unpublished observations, personal communications, etc. should not be included in the reference list but should only be mentioned in the article text (e.g., A. Kingori, University of Nairobi, Kenya, personal communication). Journal names are abbreviated according to Chemical Abstracts. Authors are fully responsible for the accuracy of the references.

Examples:


Case Studies

Case Studies include original case reports that will deepen the understanding of general medical knowledge.

The Title should be a brief phrase describing the contents of the paper. The Title Page should include the authors' full names and affiliations, the name of the corresponding author along with phone, fax and E-mail information. Present addresses of authors should appear as a footnote.

The Abstract should be informative and completely self-explanatory, briefly present the topic, state the scope of the experiments, indicate significant data, and point out major findings and conclusions. The Abstract should be 100 to 200 words in length. Complete sentences, active verbs, and the third person should be used, and the abstract should be written in the past tense. Standard nomenclature should be used and abbreviations should be avoided. No literature should be cited.

Following the abstract, about 3 to 10 key words that will provide indexing references should be listed.

A list of non-standard Abbreviations should be added. In general, non-standard abbreviations should be used only when the full term is very long and used often. Each abbreviation should be spelled out and introduced in parentheses the first time it is used in the text. Only recommended SI units should be used. Authors should use the solidus presentation (mg/ml).

The Introduction should provide a clear statement of the problem, the relevant literature on the subject, and the proposed approach or solution. It should be understandable to colleagues from a broad range of scientific disciplines.

The presentation of the case study should include the important information regarding the case. This must include the medical history, demographics, symptoms, tests etc. Kindly note that all information that will lead to the identification of the particular patient(s) must be excluded.

The conclusion should highlight the contribution of the study and its relevance in general medical knowledge.

The Acknowledgments of people, grants, funds, etc should be brief.

References: Same as in regular articles.

Short Communications

Short Communications are limited to a maximum of two figures and one table. They should present a complete study that is more limited in scope than is found in full-length papers. The items of manuscript preparation listed above apply to Short Communications with the following differences: (1) Abstracts are limited to 100 words; (2) instead of a separate Materials and Methods section, experimental procedures may be incorporated into Figure Legends and Table footnotes; (3) Results and Discussion should be combined into a single section.

Proofs and Reprints: Electronic proofs will be sent (e-mail attachment) to the corresponding author as a PDF file. Page proofs are considered to be the final version of the manuscript. With the exception of typographical or minor clerical errors, no changes will be made in the manuscript at the proof stage. Because IJMMS will be published freely online to attract a wide audience, authors will have free electronic access to the full text (in both HTML and PDF) of the article. Authors can freely download the PDF file from which they can print unlimited copies of their articles.

Copyright: 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, or thesis) that it is not under consideration for publication elsewhere; that if and when the Manuscript is accepted for publication, the authors agree to automatic transfer of the copyright to the publisher.
ARTICLES

Effect of chronic administration of Aloe vera extract on plasma biochemistry in rabbits

The effect of ozonized saline solutions processed under intense electric fields in the treatment of infected necrotizing acute pancreatitis: An experimental mode
Nadim Al-Hajjar

Anatomical variants, clinical presentation and pathological findings in patients suffering from chronic rhinosinusitis underwent functional endoscopic sinus surgery
Seyyed Abdollah Madani, Seyyed Abbas Hashemi, Shahzad Javan and Akram Alsadat Hoseini
The effect of ozonized saline solutions processed under intense electric fields in the treatment of infected necrotizing acute pancreatitis: An experimental mode

Nadim Al-Hajjar

Department of Surgery, UMF Iuliu Hatieganu Cluj-Napoca, Romania.

Received 20 August, 2012; Accepted 16 June, 2014

The aim of this study was to study the effect of ozonized saline solutions administered intraperitoneally in rabbits suffering from severe acute pancreatitis with infected necrosis. The acute pancreatitis was induced by infusion of sodium taurocholate into the main pancreatic duct and in the next 24 hours, a cecal fistula was created to obtain infection of the pancreatic necrosis with intestinal germs. The rabbits were divided into 3 groups of study and each of them underwent continuous peritoneal lavage 2 times a day for 5 days with simple saline (control group), ozonized saline solution 5 mg/L (group A) and respectively ozonized saline solution 9 mg/L processed under an intense electric field (group B). Serum level of C-reactive protein and total peroxides, bacterial content of the peritoneal liquid and tissue specimens from the pancreas and the peripancreatic area were evaluated. Six days after the induction of pancreatitis all surviving animals were sacrificed. Serum levels of C-reactive protein and total peroxides in rabbits treated with ozonized saline solution were significantly lower as compared to those of rabbits belonging to the control group. Regarding bacterial growth, a significant decrease was obtained in the peritoneal liquid or even no bacterial growth in groups A and B. Histological examination of the tissue showed specific pancreatic changes in all the groups, but less expressed in groups A and B. No significant differences were encountered between rabbits treated with different ozone concentrations. Intraperitoneal ozone therapy is effective in the amelioration of acute pancreatitis by means of laboratory analysis and decreased bacterial growths.

Key words: Severe acute pancreatitis, ozone-therapy, infection of pancreatic necrotic tissues, intense electric fields.

INTRODUCTION

Acute pancreatitis, one of the most frequent gastroenterological diseases, is a leading cause of morbidity and mortality, still having a major economical and emotional impact. Despite later progress in understanding its mechanisms and improving the management strategies, its prognosis did not change significantly as the mortality rates are still high (10 to 50%) (Bhatia, 2002).

Generally, the disease progresses in two phases. The early phase, in the first week, is characterized by a systemic inflammatory response syndrome (SIRS) where the
late phase is characterized by the presence of complications and it appears only in severe forms of pancreatitis. Infectious complication of the necrotic tissues that occurs in 50% of the cases of severe acute pancreatitis is the leading cause of death, responsible for 70 to 80% of all deaths due to acute pancreatitis (Schmid et al., 1999). Failure in prevention of infection and management of sepsis (Dellinger et al., 2007; Marincaş et al., 2006) shows the need for new therapeutic agents.

Used initially for potable water treatment, ozone is now regarded as one of the most powerful oxidizing agents having important antiseptic and antibacterial effects (Bialoszewski et al., 2010).

Proven antibacterial properties in *vitro*, ozone is recommended for application in medicine as an adjunct or alternative treatment to combat various local or systemic infections, especially in those cases where traditional therapy has not given satisfactory results (Bocci, 2007; Dyas et al., 1983; Bialoszewski and Kowalewski, 2003; Lipatov et al., 2002; Silva et al., 2009; Parkhisenko and Glukhov, 2001). Use of ozone in medicine remains controversial because in high concentrations in gaseous form, the compound has a toxic effect on the human body, more pronounced in the respiratory system (Guanche et al., 2010; Bocci et al., 2009). On the other hand, the use of aqueous ozone in different environments, in low concentrations has a beneficial effect on the body both by stimulating host defense mechanisms, and through a direct antimicrobial effect (Oizumi et al., 1998).

