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Mussel RL, De Sa Silva E, Costa AM, Mandarim-De-Lacerda CA (2003). Mast cells in tissue response to dentistry materials: an adhesive resin, a calcium hydroxide and a glass ionomer cement. J. Cell. Mol. Med. 7:171-178.

Booth M, Bundy DA, Albonico P, Chwaya M, Alawi K (1998). Associations among multiple geohelminth infections in school children from Pemba Island. Parasitol. 116: 85-93.0.

Fransiscus RG, Long JC (1991). Variation in human nasal height and breath, Am. J. Phys. Anthropol. 85(4):419-427.

Stanislawski L, Lefeuvre M, Bourd K, Soheili-Majd E, Goldberg M, Perianin A (2003). TEGDMA-induced toxicity in human fibroblasts is associated with early and drastic glutathione depletion with subsequent production of oxygen reactive species. J. Biomed. Res. 66:476-82.

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Full Length Research Paper

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

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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 h, 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

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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 oxidating 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 O_3 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 Eliason, 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 Haţieganu" 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 to2950 g housed in standard individual cages, with an average temperature of 22°C.

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 intraperitoneral 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 O_2 flow=50 l/h, at a 0.2 atmospheric pressure of oxygen, bubbling time of O_3 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.

Score	0	1	2	3
Edema	Absent	In the interlobular septum	Mild or interacinar	Diffuse
Hemorrhage	Absent	In the interlobular septum	Mild or interglandular	Diffuse
Necrosis	Absent	In 1-2 lobules	In 3-4 lobules	More than 5 lobules
Leukocytes infiltration	Absent	In 1-2 lobules	In 3-4 lobules	More than 5 lobules
Citosteatonecrosis	Absent	Mild	Moderate	Severe
Fibrosis	Absent	In 1-2 lobules	In 3-4 lobules	More than 5 lobules

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 were 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 were mannitol 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, analised on the 6th day from the induction of acute pancreatitis, were significantly lower in groups A (0.5083±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

