The Beneficial Effects of Lipoic Acid Versus Coenzyme Q10 on Arginine-Induced Acute Pancreatitis in Albino Rats: Histological and Immunohistochemical study

Original Article

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ABSTRACT

Background: Acute pancreatitis (AP) is a self-limiting condition to a widespread, deadly multi-organ failure that can result in death.

Objectives: To compare the protective effect of alpha lipoic acid (ALA) and Coenzyme (Q10) in an experimental rat model of L-arginine-induced AP.

Materials (Subjects): Forty adult male albino rats were randomized equally into 4 groups: (i) The control group; (ii) The AP group received L-Arginine by intraperitoneal (IP) injection in two doses of 2 g/kg body weight each, 1 hour apart, (iii) The ALA+AP group was injected with L-Arginine and intramuscular (IM) injection of ALA in two doses of 60mg/kg body weight; 1 hour before L-arginine injection and 24 hours after the second dose of L-arginine; (iii) The Coenzyme Q10+AP group was injected with L-Arginine and IM injection of Q10 in two doses of 30mg/kg body weight; 1 hour before L-arginine injection and 24 hours after the second dose of L-arginine. Histological, immunohistochemical for HSP70 and NF-κB, and morphometric studies were done followed by statistical analysis.

Results: AP group showed significant distortion of the pancreatic acini with obvious cellular damage while those treated by ALA and Coenzyme Q 10 especially ALA showed significant pancreatic tissue improvement as shown by HSP70 and NF-κB immunoexpressing.

Conclusions: ALA and Q 10 therapy proved a clear protective effect in suppressing experimentally generated AP in male albino rats, but the improvement was more obvious with ALA.

Key Words: Acute Pancreatitis, Coenzyme Q10, Lipoic Acid and HSP70, NF-kB.

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INTRODUCTION

Acute pancreatitis is a non-infectious inflammatory disease of the pancreas. Complications and death from AP may be experienced by as many as 20 % of patients[1]. Multiple organ failure is frequently brought on by severe AP[2]. Excessive changes in cytokine levels and free oxygen formation have been shown to play a role in the pathogenesis of AP[3]. A prior study has shown that free radicals can damage pancreatic acinar cells by lowering antioxidants in tissues, resulting in ischemia-reperfusion injury[4]. Despite being widely prevalent, there is currently no pharmacological treatment that can stop the disease's progression[5]. The acute phase of pancreatitis strongly alters gene expression, which can help the pancreas by protecting itself from acute attacks or by participating conversely in the disease mechanism. Hence, new strategies for the treatment of this disease can be discovered by identifying genes involved in cellular responses[6].

L-arginine (LA) is an essential amino acid that has shown a vasodilator effect. It is the precursor of nitric oxide (NO)[7]. LA has a positively charged guanidine group, binds the phosphate anion, and is often found in the active centers of proteins that bind phosphorylated substrates[8]. The mechanism by which LA causes pancreatitis is not exactly known. Many suggested mechanisms include oxygen free radicals[9] nitric oxide[10] inflammatory mediators[11], and autophagy[12]. As reports revealed, autophagy controls the signaling of oxidative stress and inflammation. Dysregulated autophagy may encourage the pancreatic inflammatory response[13].

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Alpha lipoic acid is a strong antioxidant with anti-inflammatory effects. It has been shown that ALA prevents free radicals from causing oxidative damage[14].

Coenzyme Q10 (ubiquinone) is a fat-soluble antioxidant with a core of 2, 3-dimethoxy-5-methylbenzoquinone and is the most common type of coenzyme Q in human tissues[15]. Antioxidant and regenerating antioxidant activity is one of its fundamental roles, stimulating cell growth and inhibiting cell death[14], and acting as an essential ROS scavenger[16]. Multiple heat shock proteins (HSPs) may be involved in the cytoprotective effect in the rat pancreas[6]. Stress-induced HSPs can safeguard against resulting cellular damage. Pancreatic heat shock protein expression was affected by hyperthermia and there was a large rise in HSP70 and HPS27; but no change in HSP60 or HSP90. Hyperthermia has significant protection against subsequent arginine-induced acute pancreatitis. More specifically, the degradation and disorganization of the actin cytoskeleton, an important early component of acute pancreatitis, was prevented[6]. The purpose of the study is to compare the protective effects of ALA and Coenzyme Q10 in an experimental rat model of L-arginine-induced AP.