Although in *vitro* antibacterial activity of O₃ is obvious even just a few minutes after application (Bocci, 1996), its short life and limited concentration that can be obtained in aqueous solutions significantly decreases its potential action in *vivo*. Applying a high frequency current between certain parameters of intensity, pulse shape, temperature and time of action upon aqueous solutions or upon biological materials generates ozone by corona discharge effect (Kogelschatz and Eliaison, 1995).

This study aimed to investigate the anti-inflammatory and antibacterial effects of ozonized saline solution on an experimental model of severe acute pancreatitis infected with intestinal flora. In different concentrations of ozone, maximally ozonized saline solution produced by ozonator will be compared with similar solution further processed in intense electric fields to assess possible differences between their therapeutic effects.

**MATERIALS AND METHODS**

Surgical interventions in this study were performed at the Center for Experimental Medicine of the University of Medicine and Pharmacy "Iuliu Hatieganu" Cluj-Napoca. The Ethical Commission of "Iuliu Hatieganu" University of Medicine and Pharmacy Cluj-Napoca approved the experimental procedures of the study. A total of 30 Belgian white rabbits were included in the study, weighing 2400 to 2950 g housed in standard individual cages, with an average temperature of 22°C. In all 30 rabbits, we induced severe acute pancreatitis by injecting sodium taurocholate in the main pancreatic duct, under high pressure (Kudari et al., 2007). The animals received intramuscular acepromazine 1%, 10 mg/kg for sedation and after 10 min; anesthesia was induced by intramuscularly administration of a mixture of xylazine with ketamine (dosages of 10 and 50 mg/kg body weight, respectively). The maintenance during surgery was obtained by intravenous administration of xylazine and ketamine, with a dosage of 5 mg/kg each, oxygen supply through a mask for spontaneous breath.

A median subxiphoidian incision of around 10 to 12 cm was done followed by identification of the main pancreatic duct at approximately 2 cm from the pylorus near the opening in the jejunum and cannulation with a 30 G needle. A total of 2 ml of 5% sodium taurocholate in saline was slowly infused. The intestinal loops were reintroduced in the peritoneal cavity and a double layer laparoraphy was performed with continuous suture using absorbable thread (Mersilene 1). After surgery, the animals were reintroduced in cages with free access to food and water.

**Induction of acute pancreatitis**

In all 30 rabbits, we induced severe acute pancreatitis by injecting sodium taurocholate in the main pancreatic duct, under high pressure (Kudari et al., 2007). The animals received intramuscular acepromazine 1%, 10 mg/kg for sedation and after 10 min; anesthesia was induced by intramuscularly administration of a mixture of xylazine with ketamine (dosages of 10 and 50 mg/kg body weight, respectively). The maintenance during surgery was obtained by intravenous administration of xylazine and ketamine, with a dosage of 5 mg/kg each, oxygen supply through a mask for spontaneous breath.

A median subxiphoidian incision of around 10 to 12 cm was done followed by identification of the main pancreatic duct at approximately 2 cm from the pylorus near the opening in the jejunum and cannulation with a 30 G needle. A total of 2 ml of 5% sodium taurocholate in saline was slowly infused. The intestinal loops were reintroduced in the peritoneal cavity and a double layer laparoraphy was performed with continuous suture using absorbable thread (Mersilene 1). After surgery, the animals were reintroduced in cages with free access to food and water.

**Infection of the necrotizing acute pancreatitis**

After 24 h, experimental animals were subjected to a new laparotomy under same anesthesia. Induction of intraperitoneal infection was achieved by creating a cecal fistula with a 16 G needle. Viscera were reintroduced inside the peritoneal cavity.

**Treatment groups**

A system of wash-drainage of the peritoneal cavity consisting of 2 silicone tubes 18 Ch placed near by the pancreas was performed. The 30 rabbits with acute pancreatitis and cecal fistula were randomized into 3 groups and subjected to peritoneal lavage treatment as follows: Control group of 6 rabbits who underwent intraperitoneal lavage with isotonic saline solution (saline); group A of 12 rabbits undergoing intraperitoneal lavage with 5% ozonized saline solution; group B of 12 rabbits who underwent intraperitoneal lavage with ozonized saline processed in intense electric fields.

Peritoneal lavage was initiated 2 h after the creation of the cecal fistula being performed twice a day (every 12 h) for 4 days, 200 ml per session. Duration of each lavage session was 10 min with the solution maintained at a temperature of 36°C. Ozonized saline solution 5% for group A was obtained in an ozone generator type COM-AD-01-IP (Anser, Germany) with an ozonometer type GM-6000-OEM (Anser, Germany) regulating the inflow of 100% oxygen, a flux of O₂ flow=50 l/h, at a 0.2 atmospheric pressure of oxygen, bubbling time of O₂ in aqueous solution=10 min at 20 to 22°C.

For group B, saline solution (normal saline) was used in which ozone produced by the generator (concentration 5 mg/l) was have bubbled, and was immediately subjected to a corona discharge in an oxygen environment using an alternating power (U = 20 kV, I = 0.1 mA and a discharge gap of 20 mm), thus obtaining a concentration of ozone of about 9 mg/L.

**Sample collection (blood, histology, cultures)**

Five days after the second laparotomy, all animals were subjected to a new intervention. After onset of anesthesia, blood samples (2 ml) were obtained from each rabbit by venous puncture of one of the limbs. Pancreatic and peripancreatic tissue was taken for histopathological examination and intraperitoneal fluid samples for bacteriological examination. All animals were sacrificed afterwards receiving an overdose of anesthetic agent.
**Table 1. Score of histopathological modifications.**

<table>
<thead>
<tr>
<th>Score</th>
<th>0</th>
<th>1</th>
<th>2</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Edema</td>
<td>Absent</td>
<td>In the interlobular septum</td>
<td>Mild or interacin</td>
<td>Diffuse</td>
</tr>
<tr>
<td>Hemorrhage</td>
<td>Absent</td>
<td>In the interlobular septum</td>
<td>Mild or interglandular</td>
<td>Diffuse</td>
</tr>
<tr>
<td>Necrosis</td>
<td>Absent</td>
<td>In 1-2 lobules</td>
<td>In 3-4 lobules</td>
<td>More than 5 lobules</td>
</tr>
<tr>
<td>Leukocytes infiltration</td>
<td>Absent</td>
<td>In 1-2 lobules</td>
<td>In 3-4 lobules</td>
<td>More than 5 lobules</td>
</tr>
<tr>
<td>Citosteatonecrosis</td>
<td>Absent</td>
<td>Mild</td>
<td>Moderate</td>
<td>Severe</td>
</tr>
<tr>
<td>Fibrosis</td>
<td>Absent</td>
<td>In 1-2 lobules</td>
<td>In 3-4 lobules</td>
<td>More than 5 lobules</td>
</tr>
</tbody>
</table>

Hematological and biochemical analysis were performed in the laboratory of medical analysis at the Cluj-Napoca Rehabilitation Center, bacteriological examinations in the Department of Microbiology of University of Veterinary Medicine, Cluj-Napoca.