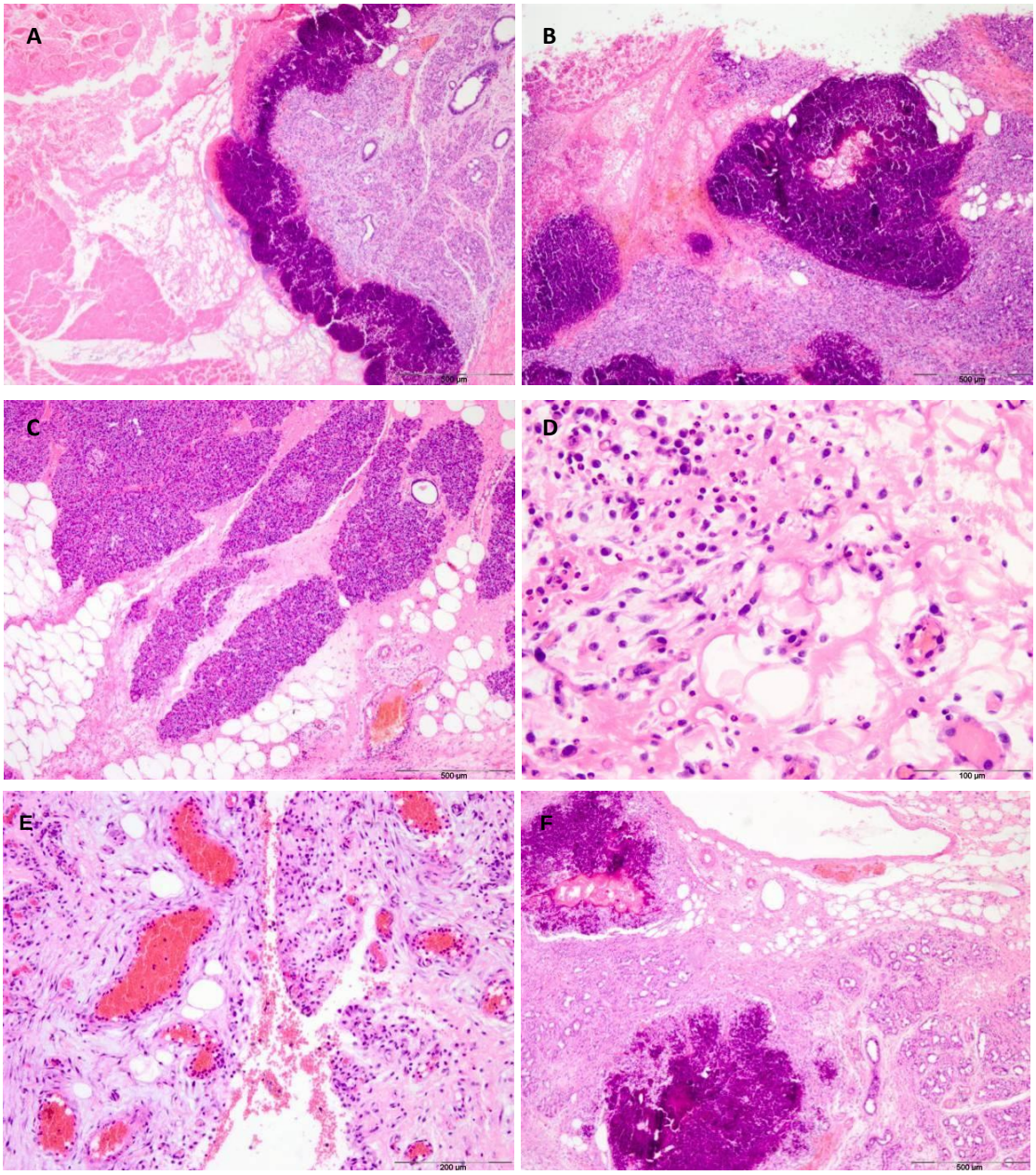

Figure 1. Histopathological aspects of pancreatic and peripancreatic tissues belonging to the rabbits in the control group I, without treatment: (A) diffuse parenchymal necrosis, severe (*), demarcated from normal tissue by a band of neutrophilic and macrophagic leukocyte, HE Bar = 500, (B) necrotico-purulent 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).

the groups. 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

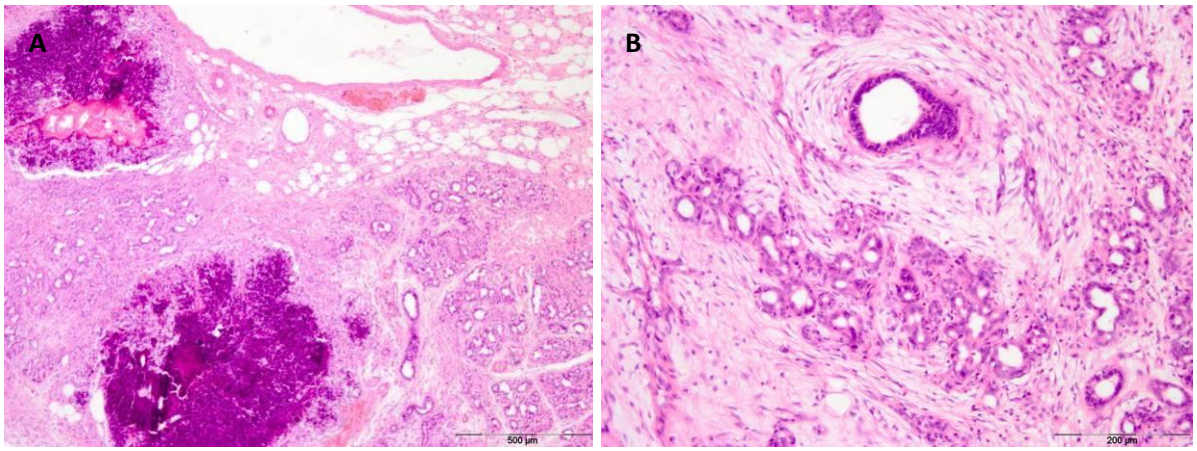

Figure 2. Histopathological aspects of the pancreatic tissues of the rabbits belonging to the study group treated with ozonated saline: (A) focal parenchymal necrosis with the formation of microabceses, edema, hemorrhage, mild neutrophilic inflammatory infiltrate and fibroblast proliferation in adjacent 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.