MATERIALS AND METHODS

Ethical Approval:

The Research Ethics Committee (REC) for Human and Animal Research of the Faculty of Medicine, Helwan University, Cairo, Egypt, serial: 72- 2021, approved the study protocol.

Animals:

For this study, 40 mature male albino rats, with an average weight of 200250- gm, were locally raised at the Animal Care Modern Center in the National Research Center. The animals were kept in standard room temperature housing, exposed to daily natural light-dark cycles, and given unlimited access to food and drink.

Experimental design:

Ten rats were placed in one of four groups:

The control group (Gp): The rats were subdivided equivalently into negative and positive control. The negative control received nothing. The positive control was given two IP injections of normal saline spaced by one hour.

AP Gp: The rats were injected IP with LA (Sigma-Aldrich, St. Louis, MO, USA). The injection was done in two doses of 2 g/kg body weight, each spaced by one hour. LA was dissolved in normal saline[17].

ALA+AP Gp: The rats were injected IP with LA as in the AP group. In addition, IM injection with ALA (Empower pharmacy) was done. ALA was dissolved in normal saline and given in two doses of 60mg/kg body weight one hour before the first dose of LA and 24 hours after the second dose of LA[18].

Coenzyme Q 10+AP Gp: The rats were injected IP with LA as in the AP group, in addition to an IM injection of coenzyme Q10 (Empower pharmacy). Coenzyme Q10 was dissolved in normal saline and given in two doses of 30mg/kg body weight, one hour before the first dose of LA and 24 hours after the second dose of LA[19].

Histological study:

After 48 hours of the second injection of LA, thiopental sodium 50 mg/kg subcutaneous injections were used to anesthetize the rats before they were sacrificed by cervical dislocation[20]. After being removed from the body, the pancreas was fixed in a 10 % formalin solution. Dehydrated specimens were treated to create paraffin blocks. Sections five µm thick were subjected to the following studies:

I- Hematoxylin and Eosin (H&E) staining[21].

II- Immunohistochemical staining was performed using HSP70 Ab-2 (Cat. # MS-482) and NF-κB /p65 (Cat. #RB-1638) (Lab Vision, Thermo Scientific, USA). HSP70 is a mouse monoclonal antibody with a nuclear and cytoplasmic reaction. Its positive control was breast cancer. Its increased expression was used as a marker for tissue protection against stress[22]. NF-κB p65 is a rabbit polyclonal antibody with a cytoplasmic reaction. It was used for the detection of tissue inflammation[23]. Pancreatic tissue samples were fixed in neutral buffered formalin, embedded in paraffin, sectioned, and mounted on slides. After being deparaffinized, endogenous peroxidase activity was inhibited with 10 % H2O2 for 10 min. After being blocked with goat serum, the sections were incubated with HSP70 diluted primary antibody (1:100) at 37 °C for 40 minutes. Other slides were incubated with NF-κB p65 diluted primary antibody (1:50) at 37 °C for 30 minutes. Slides were then counterstained with hematoxylin for 1 min. After dehydration, the slides were coverslipped. To ensure specificity of the

immunoreactions, control sections were subjected to the same immunohistochemical method with the exception that the primary Ab antibody was replaced with PBS. An additional slide of the pancreatic specimen was treated with buffer solution instead of the same concentration of primary antibody in every run as the negative control.

Histopathological scoring:

H&E stained sections were blindly assessed for each group by three examiners according to the following scoring criteria[24]: i) interstitial edema: 0 points= none, 1 point= interlobular, 2 points= lobule involvement, and 3 points= isolated island-like acinar cells; ii) leukocyte infiltration: 0 points= none, 1 point=< 20 %, 2 points= 20 - 50 %, and 3 points=> 50 %; iii) acinar cell necrosis: 0 points=none, 1 point=<5%, 2 points= 5 - 20 %, and 3 points=>20%; (iv) hemorrhage: 0 points= none, 1 point= 12 ,2- points= 3 - 5 and 3 points=> 20 %.

Morphometric study:

The computer system with the image analyzer (Leica Qwin 500, England) was used to get the morphometric data. The area percentage of HSP70 and NF-κB immunoexpression (IE) were measured in 10 high-power non-overlapping fields in each specimen using binary mode.

Statistical analysis:

The data were expressed as group means ± SD. The statistical analysis was carried out using a one-way analysis of variance (ANOVA). A value of $p \leq 0.05$ was accepted as statistically significant[25]. with SPSS version 10 (SPSS, Chicago, IL, USA). Followed by Bonferroni post-hoc test to detect which pairs of groups are significant.