C-reactive protein and the total amount of peroxides for indirect assessment of oxidative stress was investigated. Identification of the pathogenic agents was performed by inoculating the samples on sheep blood nutrient agar and Mueller Hinton agar and then incubated at 37 ±10°C for 24 h. Following the differentiation between morphology, culture and biochemical preliminaries, colonies were passed on API galleries (API Staph, API 20 Strep, API 20E, API 20 NE). Galleries were incubated at 37 ±10°C for 24 h and then read by the APIWEB program. Afterwards, they were included under different categories of species according to the codes obtained on reading of the galleries. The tests used to identify each type of microorganism was mannoit fermentation test, citrate plasma clotting test, catalase test and oxidase test.

Histopathological examinations were performed in the Department of Pathology of the University of Veterinary Medicine, Cluj-Napoca. They analyzed macroscopic changes occurring in pancreatic and peripancreatic tissues and in the intraperitoneal fluid.

Tissue specimens were fixed in formalin (10%, pH=7) for 24 h, then embedded in paraffin and cut into 4-μm; slides were stained with hematoxylin and eosin. Microscopic examination described specific histological features found in acute pancreatitis: edema, inflammatory infiltration, fat necrosis, parenchymal necrosis and hemorrhagic vascular lesions and fibrosis. Microscopic images were obtained with an Olympus BX 51 microscope, captured with an Olympus SP 350 digital camera and processed using the "Cell B software" program.

Statistical analysis

The results were expressed as mean ± standard deviation (SD). Student t test was used to assess statistical significance between groups; correlations between different variables were studied using Mann-Whitney U test (SPSS software). P values were considered statistical significant at <0.05.

**RESULTS**

The observation period for each animal included in the study was 6 days. Out of the six rabbits in the control group, two died (33.3% death rate). One of them died 30 h after induction of pancreatitis (6 h after the cecal fistula) and the other on the third day after creation of the cecal fistula. In group A, two rabbits died on the 4 and 5th days (death rate 16.6%) while in group B there has been a single death, on the 4th day (8.33% death rate) from the beginning of the study.

Serum levels of C-reactive protein, analysed on the 6th day from the induction of acute pancreatitis, were significantly lower in groups A (0.508±0.267 vs. 1.140±0.114 mg/dl, p=0.0001) and B (0.550±0.124 vs. 1.140±0.114 mg/dl, p=0.0001) than controls, but without statistical significance between the two groups treated with ozonized solution. The values of total peroxides were significantly lower in groups A (758.08±196.75 mmol/L, p=0.021) and B (627.716±171.95, p=0.001) compared to the control group, without significant differences in antioxidant property of the two ozonized solutions.

Bacteriological examination of intraperitoneal fluid harvested five days after contamination of the pancreatic necrotic tissues showed the presence of infection in all surviving rabbits in the control group. The organisms identified in various combinations include *Escherichia coli, Staphylococcus aureus, Enterococcus faecalis* and *Pseudomonas aeruginosa*. Bacterial cultures obtained from two rabbits in group A and three rabbits in group B were negative. In all positive samples obtained from groups A and B, infection was monomicrobial or with two germs, unlike the control group where we found up to four different species in the same sample. Thus, peritoneal contamination was significantly lower in group A (p=0.002) and group B (p=0.001) compared to the control group. No statistical significance was found between the number of bacterial strains in peritoneal fluid from animals in groups A and B (p=0.603), and no absolute antibacterial effect of ozonized saline drips were observed on any of the four bacteria present.

The global effects of the two ozonized solutions tested on necrotic acute pancreatitis infected with intestinal flora were assessed by means of pancreatic histopathological changes. Histopathological criteria followed were edema, hemorrhage, leukocyte infiltration, pancreatic necrosis, peripancreatic fat necrosis and fibrosis (Yilmaz et al., 2009). Each parameter received a grade from 0 to 3 (Table 1), and by adding the score, total histopathological score was obtained. Histopathological changes as edema, hemorrhage and necrosis of pancreatic parenchyma were significantly more pronounced in the control group than the two other groups. Regarding the fibroblast proliferation, there were no major differences between
Figure 1. Histopathological aspects of pancreatic and peripancreatic tissues belonging to the rabbits in the control group I, without treatment: (A) diffuse parenchymal necrosis, severe (*), demarcated from normal tissue by a band of neutrophilic and macrophagic leukocyte, HE Bar = 500, (B) necrotico-purulent foci with abscess formation, inflammatory edema, hemorrhage, massive infiltration of neutrophils and macrophages, HE Bar = 500, (C) interlobular, interstitial and intraglandular serofibrinous exudates with parenchymal dissociation, HE Bar = 500, (D) diffuse peripancreatic citosteatonecroză with neutrophilic and macrophagic infiltration, HE Bar = 100, (E) septal fibroblast proliferation at interlobular and interacinar level with parenchymal atrophy, vacuolar degeneration of pancreatic cells, moderate inflammatory infiltrate with neutrophils and rare eosinophils, HE Bar = 100, (F) congestion, edema, leucodiapedesis and perivascular fibroplasia in the peripancreatic tissues (bar, 200 μm).

Overall, the pancreatic and peripancreatic histological characteristics were statistical significant more altered in the control group than the group A (U=0, p=0.001) and B (U=0, p=0.001) (Table 1, Figures 1A, B, C, D, E, F, 2A, B, 3A, B, C, and D).

DISCUSSION

The most important factor in the evolution of acute severe pancreatitis is infection (Bourgaux et al., 2007). Mortality associated with infection of the pancreatic and
Figure 2. Histopathological aspects of the pancreatic tissues of the rabbits belonging to the study group treated with ozonated saline: (A) focal parenchymal necrosis with the formation of microabceses, edema, hemorrhage, mild neutrophilic inflammatory infiltrate and fibroblast proliferation in adjacent areas, HE Bar = 500, (B) fibroblast proliferation and edema at interlobular interstitial and intraglandular level with ductal swelling, reduced inflammatory infiltrate of neutrophils and macrophages, HE Bar = 200.

Figure 3. Histopathological aspects of pancreatic tissues of the rabbits belonging to the study group B treated with ozonated saline processed in intense electric fields: (A and B) focal parenchymal necrosis, edema, hemorrhage, moderate neutrophilic inflammatory infiltrate and fibroblast proliferation in adjacent areas, HE Bar = 500, (C) focal parenchymal necrosis, edema, hemorrhage and discrete inflammatory infiltrate, HE Bar = 200, (D) fibroblast proliferation edema at interlobular interstitial and intraglandular level with swelling of the ducts, HE Bar.
peripancreatic necrotic tissues remains high, despite administration of expensive but highly active broad spectrum antibiotics (De Waele et al., 2004; Lilja et al., 2008). Finding new means of treatment to control infection of the necrotic acute pancreatitis is still a challenge today.

Ozone is a known oxidizing agent in aqueous solutions and a reliable antimicrobial agent. In acute pancreatitis with infected necrosis, peritoneal lavage is very important; as it reduces the microbial load as well as the amount of inflammatory mediators arrived in the extravascular space along with the reduction of intra-abdominal pressure (Georgescu et al., 2005; D'Egido, 1991). In this study, we obtained a significant reduction in C-reactive protein levels and total peroxides amount in those animals treated with ozone compared with the animals belonging to the saline control group. C-reactive protein (CRP) is a simple, cheap and reliable test for indirect assessment of severity of acute pancreatitis offering a good prognostic accuracy for pancreatic necrosis and mortality (Cardoso et al., 2013).