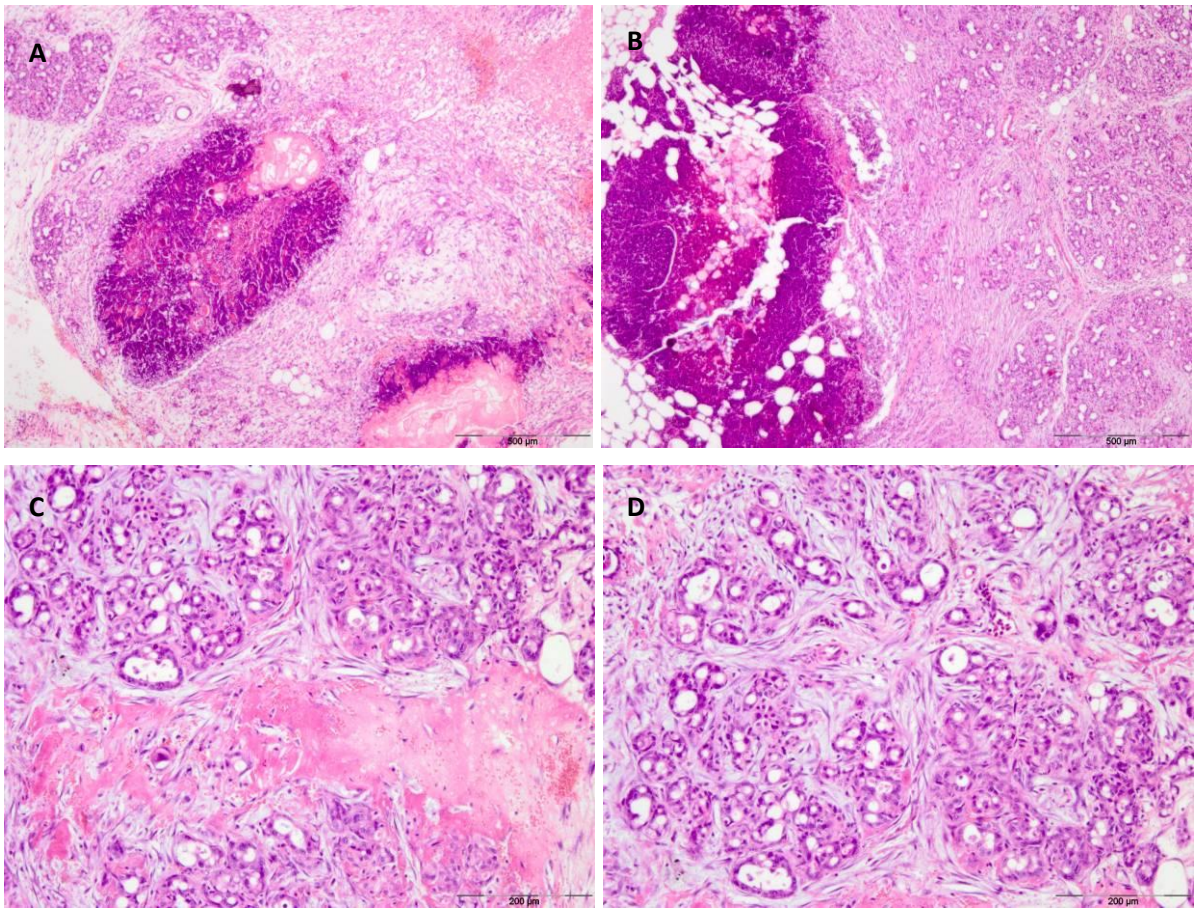

Figure 3. Histopathological aspects of pancreatic tissues of the rabbits belonging to the study group B treated with ozonated saline processed in intense electric fields: (A and B) focal parenchymal necrosis, edema, hemorrhage, moderate neutrophilic inflammatory infiltrate and fibroblast proliferation 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 fasciitis, 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.

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Conflict of Interests

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

REFERENCES

Bhatia M (2002). Novel therapeutic targets for acute pancreatitis and associated multiple organ dysfunction syndrome. Curr. Drug Targets Inflamm. Allergy 1:343–351.

Bialoszewski D, Bocian E, Bukowski B, Czajkowski M, Sokol-Leszczynska B, Tyska S (2010). Antimicrobial activity of ozonated water. Med. Sci. Monitor. 16(9):MT71-5.

Bialoszewski D, Kowalewski M (2003). Superficially, Longer, Intermittent Ozone Theraphy in the Treatment of the Chronic Infected Wounds. Ortop. Traumatol. Rehabilitated. 30(5):652-8.

Bocci V (1996). Ozone as bioregulator: Pharmacology and Toxicology of ozonetherapy today. J. Biol. Rules. Homeost. Agents 10:31-53.

Bocci V (2007). The case for oxygen-ozonetherapy. Br. J. Biomed. Sci. 64(1):449.

Bocci V, Borrell E, Travagli V, Zanardi I (2009). The ozone paradox: Ozone is a strong oxidant as well as a medical drug. Med. Res. Rev. 29(4):646-82.

Bourgaux JF, Defez C, Muller L, Vivancos J, Prudhomme M, Navarro F, Pouderoux P, Sotto A (2007). Infectious complications, prognostic factors and assessment of antiinfectious management of 212 consecutive patients with acute pancreatitis. Gastroenterol. Clin. Biol. 31:431-5.

Cardoso FS, Ricardo LB, Oliveira AM, Canena JM, Horta DV, Papoila AL, Deus JR (2013). C-reactive protein prognostic accuracy in acute pancreatitis: timing of measurement and cutoff points. Eur. J. Gastroenterol. Hepatol. 25(7):784-9. doi: 10.1097.

De Waele JJ, Vogelaers D, Host E, Blot S, Colardyn F (2004). Emergence of antibiotic resistance in infected pancreatic necrosis. Arch. Surg. 139:1371-6.

D'Egido SM (1991). Surgical treatment strategies in the infection of pancreatic necrosis. Br. J. Surg. 78:133.

Dellinger EP, Tellado JM, Soto NE, Ashley SW, Barie PS, Dugernier T, Imrie CW, Johnson CD, Knaebel HP, Laterre PF, Maravi-Poma E, Kissler JJ, Sanchez-Garcia M, Utzolino S (2007). Early antibiotic treatment for severe acute necrotizing pancreatitis. A randomized, double-blind, placebo-controlled study. Ann. Surg. 245:674-83.

Dyas A, Boughton BJ, Das BC (1983). Ozone Killing Action Against Bacterial and Fungal Species: Microbiological Testing of a Domestic Ozone Generator. J. Clin. Pathol. 36:1102-4.