RESULTS

Histological and immunohistochemical results:

A- Hematoxylin and Eosin staining:

The control Gp showed the characteristic normal appearance of pancreatic acini with apical acidophilic granules, basal basophilia, and basal round nuclei. In addition, lightly stained islets of Langerhans appear between the acini (Figure 1a). The pancreatic intralobular ducts lined by cuboidal epithelium are demonstrated (Figure 1b). AP Gp showed distortion, and vacuolation of pancreatic acinar cells and darkly eosinophilic cells with small pyknotic nuclei in addition to karyolysis in multiple nuclei (Figure 2). Congested blood vessels and mononuclear inflammatory cells infiltration were observed (Figure 3). There were dilated intralobular pancreatic ducts (Figure 4). ALA+ AP Gp showed apparently normal architecture of pancreatic acini with apical acidophilic granules, basal basophilia, and basal round nuclei. In addition, lightly stained islets of Langerhans appeared between the acini (Figure 5). Coenzyme Q10+AP Gp showed many improved pancreatic acini and apparently normal intralobular duct. Lightly stained islets of Langerhans in between the acini were present. Vacuolation of a few pancreatic acinar cells, darkly eosinophilic cells with a small pyknotic nucleus, and eosinophilic material in degenerated acini were still noted (Figure 6).

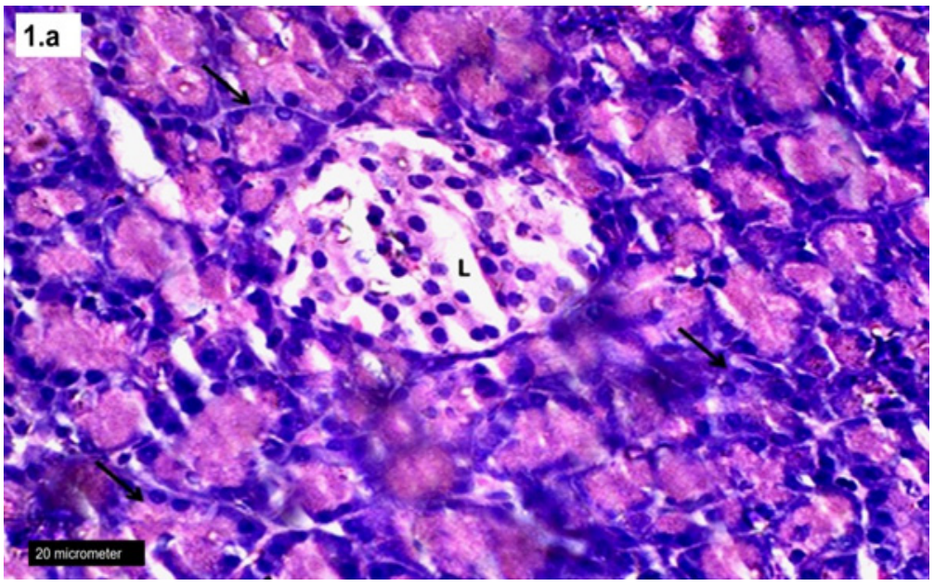

Figure 1(a): A photomicrograph of a section in the rat pancreas of the control group showing the characteristic normal appearance of pancreatic acini (arrows) with apical acidophilic granules, basal basophilia, and basal round nuclei. lightly stained islets of Langerhans (L) are seen in between the acini. (H&E, 400x).