These results correlate with the histopathological changes occurring in the pancreas. Potential augmentation of the inflammatory process and oxidative stress caused by a larger amount of free oxygen radicals corresponding to a higher concentration of ozone was not confirmed between groups treated with ozonized saline 5 and 9% as this was not statistical significant. The continuous peritoneal lavage with ozonized solution which reduces the action of free oxygen radicals has a beneficiary effect on microcirculation and activation of the immune system (Bourgaux et al., 2007).

Various studies have investigated the antibacterial effect of ozone in infectious diseases, with the premise that reactive oxygen species generated by the interaction of ozone with pathogenic microorganisms is the most natural and effective antimicrobial agent. In conditions such as abscesses, perianal fistulas, furunculosis, osteomyelitis, vulvovaginitis, and necrotizing fascitis, ozone therapy had dramatic effects on eradication of the infection and on fastening the healing (Madej et al., 1995). In our study, ozonized saline drips showed a significant antibacterial activity. Administered intraperitoneally, ozone significantly reduced the number of bacterial strains from infected pancreatitis in the groups treated as compared to the control group. Higher concentration of ozone used in animals belonging to group B showed a more pronounced bactericidal effect as compared to 5% ozonized serum used in group A (but not statistical significant).

A research group showed in a study that ozone reduced the severity of pancreatitis and prevented bacterial translocation to the pancreas, liver, cecum, and peritoneum besides improving the survival rates (Uysal et al., 2010). There is only one human study about the efficacy of ozone treatment in acute pancreatitis (Kopchak et al., 2008).

Conclusions

In this study, all the biochemical and histological data indicate that ozone therapy had ameliorative effects on acute pancreatitis. Intraperitoneal continuous lavage with saline drips processed in intense electric fields has real benefits in acute pancreatitis with infected necrosis through significant antimicrobial action, consecutively diminishing tissue injury induced by inflammation mediators. Further research needs to be done regarding different concentrations of ozone related to increased antiinflammatory and antibacterial effect.

ACKNOWLEDGEMENTS

The research was funded by PNII grant - Ideas Code 829/2008 by CNCSIS-UEFISCDI.

Conflict of Interests

The author(s) have not declared any conflict of interests.

REFERENCES


Georgescu I, Nemes R, Cartidges D, Cârju D, Surîn V, Mârgăritescu Al-Hajjar 181
Effect of chronic administration of Aloe vera extract on plasma biochemistry in rabbits

Amjad Ali Channa¹*, Saeed Ahmed Soomro², Roshan Ali Korejo¹, Band-e-Ali Khaskeli¹, Tofique Ahmed Qureshi³, Imtiaz Ahmed Shah¹, Nawab Ali Kalhoro¹ and Hinesh Kumar Maheshwari¹

¹Faculty of Animal Husbandry and Veterinary Sciences, Sindh Agriculture University Tandojam, Pakistan.
²Department of Veterinary Physiology and Biochemistry, Faculty of Animal Husbandry and Veterinary Sciences, Sindh Agriculture University Tandojam, Pakistan.
³Department of Pharmacology, Faculty of Animal Husbandry and Veterinary Sciences, Sindh Agriculture University Tandojam, Pakistan.

Received 2 June, 2014; Accepted 24 June, 2014

Due to excessive use, synthetic medicines are going to be expensive and resistant, their residues accumulate in blood, that is why slowly and gradually they become resistant. Alternatives to synthetic medicines are herbal medicines that are cheap, with minimum or no side effect. Previous studies found that liquid extract of Aloe vera plant proved beneficial for animal’s health as it has immune-modulating, hypolipidemic, hypoglycemic and haematinic effects. The aim of this study was to investigate the effect of A. vera extract on plasma biochemistry. A total of twenty (n = 20) male rabbits were selected and divided into 4 groups; P, Q, R and S with five (n = 5) in each group. The group P was kept as control while group Q, R and S were given oral A. vera extract at the dose of 200, 300 and 400 mg/kg body weight, respectively daily for 21 days. Blood samples were taken on various days; 0, 7, 14 and 21 of treatment. Research has found that A. vera significantly (p < 0.05) decrease cholesterol level, that is indication that A. vera has influence on adipose tissues.

Key words: Aloe vera, haematology, plasma biochemistry.

INTRODUCTION

Herbal treatment has been used from ancient times for the remedies of many pathological lesions and pathologies. There are many plants with medicinal characteristics; Trigonella foenum graecum, Allium sativum, Gymnema sylvestre, Syzygium cumini and Aloe vera (Saif-ur-rehman et al., 2011). A. vera is a well know plant that has been
grown in houses as first aid in many countries. It has been reported that there are 275 species of *A. vera*, out of that, 100 occur in South Africa (Maphosa and Masika, 2010), 4 occur in India that are *Aloe ferox*, *Aloe inermis*, *Aloe forbesii* and *Aloe barbadensis* and 12 to 15 occur in Arabian Peninsula (Urvashi and Raju, 2012). *A. vera* is a succulent plant that is mainly composed of inner liquid portion and outer greenish covering that has small spines. The former is about 99.5% and the latter is about 0.5% that constitutes the whole solid portion (Hamman, 2008).

Research has investigated that there are about 200 compounds in *A. vera* out of them, 75 are well known for their biological active ingredients. These active ingredients are *aloe polysaccharides* (Jun et al., 2005), cholesterol reducing ingredients (anthraquinones, isoAloeresin-D, iso-rabaichromone, neoAloesin-A) (Ni and Tizard, 2004) tannins, sterols (lupeol, Aloetinic acid, choline and choline salicylate, complex mucopolysaccharides similar to hyaluronic acid, sapogenins), enzymes such as catalase, alliinase, amylase and cellulose. *A. vera* gel contains anthrones and anthraquinones acetylated mannans, anthraquinone C-glycosides, polymannans and lectins (Boudreau and Beland, 2006).

It has been reported that *A. vera* has been used for remedies of many non infectious infestations such as enhance wound healing by proliferation of epithelial and fibrous tissue (Reddy et al., 2011). It has been used to prevent and treat various lesions of gastro intestinal parasites mainly helminths (Maphosa and Masika, 2010). Research has investigated that *A. vera* has influence on central nervous system and enhancing ependymal cells of brain that are source of cerebro spinal fluid in cavities called ventricles (Kosif et al., 2008). *A. vera* promote the function of liver and pancreas by enhancing aspartate amino transferase (AST) and alanine transferase (ALT) that have been investigated as hepato-specific enzymes and cause major damage to hepatocytes (Iji et al., 2010). It has been investigated that *A. vera* has antifungal properties and used to prevent many fungal infestations and fungal diseases. Instead of this, *A. vera* has been used as preventive measure as well as to treat variety of infectious disease. It has been used as an active antibacterial agent against bacteria such as *Klebsella*, *Pseudomonas*, *Streptococcus*, *Staphylococcus*, *Escherichia coli*, *Salmonilla* etc (Hamman, 2008). Research has investigated that the vital medicinal *A. vera* plant has been used for remedies of various viral diseases. It has been used to prevent animal and humans from hepatitis by enhancing function of liver and inhabiting hepatitis virus (Rabe et al., 2005). It has also been used to enhance immune system; mannose polymers are special polysaccharides derived from *A. vera* with immune modulating properties (Leung et al., 2004). It has also been investigated that aloe polysaccharides are used to stimulate hematopoietic stem cell, myeloid and erythroid colony forming cell and macrophage colony forming cell (Im et al., 2005). Considering its hypoglycaemic effect, *A. vera* has been used to cure diabetic as well as normal rats (Saif-ur-Rehman et al., 2011). By considering clinical and therapeutic importance of *A. vera*, the present study is proposed to know safe and effective dose of *A. vera* extract; and also to know its biochemical and haematological properties.