Georgescu I, Nemes R, Cartridges D, Cârţu D, Surlin V, Mărgăritescu

D, Dumitrescu D, Chiuţu L, Ciurea M, Georgescu E (2005). Severe acute pancreatitis-diagnose strategy and therapeutics. Surgery 100:557-62.
Guanche D, Zamora Z, Hernández F, Mena K, Alonso Y, Roda M, Gonzáles M, Gonzales R (2010). Effect of ozone/oxygen mixture on systemic oxidative stress and organic damage. Toxicol. Mech. Methods 20(1):25-30.
Kogelschatz U, Eliason B (1995). Ozone Generation and Application. In: Marcel Dekker. Handbook of Electrostatic Processes. New York 581-605.
Kopchak VM, Khomiak IV, Duvalko AV, Stasenko AA, Dieiev VA (2008). [Application of ozone therapy in complex treatment of patients with acute necrotizing pancreatitis]. Klin Khir pp. 28–31.
Kudari A, Wig DJ, Vaiphei K, Kochhar R, Majumdar S, Gupta R, Yadav TD, Doley RP (2007). Histopathological Sequential Changes in Sodium-Induced Acute Pancreatitis Taurocholat. Pancreas J. 8(5):564-72.
Lilja H, Leppaniemi A, Kemppainen E (2008). Utilization of Intensive Care Unit Resources in Severe Acute Pancreatitis. J. Pancreas 9(2):179-84.
Lipatov KV, Sopromadze MA, Shekhter AB, Rudenko TG, Emelianov IA (2002). Ozone-ultrasonic Therapy in the Treatment of Wounds Purulent. Khirurgiia 1:36-9.
Madej P, Antoszewski Z, Madej JA (1995). Ozonotherapy. Materia Medica Poland. Polish J. Med. Pharm. 27:53-6.
Marincaş M, Brătucu E, Drum Madalina, Cirimbei C (2006). Peacocks Ligia. Surgical attitude in acute pancreatitis. Surgery 101:237-47.
Oizumi M, Suzuki T, Uchida M, Furuya J, Okamoto Y (1998). *In vitro* testing of a denture cleaning method using ozone. J. Med. Dent. Sci. 45:135-139.
Parkhisenko IA, Glukhov AA (2001). Use of ozone therapy and hydro-pressure technologies in complex intensive therapy of surgical sepsis. Khirurgiia 4:55-8.
Schmid SW, Uhl W, Friess H, Malfertheine P (1999). The role of infection in acute pancreatitis. Gut 45:311-6.
Silva RA, Garotti, JE, Silva RSB, Navarini A, Pacheco AM (2009). Analysis of the bactericidal effect of ozone pneumoperitoneum. Acta Cir Bras 24:124-7.
Uysal B, Yasar M, Ersoz N, Coskun O, Kilic A, Cayc T, Kurt B, Oter S, Korkmaz A, Guven A (2010). Efficacy of hyperbaric oxygen therapy and medical ozone therapy in experimental acute necrotizing pancreatitis. Pancreas 39:9–15.
Yilmaz M, Topsakal S, Herek O, Ozmen O, Sahinduran S, Buyukoglu T, Yonetci N (2009). Effects of etanercept on sodium taurocholate-induced acute pancreatitis in rats. Translational Res.154(5):241-8.

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Full Length Research Paper

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

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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, Gymneema slyvestre, Syzigium cumini* and *Aloe vera* (Saif-ur-rehman et al., 2011). *A. vera* is a well know plant that has been

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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 ingedients (anthraquinones, isoAloeresin-D, iso-rabaichromone, neoAloesin-A) (Ni and Tizard, 2004) tannins, sterols (lupeol, Aloetinic acid, choline and choline salicylate, complex mucopoly-saccharides similar to hyaluronic acid, sapogenins), enzymes such as catalase, alliinase, amylase and cellulose. *A. vera* gel contains anthrones and anthraxquinones 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 nervious system and enhancing ependymal cells of brain that are source of cerebro spinal fluid in cavaties 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 antibac-terial 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 (lm et al., 2005). Considering its hypogly-caemic 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 splitted 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).