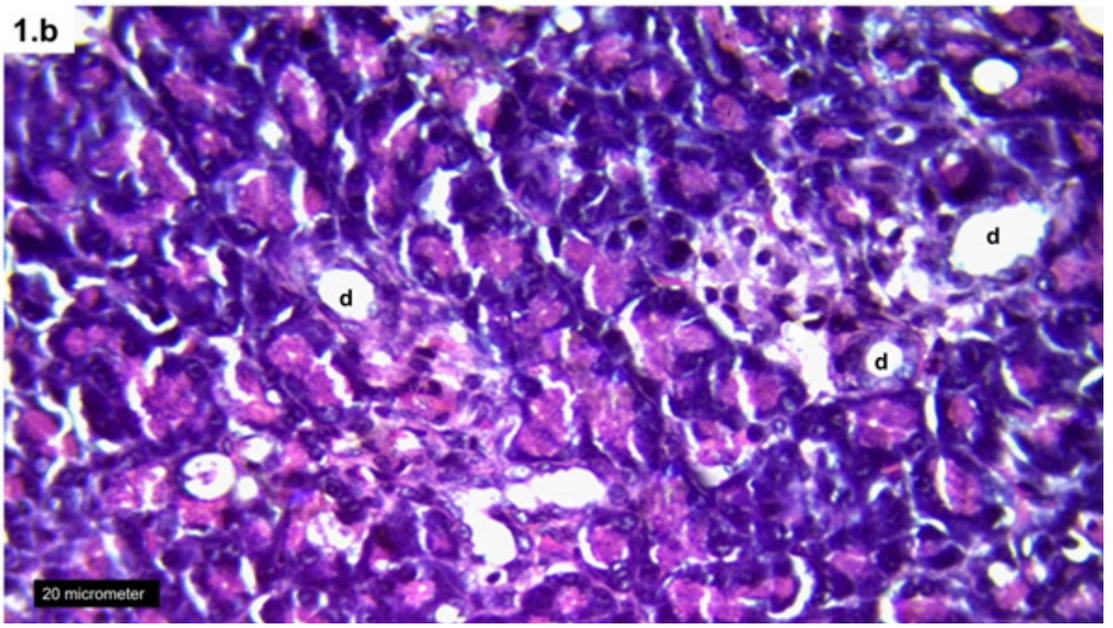

Figure 1(b): A photomicrograph of a section in the rat pancreas of the control group showing pancreatic intralobular ducts (d) in-between the acini and lined by cuboidal epithelium. (H&E, 400x).

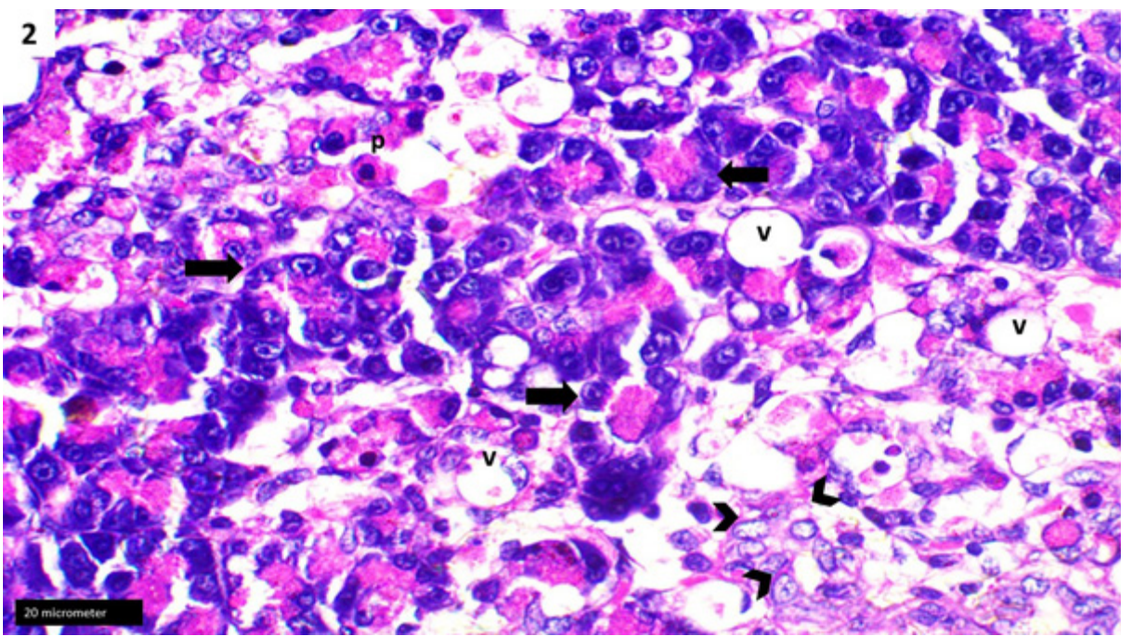

Figure 2: A photomicrograph of a section in the rat pancreas of the AP group showing distorted acini (thick arrows), vacuolation of pancreatic acinar cells (V), darkly eosinophilic cell with a small pyknotic nucleus (P) and karyolysis (arrowheads) in multiple nuclei. (H&E, 400x).