**MATERIALS AND METHODS**

*A. vera* plant was brought from Hyderabad district of Sindh province, fresh leaves of *A. vera* was separated and washed. A leaf was split in two parts by removing spiny margins, white transparent inner gel of *A. vera* was obtained, blended and weighed. Two hundred grams (200 g) of blended *A. vera* liquid was boiled with 400 ml of distilled water for 20 min. After boiling, the extract was cooled, filtered and stored in a refrigerator (4°C) until further use. A total of 20 male rabbits (average body weight: 1·5 kg) were selected and divided into four groups; P, Q, R and S (with 5 rabbits in each group). Feed (rice and green grass) and water was given to them according to free choice of feeding. Group P was kept as control while group Q, R and S were given oral *A. vera* extract at the dose of 200, 300 and 400 mg/kg body weight, respectively daily for 21 days. Blood was collected (on 0, 7, 14, and 21st day) of treatment from central ear vein and cephalic vein, and transferred to test tubes containing anticoagulant (EDTA; Ethylene diamine tetra acetate). The blood samples were brought to Post Graduate Laboratory of Veterinary Physiology, Sindh Agriculture University, Tandojam for analysis and further investigations. Serum protein, globulin, albumin, urea and creatinine were investigated according to Ogunsanmi et al. (1994). The plasma triglyceride and cholesterol were determined according to Toro and Ackermann (1975).

**RESULT**

Total protein, albumin and globulin in various groups; P, Q, R and S on various days (1, 7, 14 and 21) of treatment are significantly different (Table 1 and Figures 1 to 3). It has also been investigated that *A. vera* extract have significant influence (p < 0.05) on urea and creatinine, in various groups (Q, R and S), on various days of treatment, by various doses of 200, 300 and 400 mg/kg (Table 2 and Figures 4 and 6). Furthermore it has been found that *A. vera* extract significantly (p < 0.05) lowers cholesterol level in all groups (Q, R and S) on various days of treatment, respectively (Table 2 and Figure 5). It is found that the dose of 300 mg/kg and the dose of 400 mg/kg brought equal decrease in cholesterol level on 7th day, while on 14th day there was gradual decrease caused
Table 1. Plasma biochemistry of rabbits given oral administration of *A. vera* extract (Mean ± SD).

<table>
<thead>
<tr>
<th>Day</th>
<th>Rabbit group</th>
<th>Total protein</th>
<th>Albumin</th>
<th>Globumin</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1</td>
<td>6.34±0.32ᵃ</td>
<td>3.62±0.04ᵃ</td>
<td>3.38±0.08ᵃ</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>6.42±0.16ᵃ</td>
<td>2.94±0.34ᵇ</td>
<td>3.40±0.15ᵇ</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>6.28±0.19ᵃ</td>
<td>3.84±0.33ᵇ</td>
<td>3.30±0.10ᵇ</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>6.04±0.32ᵃ</td>
<td>2.96±0.27ᵇ</td>
<td>3.36±0.08ᵃ</td>
</tr>
<tr>
<td>7</td>
<td>1</td>
<td>6.44±0.05ᵃ</td>
<td>3.58±0.04ᵃ</td>
<td>3.40±0.10ᵃ</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>6.40±0.10ᵇ</td>
<td>2.94±0.32ᵇ</td>
<td>3.36±0.11ᵇ</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>6.32±0.13ᵃ</td>
<td>2.88±0.27ᵇ</td>
<td>3.30±0.10ᵇ</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>6.16±0.41ᵃ</td>
<td>2.96±0.33ᵇ</td>
<td>3.34±0.08ᵃ</td>
</tr>
<tr>
<td>14</td>
<td>1</td>
<td>6.46±0.16ᵃ</td>
<td>3.44±0.15ᵃ</td>
<td>3.34±0.08ᵃ</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>6.50±0.07ᵃ</td>
<td>2.94±0.36ᵇ</td>
<td>3.28±0.08ᵃ</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>6.48±0.08ᵃ</td>
<td>2.94±0.31ᵇ</td>
<td>3.28±0.13ᵇ</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>6.12±0.50ᵃ</td>
<td>2.86±0.30ᵇ</td>
<td>3.30±0.12ᵇ</td>
</tr>
<tr>
<td>21</td>
<td>1</td>
<td>6.44±0.13ᵃ</td>
<td>3.44±0.19ᵇ</td>
<td>3.34±0.11ᵇ</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>6.30±0.51ᵇ</td>
<td>2.92±0.36ᵇ</td>
<td>3.38±0.13ᵇ</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>6.50±0.18ᵃ</td>
<td>2.96±0.27ᵇ</td>
<td>3.36±0.13ᵇ</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>6.34±0.24ᵇ</td>
<td>2.94±0.36ᵇ</td>
<td>3.32±0.10ᵇ</td>
</tr>
</tbody>
</table>

**Figure 1.** Total protein of various groups on various days.

by the dose of 300 and 400 mg/kg and highest decrease caused by 200 mg/kg. Finally, on 21st day the dose of 300 and 400 mg/kg brought equal changes in cholesterol level (Table 2 and Figure 5).

**DISCUSSION**

Haematology and plasma biochemistry are basic indictors and fundamental markers of various pathologies.
and pathogenic agents. Therefore investigators choose haematology and plasma biochemistry to diagnose disease within short period of time. Previous investigations found that *A. vera* extract improved immune cells and complement system (Amjad et al., 2014; Ghasem et al., 2011; Leung et al., 2004; Im et al., 2005), active antibacterial agent (Hamman., 2008) improves ventricles key component of producing cerebrospinal fluid (Kosif et al., 2008). It is hypothetised that *A. vera* improves plasma proteins for that reason, plasma contents are assessed and it was found that it causes gradual changes in the means of total protein, albumin, globulin, urea, and creatinine in different groups were not significantly different (*p* < 0.05) but it shows regular variations, gradual increase or decrease in all the three doses on 200, 300 and 400 mg/kg on different days of treatment, respectively. Besides

Table 1: Plasma protein contents of various groups on various days.