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

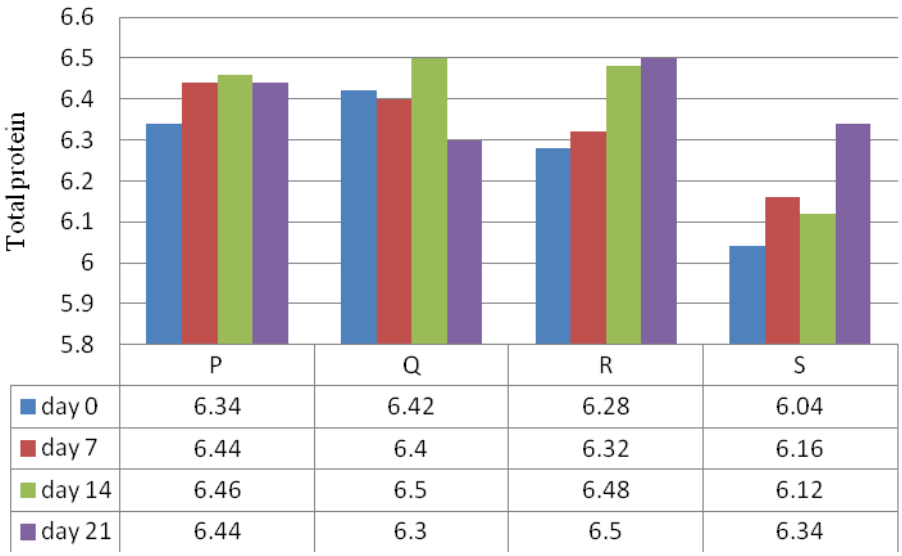
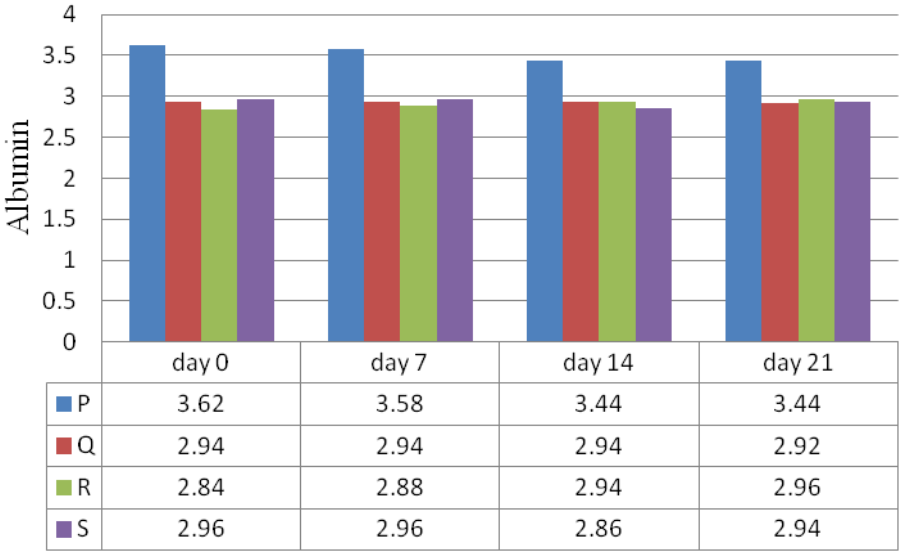


Figure 1. Total protein of various groups on various days.

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).

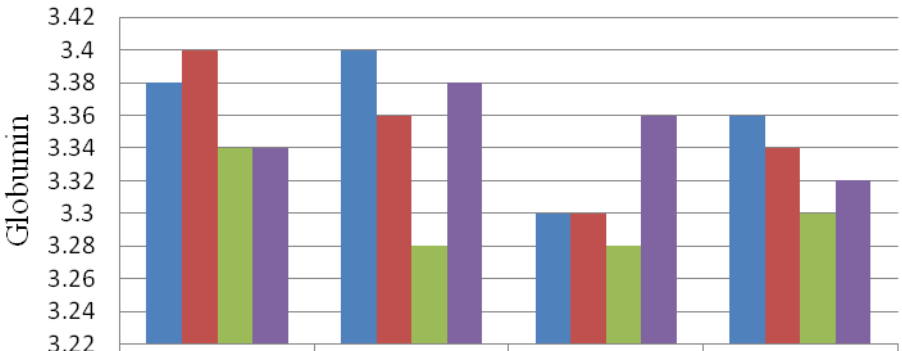
DISCUSSION

Haematology and plasma biochemistry are basic indictors and fundamental markers of various pathologies



	day 0	day 7	day 14	day 21
P	3.62	3.58	3.44	3.44
Q	2.94	2.94	2.94	2.92
R	2.84	2.88	2.94	2.96
S	2.96	2.96	2.86	2.94

Figure 2. Albumen of various groups on various days.



	P	Q	R	S
day 0	3.38	3.4	3.3	3.36
day 7	3.4	3.36	3.3	3.34
day 14	3.34	3.28	3.28	3.3
day 21	3.34	3.38	3.36	3.32

Figure 3. Globulin of various groups on various days.

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; lm et al., 2005), active antibacteral 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 2. Plasma lipid and metabolites (mg/dl) of rabbits given oral administration of *Aloe vera* extract (Mean ± SD).