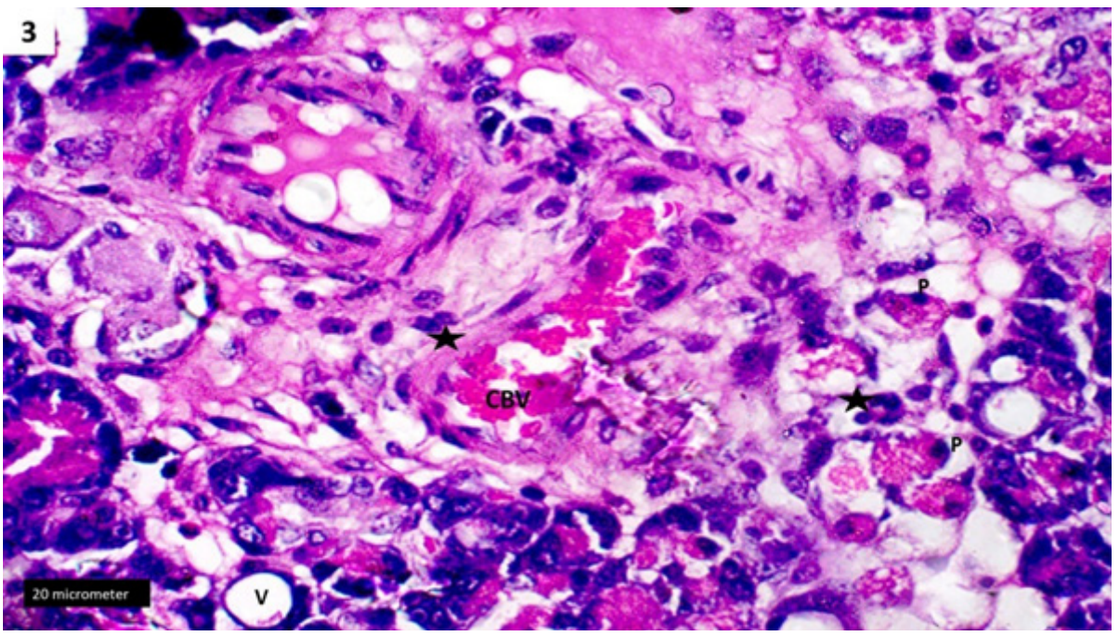

Figure 3: A photomicrograph of a section in the rat pancreas of the AP group showing distortion, vacuolation of the pancreatic acinar cell (V), destroyed darkly eosinophilic cells with small pyknotic nuclei (P), congested blood vessel (CBV), infiltration of mononuclear inflammatory cells (star). (H&E, 400x).