<table>
<thead>
<tr>
<th>Group</th>
<th>Day 0</th>
<th>Day 7</th>
<th>Day 14</th>
<th>Day 21</th>
</tr>
</thead>
<tbody>
<tr>
<td>P</td>
<td>3.62</td>
<td>3.58</td>
<td>3.44</td>
<td>3.44</td>
</tr>
<tr>
<td>Q</td>
<td>2.94</td>
<td>2.94</td>
<td>2.94</td>
<td>2.92</td>
</tr>
<tr>
<td>R</td>
<td>2.84</td>
<td>2.88</td>
<td>2.94</td>
<td>2.96</td>
</tr>
<tr>
<td>S</td>
<td>2.96</td>
<td>2.96</td>
<td>2.86</td>
<td>2.94</td>
</tr>
</tbody>
</table>

Figure 2. Albumen of various groups on various days.

Figure 3. Globulin of various groups on various days.
Table 2. Plasma lipid and metabolites (mg/dl) of rabbits given oral administration of Aloe vera extract (Mean ± SD).

<table>
<thead>
<tr>
<th>Day</th>
<th>Rabbit group</th>
<th>Cholesterol</th>
<th>Urea</th>
<th>Creatinine</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>1</td>
<td>44.96±0.18a</td>
<td>22.04±0.72b</td>
<td>1.02±0.10ab</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>43.06±2.88abc</td>
<td>23.68±1.94ab</td>
<td>1.14±0.23abc</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>41.68±3.19abc</td>
<td>23.78±2.20ab</td>
<td>1.22±0.19ab</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>39.58±4.51abc</td>
<td>23.58±1.78ab</td>
<td>1.24±0.11a</td>
</tr>
<tr>
<td>7</td>
<td>1</td>
<td>44.42±0.40abc</td>
<td>22.48±0.77ab</td>
<td>1.00±0.07ab</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>42.58±2.34abc</td>
<td>23.38±1.12ab</td>
<td>1.10±0.15abc</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>38.34±8.43abc</td>
<td>23.68±1.94ab</td>
<td>1.22±0.14ab</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>38.40±3.91abc</td>
<td>24.90±2.16ab</td>
<td>1.16±0.08ab</td>
</tr>
<tr>
<td>14</td>
<td>1</td>
<td>44.86±0.20a</td>
<td>23.2±1.30ab</td>
<td>0.96±0.05b</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>41.44±2.66abc</td>
<td>23.98±1.34ab</td>
<td>1.06±0.18abc</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>39.30±3.28abc</td>
<td>24.62±1.78ab</td>
<td>1.20±0.21ab</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>38.44±4.38abc</td>
<td>24.24±1.32ab</td>
<td>0.98±0.13ab</td>
</tr>
<tr>
<td>21</td>
<td>1</td>
<td>44.26±0.73abc</td>
<td>24.16±1.09ab</td>
<td>1.00±0.12ab</td>
</tr>
<tr>
<td></td>
<td>2</td>
<td>41.54±2.41abc</td>
<td>24.78±1.44ab</td>
<td>1.12±0.13ab</td>
</tr>
<tr>
<td></td>
<td>3</td>
<td>37.96±2.98c</td>
<td>25.36±3.21ab</td>
<td>1.12±0.17ab</td>
</tr>
<tr>
<td></td>
<td>4</td>
<td>38.58±4.06bc</td>
<td>25.84±1.77a</td>
<td>1.10±0.07ab</td>
</tr>
</tbody>
</table>

Figure 4. Urea of various groups on various days.

Besides this, it was found that urea is increased in all three doses on all three treated groups and highest increase was noted in group S that was given 400 mg/kg of A. vera extract (Tables 1 and 2 and Figures 1, 2, 3, 4 and 6), these investigation are contrary to Iji et al. (2010). Regular administration of A. vera extract significantly decreased (p < 0.05) cholesterol level in various groups (P, Q, R and S), respectively. Highly significant decrease occur in groups R and S on day 7, 14 and 21 that was 38.40 ± 3.91, 39.30 ± 3.28, 37.96 ± 2.98 and 38.40 ± 3.91, 38.44 ± 4.38 and 38.58 ± 4.03, respectively (Table 2 and Figure 5). Research has proved that decrease in cholesterol is due to lower production of endogenous cholesterol transporter. It may be due to mannans that
inhibit cholesterol absorption (Sikarwar et al., 2010) or active involvement of liver tissues in fatty acid oxidation and formation of lipoproteins (Rajasekaran et al., 2006).

Conclusion

The study therefore concluded that chronic oral administration of A. vera extract has decreasing effects on cholesterol level and improves haematology. We believe further investigations with similar results will be helpful to know the mechanism of these modifications in the level of cholesterol and haematological parameters.

Conflict of Interests

The author(s) have not declared any conflict of interests.

REFERENCES

Amjad AC, Izhar HQ, Saeed AS, Alta HS, Jameel AG, Roshan AK,


Anatomical variants, clinical presentation and pathological findings in patients suffering from chronic rhinosinusitis underwent functional endoscopic sinus surgery

Seyyed Abdollah Madani, Seyyed Abbas Hashemi, Shahzad Javan and Akram Alsadat Hoseini

Department of Otorhinolaryngology, Head and Neck Surgery, Traditional and Complementary Medicine Research Center, Mazandaran University of Medical Sciences, Sari, Iran.

Received 3 May, 2014; Accepted 23 June, 2014

In this study, we investigated the different anatomical variations and clinical modes of presentation of chronic rhinosinusitis and their association with final histopathological diagnosis. This prospective randomization research was conducted on a total of 284 patients with chronic rhinosinusitis who underwent functional endoscopic sinus surgery from March, 2009 to September, 2012. The study population (284 patients) included 170 males (59.8%) and 114 females (40.1%), with a mean age of 29 years. The most frequent symptoms were nasal obstruction (24.3%); headache (21.9%), nasal congestion (18.5%) and post nasal discharge (16.6%). The nasal septums were significantly deviated in 207 (79.2%) subjects. Inferior turbinate hypertrophy was observed in 102 (35.9%) patients. Bulla ethmoidalis was reported in 32 (11.3%) participants. Uncinate bulla and concha bullosa were identified in 12 (4.2%), 12 (4.2%) patients, respectively. According to pathological report, majority of the patients (184 patients, 64.7%) had chronic inflammation in sinuses went after polyp in 46 patients (16.9%). Our study revealed anatomical variations were common in patients with chronic rhinosinusitis. Identification of different variations will guide the surgeons during functional endoscopic sinus surgery.

Key words: Chronic rhinosinusitis, functional endoscopic sinus surgery, anatomical variations.

INTRODUCTION

Based on the National Health Interview Survey of 1996, chronic rhinosinusitis (CRS) was the second chronic disease in USA imposing 12.5% of the US population or nearly 31 million subjects annually (Adams et al., 1999; Anand 2004). In this regard, according to 2008 National Health Interview Survey information, rhinosinusitis imposed 1 in 7 adults (Pleis et al., 2009). Since CRS was established on symptomatic criteria, this prevalence was
probably overestimated in these studies. Due to coexisting inflammation of the nasal and sinus mucosa, the present terminology is “rhinosinusitis”; if the clinical symptoms of this inflammation exist for at least 12 weeks with no complete resolution, we call it chronic (Koen et al., 2011; Hashemi et al., 2012).