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

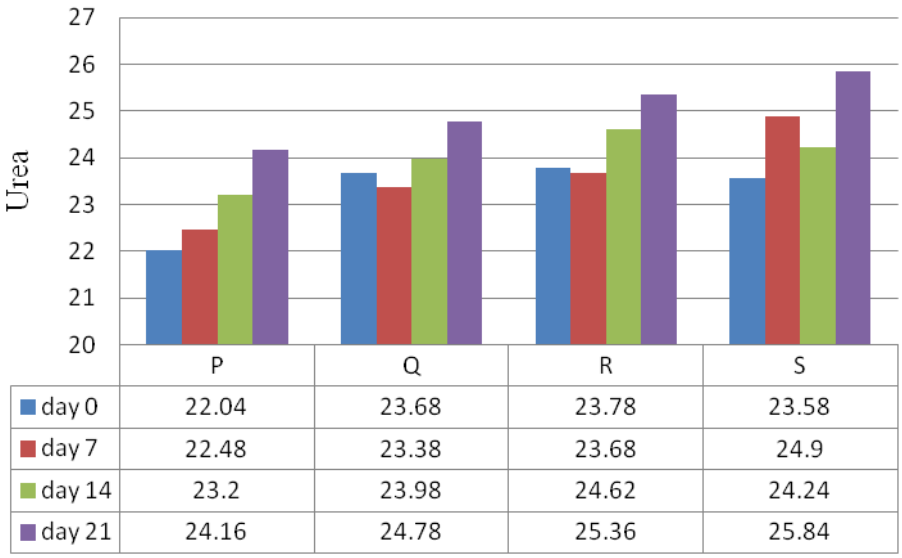
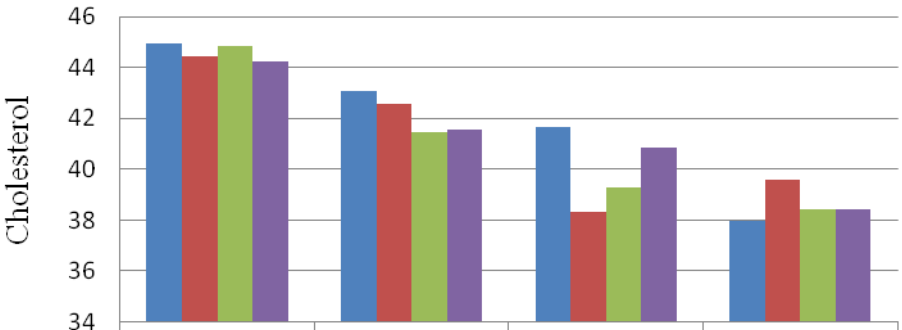


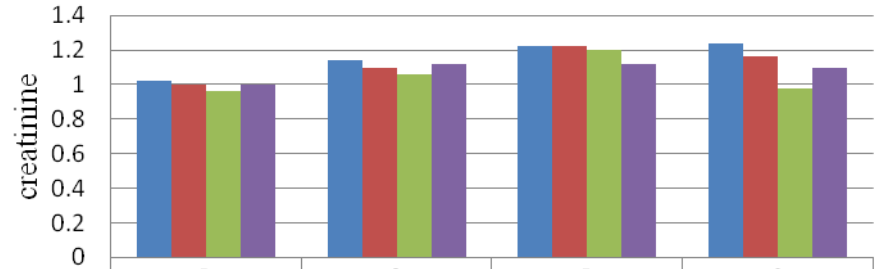
Figure 4. Urea of various groups on various days.

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



	P	Q	R	S
day 0	44.96	43.06	41.68	37.96
day 7	44.42	42.58	38.34	39.58
day 14	44.86	41.44	39.3	38.4
day 21	44.26	41.54	40.84	38.44

Figure 5. Cholesterol of various groups on various days.



	P	Q	R	S
day 0	1.02	1.14	1.22	1.24
day 7	1	1.1	1.22	1.16
day 14	0.96	1.06	1.2	0.98
day 21	1	1.12	1.12	1.1

Figure 6. Creatinine of various groups on various days.

inhabit cholesterol absoption (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, Atta HS, Jameel AG, Roshan AK,

Imtiaz AS, Nawab AK, Band-e-ali K (2014). Effect of oral supplements of aloe vera extract on hematology and immune cells in rabbits. Afr. J. Pharm. Pharm. 8(19):497-501.
Boudreau MD, Beland FA (2006). An evaluation of the biological and toxicological properties of *Aloe barbadensis* (Miller), *Aloe vera*. J. Environ. Sci. HealthC. 24:103-154.
Ghasem V, Mehdi T, Amin KM, Reza H (2011). The effect of Aloe vera extract on humoral and cellular immune response in rabbit. Afr. J. Biotech. 10(26):5225-5228.
Hamman JH (2008). Composition and applications of *Aloe vera* leaf gel. Molecules 13:1599-1616.
Iji OT, Oyagbemi AA, Azeez OI (2010). Assessment of Chronic Administration of *Aloe Vera* Gel On Haematology, Plasma Biochemistry, Lipid Profiles and Erythrocyte Osmotic Resistance in Wistar Rats. Niger. J. Physiol. Sci. 25:107– 113.
Im SA, Oh ST, Song S, Kim MR, Kim DS, Woo SS, Jo YI, Lee CK (2005). Identification of optimal molecular size of modified Aloe polysaccharides with maximum immunomodulatory activity. Int. Immunopharmacol. 5:271-279.
Leung MYK, Liu C, Zhu LF, Hui YZ, Yu B, Fung KP (2004). Chemical and biological characterization of a polysaccharide biological response modifier from *Aloe vera* L. var. *chinensis* (Haw.) Berg. Glycobiol. 14:501-510.
Ni Y, Turner D, Yates KM, Tizard I (2004). Isolation and characterization of structural components of *Aloe vera* L. leaf pulp. Int. Immunopharmacol. 4:1745-1755.
Ogunsanmi AO, Akpavie SO, Anosa VO (1994). Serum biochemical changes in West African dwarf sheep experimentally infected with *Trypanosoma brucei.* Rev. Elev. Med. Vet. Pays Trop. 47:195-200.
Maphosa V, Masika PJ (2010). Ethnoveterinary uses of medicinal plants: a survey of plants used in the ethnoveterinary control of gastro-intestinal parasites of goats in the Eastern Cape Province, South Africa. Pharm. Biol. 48(6):697-702.
Rabe C, Musch A, Schirmacher P, Kruis W, Hoffmann R (2005). Acute hepatitis induced by an *Aloe vera* preparation, a case report. World J. Gastroenterol. 11(2):303-304.
Reddy Uma CH, Reddy SK, Reddy J (2011). *Aloe vera* -A wound healer. Asian J. Oral Health Allied Sci. 1:91-92.
Rajasekaran S, Sivagnanam K, Subramanian S (2006). Antioxidant effect of *Aloe vera* gel extract in streptozotocin induced diabetes in rats. Pharmacol. Rep. 57:90- 96.
Kosif R, Aktas RG, Oztekin A (2008). The effects of oral administration of Aloe vera [*barbadensis*] on rat central nervous system: An experimental preliminary study. Neuroanatomy *7*:22-27.
Saif-ur-Rehman, Saghir AJ, Sajid H, Ishtiaq A, Muhammad N (2011). Studies on antidiabetic effect of *Aloe vera* extraction alloxan induced diabetes. Libyan Agric. Res. J. Int. 2(1):29-32.
Sikarwar MS, Patil MB, Sharma S, Bhat V (2010). *Aloe vera*: Plant of Immortality. Int. J. Pharm. Sci. Res. 1:7-10.
Toro G, Ackermann PG (1975). Practical Clinical Chemistry. 1st ed., Little Brown and Co. Inc., Boston, USA.
Urvashi N, Raju LB (2012). *Aloe vera* for human nutrition health and cosmetic use. Int. Res. J. Plant Sci. 3(3):38-46.