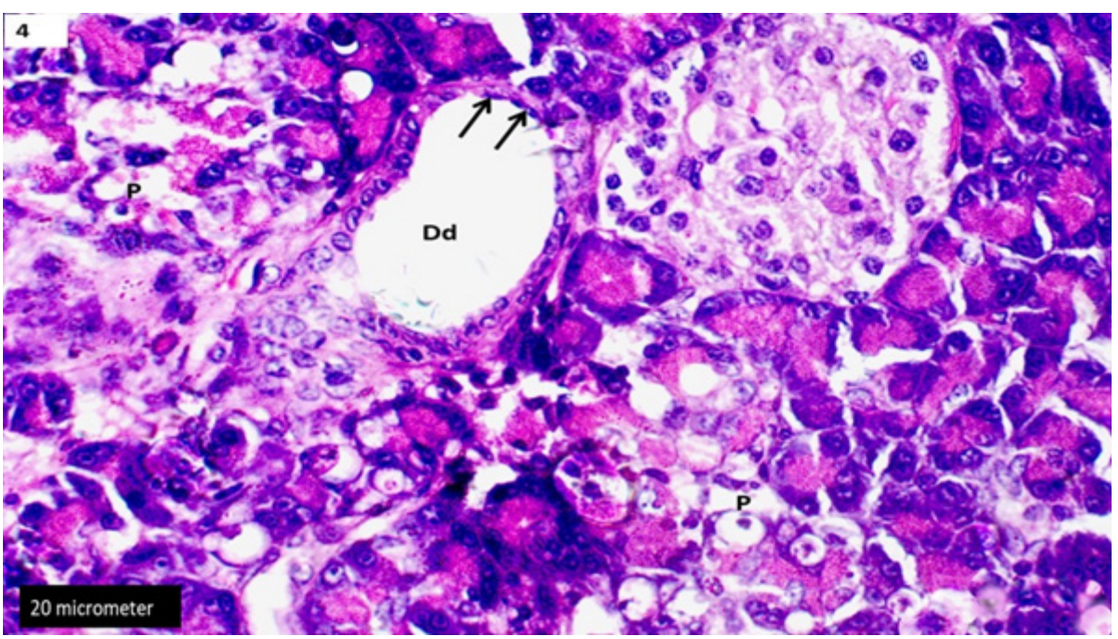

Figure 4: A photomicrograph of a section in the rat pancreas of the AP group showing dilated intralobular duct (Dd, in addition to small pyknotic nuclei (P). Note flat-lining epithelium instead of the cuboidal epithelium (arrows) (H&E, 400x).

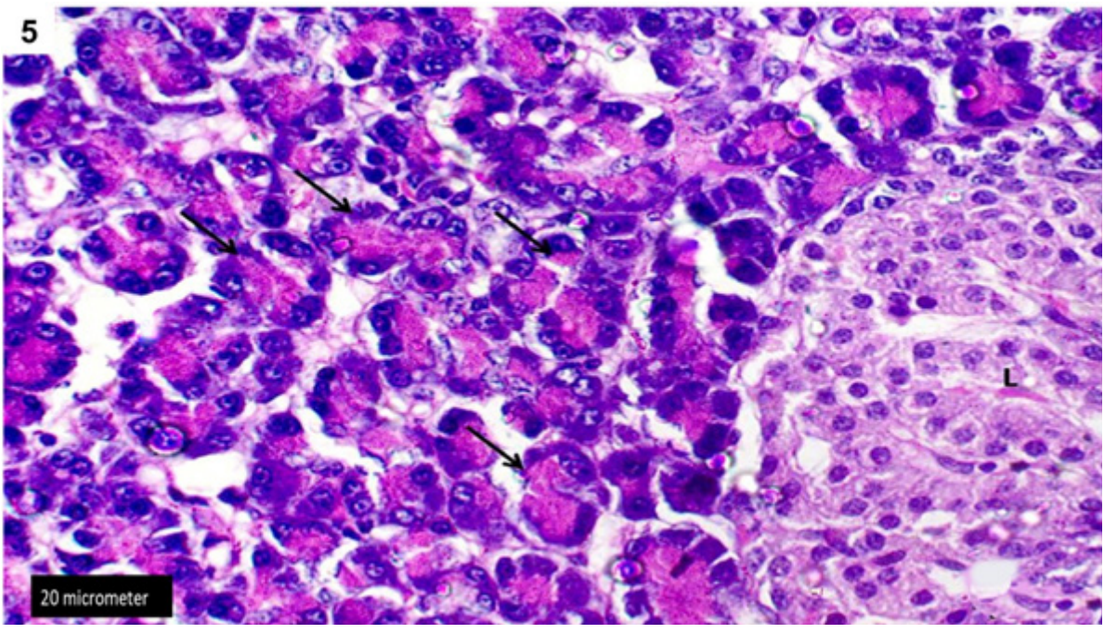

Figure 5: A photomicrograph of a section in the rat pancreas of the ALA and AP group showing the pancreatic acini appears normal with apical acidophilic granules, basal basophilia, and basal round nuclei (thin arrows) and lightly stained islets of Langerhans (L) in between the acini. (H&E, 400x).