The introduction of functional endoscopic sinus surgery (FESS) besides the medical therapy for CRS, made the interventional procedures competent (Kennedy 1985; Stammberger 1985). Short- and long-term investigations worked on FESS results, elucidated development in sinus symptoms and reduced recurrent infections (Kennedy 1992; Senior and Kennedy 1998). Endoscopic sinus surgery has been established as a safe method and complications prevalence is indicated to be less than 1% (Chiu and Kennedy 2004). Numbers of complications include blindness, intracranial injury, orbital hematoma, stroke and cerebrospinal fluid leak (Luong and Bradley, 2006). In this relation, there are pathologic situations which need a more aggressive FESS. For instance, extensive nasal polyposis affecting middle turbinate required to be removed partially since post surgery reduced prevalence of synechiae, long-term patency of middle meatus antrostomy, developed nasal airflow, reduced nasal resistance and developed intrasurgery and postsurgery access to the ethmoidal labyrinth (LaMear 1992; Lawson 1994; Cook et al., 1995; Stewart 1998; Giacchi et al., 2000).

In this study, we attempted to explore the different anatomical variations and clinical modes of presentation of CRS and their relation with final histopathological diagnosis and to clarify these conditions from other situations in patients who underwent FESS.

METHODOLOGY

This prospective study population involved 284 subjects including 170 male and 114 female subjects, aged 5 to 70 years, who underwent FESS for CRS from March, 2009 to September, 2012. The ethics committee of Mazandaran University of Medical sciences (Sari IRAN) approved this study. Written informed consent was obtained from all participants prior to initiation of investigation.

Inclusion and exclusion criteria

All patients were selected according to criteria for CRS as described by (Benninger et al., 2003). Our exclusion criteria involved age younger than 5 years, history of coronary artery disease and bleeding disorders. Subsequent checkups were performed before surgery and in each visit, patients were questioned regarding nasal obstruction, headache, nasal congestion, post nasal discharge, breathing disorders, cough, facial pain, hoarseness, epistaxis and anatomical variation were examined during FESS.

Randomization

852 patients considered for the research were randomized before the study. Assignment to groups was carried out by computer-generated random numbers. The randomization process was done by a third party; all of the patients and doctors were excluded in selection section.

Statistical analysis

Descriptive analysis was performed to characterize the outcomes including demographic, anatomical variants, histological reports, clinical symptoms and any other information before and during checkups and FESS. Data were transferred to MS-excel spread sheets. The procedures involved were transcription, preliminary data inspection, content analysis and at last interpretation. Investigators used percentages (SPSS software, Version 15, Chicago, IL, USA) to interpret epidemiological variables.

RESULTS

The study group (284 patients) included 170 males (59.8%) and 114 females (40.1%), with a mean age of 29 years. The most frequent symptoms among these patients were nasal obstruction (24.3%), headache (21.9%), nasal congestion (18.5%) and post nasal discharge (16.6%) (Table 5). The nasal septums were significantly deviated in 207 (79.2%) subjects (Table 4). Inferior turbinate hypertrophy was observed in 102 (35.9%) patients (Table 4). Bulla ethmoidalis was reported in 32 (11.3%) participants (Table 4). Uncinate bulla and concha bullosa were identified in 12 (4.2%) and 12 (4.2%) patients, respectively (Table 4). Most of the patients were in the range of 10 to 20 years (83 patients, 29.4%) followed by 20 to 30 (73 patients, 25.7%) and 30 to 40 years (70 subjects, 24.6%) (Table 1). According to pathological report, majority of the patients (184 patients, 64.7%) had chronic inflammation in sinuses went after polyp in 46 patients (16.9%) (Table 3). Most of the patients (142 patients, 50%) had history of symptoms for 1 to 5 years. 63 patients (22.2%) indicated these symptoms for 5 to 10 years (Table 2).

DISCUSSION

This manuscript is divided into two broad sections. In the first part we discussed the anatomical variants in CRS and in the second section we talked about the clincopathological feature of study population. Advances in operational procedures resulted in better findings with less complications in the paranasal sinus area. Therefore in this trial, we examined the correlation of anatomical variations and presence of CRS. Functional endoscopic sinus surgery (FESS) is used for CRS refractory to medical therapy. The indications for FESS are expanding and discussion about these indications is beyond the scope of this manuscript but in this relation, some absolute indications for FESS in children are summarized (Fokkens et al., 2007):

1. Complete nasal obstruction in CF due to massive
The presence of an air cavity inside the lamina recurvata is called concha bullosa. This space is ranging from too small to considerable in size (Meloni et al., 1992). Different studies reported various frequencies of the concha bullosa, including 17, 21 and 28% (Meloni et al., 1992; Zinreich et al., 1987; Is˚yk and Bulut 1994). It has been shown that these variants may be the cause of middle meatal obstruction and recurrent ethmoiditis (Shechtamn et al., 1993). Some studies reported a correlation between the concha bullosa and sinusitis (Shin 1986; Calhoun et al., 1991), but some investigations indicated there was no significant association (Danese et al., 1997; Lam et al., 1996). In this relation, (Calhoun et al. 1991), showed there was a probable relation between concha bullosa or septal deviation and rhinosinusitis (Calhoun et al., 1991). In consistent with previous investigations, Hamdan et al., 2011 indicated there was no significant association between septal deviation and rhinosinusitis. (Hamdan et al., 2011; Jamie et al. 2004) elucidated seventy-three percent of their study participants with concha bullosa who had paranasal sinus inflammatory diseases; but 78% of patients without concha bullosa also suffered from some forms of inflammatory diseases. (Hisham et al., 2011) reviewed that the computed tomography scans of 63 subjects underwent revision FESS. They showed 15.9% of the series had significant deviation of the nasal septum. In our study, 12 patients (4.2%) were identified with concha bullosa and nasal septal deviations were highlighted in 207 (79.2%) of the subjects. Our exploration confirmed that nasal septal deviations are a significant interest in CRS. The previous studies did not discuss about the inferior turbinate hypertrophy, in contrast to former findings, in this series inferior turbinate hypertrophy was elucidated in 102 (35.9%) patients. Pneumatization of the uncinate process is named uncinate bulla, which may lead to anatomic narrowing of the infundibulum and could damage sinus ventilation (Bolger et al., 1990; Bolger et al., 1991, Rao and El-Noueam 1998). Although this variant is not well described but (Kennedy and Zinreich, 1988) reported one subject with uncinate bulla in a series of 230 participants. Bolger et al. (1991) studied

### Table 1. Age distribution of patients in this series.

<table>
<thead>
<tr>
<th>Age</th>
<th>Number</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>5-10</td>
<td>18</td>
<td>6.3</td>
</tr>
<tr>
<td>10-20</td>
<td>83</td>
<td>29.4</td>
</tr>
<tr>
<td>20-30</td>
<td>73</td>
<td>25.7</td>
</tr>
<tr>
<td>30-40</td>
<td>70</td>
<td>24.6</td>
</tr>
<tr>
<td>40-50</td>
<td>24</td>
<td>8.4</td>
</tr>
<tr>
<td>50-60</td>
<td>6</td>
<td>2.1</td>
</tr>
<tr>
<td>&gt;60</td>
<td>10</td>
<td>3.5</td>
</tr>
</tbody>
</table>