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

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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

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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 clinicopathological 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

Table 1. Age distribution of patients in this series.

Age	Number	Percent
5-10	18	6.3
10-20	83	29.4
20-30	73	25.7
30-40	70	24.6
40-50	24	8.4
50-60	6	2.1
>60	10	3.5

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

Duration (year)	Number	Percent
<1	50	17.6
1-5	142	50
5-10	63	22.2
>10	29	10.2

Table 3. Classification of pathological reports.

Pathology	Number	Percent
Chronic inflammation	184	64.7
Polyp	46	16.9
Allergic rhinosinusitis	27	9.5
Squamous cell carcinoma	8	2.81
Hemangioma	8	2.81
Craniophryngioma	6	2.1
Rhinolith	5	1.7

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

Anatomical variations	Number	Percent
Nasal septal deviation	207	79.2
Inferior turbinate hypertrophy	102	35.9
Bulla ethmoidalis	32	11.3
Concha bullosa	12	4.2
Uncinate bulla	12	4.2

polyposis or due to medialization of the lateral nasal wall;
2. Orbital abscess;
3. Intracranial complications;
4. Antrochoanal polyp;
5. Mucocoeles or mucopyocoeles;
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).

Table 5. Pre-operative symptoms in this study.

Pre-operative symptoms	Number	Percent
Nasal obstruction	69	24.3
Headache	63	21.9
Nasal congestion	53	18.5
Post nasal discharge	48	16.6
Breathing disorders	22	7.6
cough	11	3.6
Facial pain	8	2.9
hoarseness	6	1.8
Epistaxis	4	1.4

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

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 than12 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 polyp 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, craniophryngioma, 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.

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Conflict of Interests

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

REFERENCES

Adams PF, Hendershot GE, Marano MA (1999). Centers for Disease Control and Prevention/ National Center for Health Statistics. Current estimates from the National Health Interview Survey, 1996. Vital Health Stat. 10(200):1-203.

Luong A, Marple BF (2006). Sinus surgery. Clin Rev. Allergy Immunol. 30(3):217-222.

Anand VK (2004). Epidemiology and economic impact of rhinosinusitis. Ann. Otol. Rhinol. Laryngol. 193:3-5.

Benninger MS, Ferguson BJ, Hadley JA, Hamilos DL, Jacobs M, Kennedy DW, Donald CL, Bradley FM, Osguthorpe JD, Stankiewicz JA, Jack A, James D, Ivor E, Howard L (2003). Adult chronic rhinosinusitis: definitions, diagnosis, epidemiology, and pathophysiology. Otolaryngol. Head Neck Surg.129(Suppl):S1-32.

Bolger WE, Butzin CA, Parsons DS (1991). Paranasal sinus bony anatomic variations and mucosal abnormalities: CT analysis for endoscopic sinus surgery. Laryngoscope 101:56–64.

Bolger WE, Woodruff W, Parsons DS (1990). CT demonstration of pneumatization of the uncinate process. Am. J. Neuroradiol. 11:552.

Calhoun KH, Waggenspack GA, Simpson CB, Hokanson JA, Bailey BJ (1991). CT evaluation of the paranasal sinuses in symptomatic and asymptomatic populations. Otolaryngol. Head Neck Surg. 104:480–483

Calhoun KH, Waggenspack GA, Simpson CB, Hokanson JA, Bailey BJ (1991). CT evaluation of the paranasal sinuses in symptomatic and asymptomatic populations. Otolaryngol. Head Neck Surg. 104:480–483.