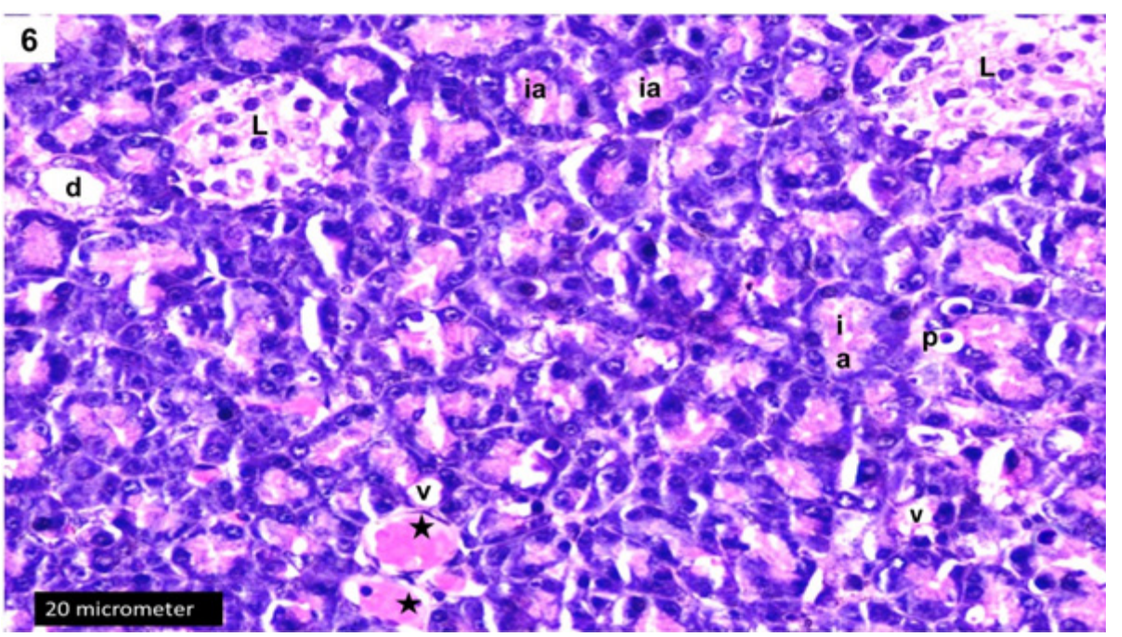

Figure 6: A photomicrograph of a section in the rat pancreas of coenzyme Q 10 and AP group showing many improved pancreatic acini (ia) and intralobular duct (d), in addition to lightly stained islets of Langerhans (L) in between the acini. Note vacuolation of a few pancreatic acinar cells (V), darkly eosinophilic cells with a small pyknotic nucleus (P), and eosinophilic material in degenerated acini (stars) (H&E, 400x).

B- Histopathological scores:

There was a significant difference in the scores of AP Gp in relation to ALA+AP Gp, and coenzyme Q10+AP Gp. Besides, the difference between ALA and AP Gp and coenzyme Q10 and AP Gp was significant Table (1).

Table 1: AP Histopathological scores:

Groups	Interstitial edema	Leucocyte infiltration	Acinar cell necrosis	Hemorrhage	Total
Group I (Control Gp)	0 ± 0.00	0 ± 0.00	0 ± 0.00	0 ± 0.00	0 ± 0.00
Group II (AP Gp)	2.91 ± 0.82[(a)]	1.4 ± 0.17[(a)]	3.00 ± 0.00[(a)]	2.15 ± 0.5[(a)]	9.46 ± 1.49[(a)]
Group III (ALA +AP Gp)	0.33 ± 0.1[(b)]	0.39 ± 0.09[(b)]	0.1 ± 0.01[(b)]	0.2± 0.03[(b)]	1.02± 0.23[(b)]
Group IV(Coenzyme Q10 and AP Gp)	1.1 ± 0.19[(c)]	0.78 ± 0.16[(c)]	0.21 ± 0.01[(c)]	1.1± 0.06[(c)]	3.19 ± 0.42[(c)]

Significant ≤ 0.05.
(a) Significant compared to groups III &IV.
(b) Significant compared to groups II &IV.
(c) Significant compared to groups II &III.

C- Immunohistochemical staining:

HSP70 IE:

The pancreatic section of the control Gp showed positive IE in the nucleus and cytoplasm of a few acinar cells (Figure 7). AP Gp showed positive IE in the nucleus and cytoplasm of some pancreatic acinar cells (Figure 8). ALA and AP Gp demonstrated positive IE in the nucleus and cytoplasm of most pancreatic acinar cells (Figure 9). However, coenzyme Q10 and AP Gp revealed positive IE in the nucleus and cytoplasm of multiple pancreatic acinar cells (Figure 10).