### Table 2. Individual differences in duration of chronic rhinosinusitis before functional endoscopic sinus surgery.

<table>
<thead>
<tr>
<th>Duration (year)</th>
<th>Number</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;1</td>
<td>50</td>
<td>17.6</td>
</tr>
<tr>
<td>1-5</td>
<td>142</td>
<td>50</td>
</tr>
<tr>
<td>5-10</td>
<td>63</td>
<td>22.2</td>
</tr>
<tr>
<td>&gt;10</td>
<td>29</td>
<td>10.2</td>
</tr>
</tbody>
</table>

### Table 3. Classification of pathological reports.

<table>
<thead>
<tr>
<th>Pathology</th>
<th>Number</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chronic inflammation</td>
<td>184</td>
<td>64.7</td>
</tr>
<tr>
<td>Polyp</td>
<td>46</td>
<td>16.9</td>
</tr>
<tr>
<td>Allergic rhinosinusitis</td>
<td>27</td>
<td>9.5</td>
</tr>
<tr>
<td>Squamous cell carcinoma</td>
<td>8</td>
<td>2.81</td>
</tr>
<tr>
<td>Hemangioma</td>
<td>8</td>
<td>2.81</td>
</tr>
<tr>
<td>Craniophyrgioma</td>
<td>6</td>
<td>2.1</td>
</tr>
<tr>
<td>Rhinolith</td>
<td>5</td>
<td>1.7</td>
</tr>
</tbody>
</table>

### Table 4. Distribution of anatomical variants in patients with chronic rhinosinusitis.

<table>
<thead>
<tr>
<th>Anatomical variations</th>
<th>Number</th>
<th>Percent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nasal septal deviation</td>
<td>207</td>
<td>79.2</td>
</tr>
<tr>
<td>Inferior turbinate hypertrophy</td>
<td>102</td>
<td>35.9</td>
</tr>
<tr>
<td>Bulla ethmoidalis</td>
<td>32</td>
<td>11.3</td>
</tr>
<tr>
<td>Concha bullosa</td>
<td>12</td>
<td>4.2</td>
</tr>
<tr>
<td>Uncinate bulla</td>
<td>12</td>
<td>4.2</td>
</tr>
</tbody>
</table>

The presence of an air cavity inside the lamina recurvata is called concha bullosa. This space is ranging from too small to considerable in size (Meloni et al., 1992). Different studies reported various frequencies of the concha bullosa, including 17, 21 and 28% (Meloni et al., 1992; Zinreich et al., 1987; Is˚yk and Bulut 1994). It has been shown that these variants may be the cause of middle meatal obstruction and recurrent ethmoiditis (Shechtamn et al., 1993). Some studies reported a correlation between the concha bullosa and sinusitis (Shin 1986; Calhoun et al., 1991), but some investigations indicated there was no significant association (Danese et al., 1997; Lam et al., 1996). In this relation, (Calhoun et al. 1991), showed there was a probable relation between concha bullosa or septal deviation and rhinosinusitis (Calhoun et al., 1991). In consistent with previous investigations, Hamdan et al., 2011 indicated there was no significant association between septal deviation and rhinosinusitis. (Hamdan et al., 2011; Jamie et al. 2004) elucidated seventy-three percent of their study participants with concha bullosa who had paranasal sinus inflammatory diseases; but 78% of patients without concha bullosa also suffered from some forms of inflammatory diseases. (Hisham et al., 2011) reviewed that the computed tomography scans of 63 subjects underwent revision FESS. They showed 15.9% of the series had significant deviation of the nasal septum. In our study, 12 patients (4.2%) were identified with concha bullosa and nasal septal deviations were highlighted in 207 (79.2%) of the subjects. Our exploration confirmed that nasal septal deviations are a significant interest in CRS. The previous studies did not discuss about the inferior turbinate hypertrophy, in contrast to former findings, in this series inferior turbinate hypertrophy was elucidated in 102 (35.9%) patients. Pneumatization of the uncinate process is named uncinate bulla, which may lead to anatomic narrowing of the infundibulum and could damage sinus ventilation (Bolger et al., 1990; Bolger et al., 1991, Rao and El-Noueam 1998). Although this variant is not well described but (Kennedy and Zinreich, 1988) reported one subject with uncinate bulla in a series of 230 participants. Bolger et al. (1991) studied

Polyposis or due to medialization of the lateral nasal wall;
2. Orbital abscess;
3. Intracranial complications;
4. Antrochoanal polyp;
5. Mucocoel or mucopyocele;
6. Fungal rhinosinusitis.

Possible indications consist of CRS with frequent exacerbations continuing despite optimal medical therapy and after exclusion of any systemic disease (Daniel, 2011).
the CT scans of 202 patients and indicated the uncinate bulla in 2.5% of study population. Likewise in these researches, uncinate bulla were reported in 12 (4.2%) of our study population.

In 2003, a consensus panel described CRS as an inflammatory disease of the nose and paranasal sinuses of not identified etiology defined on the basis of characteristic symptoms (≥ 2 as follows: nasal congestion, facial pain/pressure, anterior or posterior nasal drainage and decreased or absent sense of smell), (duration more than 12 weeks), and objective evidence of sinus disorder by means of direct visualization or imaging examination (Benninger et al., 2003). Among these 284 patients, the most frequent symptoms were nasal obstruction (24.3%), headache (21.9%), nasal congestion (18.5%) and post nasal discharge (16.6%) (Table 5). Based on pathological report, most of the patients (184 patients, 64.7%) were identified with chronic inflammation in sinuses followed by polypl in 46 patients (16.9%) (Table 3). Allergic rhinosinusitis were reported in 27(9.5%) patients according to measurement of serum immunoglobulin (IgE) (Table 3). Although based on symptoms, all of these subjects were identified with CRS but after FESS some patients showed other diseases like (based on histological report) squamous cell carcinoma, hemangioma, craniohynghytoma, rhinolith (Table 3).

Conclusion

The frequencies of anatomical variations, clinical symptoms and pathological features have been reported in various ethnics and each study indicated these numbers, and frequencies are not the same which may be the result of different genetic and environmental factors.

ACKNOWLEDGMENT

This study was supported by a grant from Mazandaran University of Medical Sciences, Sari, Iran. The authors are grateful to Mohammad Hashemi for technical assistance.

Conflict of Interests

The author(s) have not declared any conflict of interests.

REFERENCES


International Journal of Medicine and Medical Sciences

Related Journals Published by Academic Journals

- Journal of Medicinal Plant Research
- African Journal of Pharmacy and Pharmacology
- Journal of Dentistry and Oral Hygiene
- International Journal of Nursing and Midwifery
- Journal of Parasitology and Vector Biology
- Journal of Pharmacognosy and Phytotherapy
- Journal of Toxicology and Environmental Health Sciences