Chiu AG, Kennedy DW (2004). Surgical management of chronic rhinosinusitis and nasal polyposis: a review of the evidence. Curr. Allergy Asthma Rep. 4:486–489

Cook PR, Begegni A, Bryant WC, Davis WE (1995). Effect of partial middle turbinectomy on nasal airflow and resistance. Otolaryngol. Head Neck Surg.113(4):413-9.

Danese M, Duvoisin B, Agrifoglio A, Cherpillod J, Krayenbuhl M (1997). Influence of nasosinusal anatomic variants on recurrent, persistent or chronic sinusitis. X-ray computed tomographic evaluation in 112 patients. J. Radiol. 78:651–657

Daniel L (2011). Hamilos. Chronic rhinosinusitis: Epidemiology and medical management. J. Allergy Clin. Immunol. 128:693-707

Fokkens W, Lund V, Mullol J (2007). European Position Paper on Rhinosinusitis and Nasal Polyps group. Rhinol Suppl 20:1-136.

Giacchi RJ, Lebowitz RA, Jacobs JB (2000). Middle turbinate resection: issues and controversies. Am. J. Rhinol. 14:193-7.

Hamdan AL, Bizri AR, Jaber M, Hammoud D, Baino T, Fuleihan N (2001). Nasoseptal variation in relation to sinusitis: a computerized tomographic evaluation. J. Med. Liban. 49(1):2–5.

Hashemi SA, Abediankenari S, Madani SA, Akbari M (2012). Comparison of salivary IgA, tear IgA and serum IgE in patients suffering from chronic rhinosinusitis. Int. J. Med. Investig. 1:31-37.

Hisham SK, Ahmed ZE, Nicholas C (2011). Radiological findings in patients undergoing revision endoscopic sinus surgery: a retrospective case series study. BMC Ear, Nose Throat Disord. 11:4

Is¸yk AO, Bulut S (1994). Coccha bullosa. Relations with sinus disease and septal deviation. Turk. J. Diagn. Intervent. Radiol. 1:301–4.

Jamie SS, Joao NL, Peter MS (2004).The Incidence of Concha Bullosa and Its Relationship to Nasal Septal Deviation and Paranasal Sinus Disease. AJNR Am J Neuroradiol. 25:1613–1618.

Kennedy DW (1992). Prognostic factors, outcomes and staging in ethmoid sinus surgery. Laryngoscope 102(12 Pt 2 Suppl 57):1–18.

Kennedy DW (1985). Functional endoscopic sinus surgery. Technique. Arch. Otolaryngol. 111(10):643–649.

Kennedy DW, Zinreich SJ (1988). Functional endoscopic approach to inflammatory sinus disease: current perspectives and technique modifications. Am. J. Rhinol. 2:89–96.

Koen VC, Nan Z, Philippe G, Peter T, Claus B (2011). Pathogenesis of chronic rhinosinusitis: Inflammation. J. Allergy Clin. Immunol. 128(4):728-32.

Lam WW, Liang EY, Woo JK, Van Hasselt A, Metreweli C (1996). The etiological role of concha bullosa in chronic sinusitis. Eur Radiol. 6:550–552.

LaMear WR, Davis WE, Templer JW, McKinsey JP, Del Porto H (1992). Partial endoscopic middle turbinectomy augmenting functional endoscopic sinus surgery. Otolaryngol. Head Neck Surg. 107(3):382-9.

Lawson W (1994). The intranasal ethmoidectomy: evolution and an assessment of the procedure. Laryngoscope 104(6 Pt 2):1-49 .

Meloni F, Mini R, Rovasio S, Stomeo F, Teatini GP (1992). Anatomic variations of surgical importance in ethmoid labyrinth and sphenoid sinus. A study of radiological anatomy. Surg. Radiol. Anat. 14:65–70.

Pleis JR, Lucas JW, Ward BW (2009). Summary health statistics for

U.S. adults: National Health Interview Survey, 2008. Vital Health Stat 10(242):1-157.
Rao VM, El-Noueam KI (1998). Sinonasal imaging. Anatomy and pathology. Radiol. Clin. North Am. 36:921–39.
Senior BA, Kennedy DW, Tanabodee J, Kroger H, Hassab M, Lanza D (1998). Long-term Results of Functional Endoscopic Sinus Surgery. Laryngoscope 108(2):151-157.
Shechtamn FG, Kraus WM, Schaefer SD (1993). Inflammatory diseases of the sinuses. Anatomy. Otolaryngol Clin. North Am. 26:509–15.
Shin HS (1986). Clinical significance of unilateral sinusitis. J. Korean Med. Sci. 1:69–74.
Stammberger H (1985). Endoscopic surgery for mycotic and chronic recurring sinusitis. Ann. Otol. Rhinol. Laryngol. Suppl. 119:I–II.
Stewart MG (1998). Middle turbinate resection. Arch. Otolaryngol. Head Neck Surg.124:104-6.
Zinreich SJ, Kennedy DW, Rosenbaum AE, Gayler BW, Kumar AJ, Stammberger H (1987). Paranasal sinuses: CT imaging requirements for endoscopic surgery. Radiology 163:769–75.

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