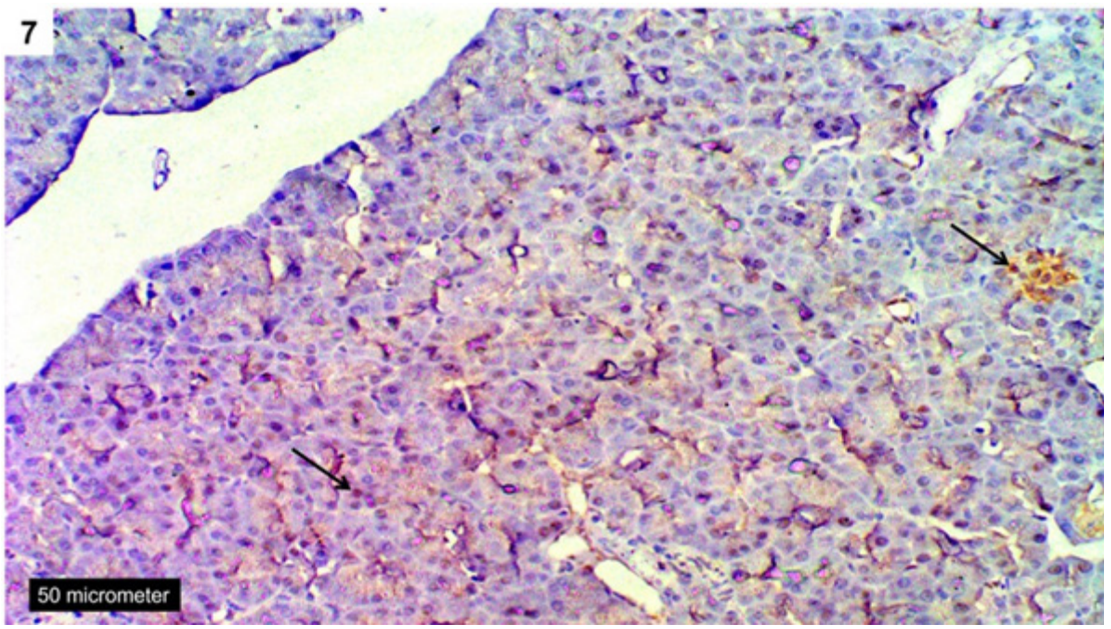

Figure 7: A photomicrograph of a section in the rat pancreas of the control group showing positive immunoexpression (IE) in the nucleus and cytoplasm of a few pancreatic acini (arrow). (HSP70, 200x).

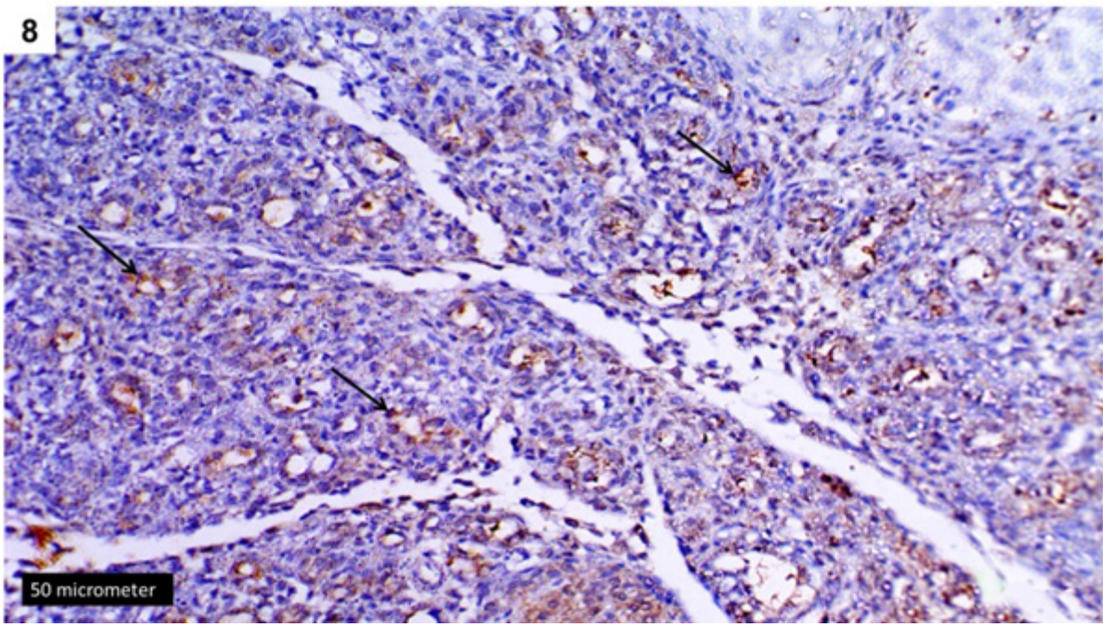

Figure 8: A photomicrograph of a section in the rat pancreas of the AP group showing positive IE in the nucleus and cytoplasm of some pancreatic acinar cells (arrows). (HSP70, 200x).

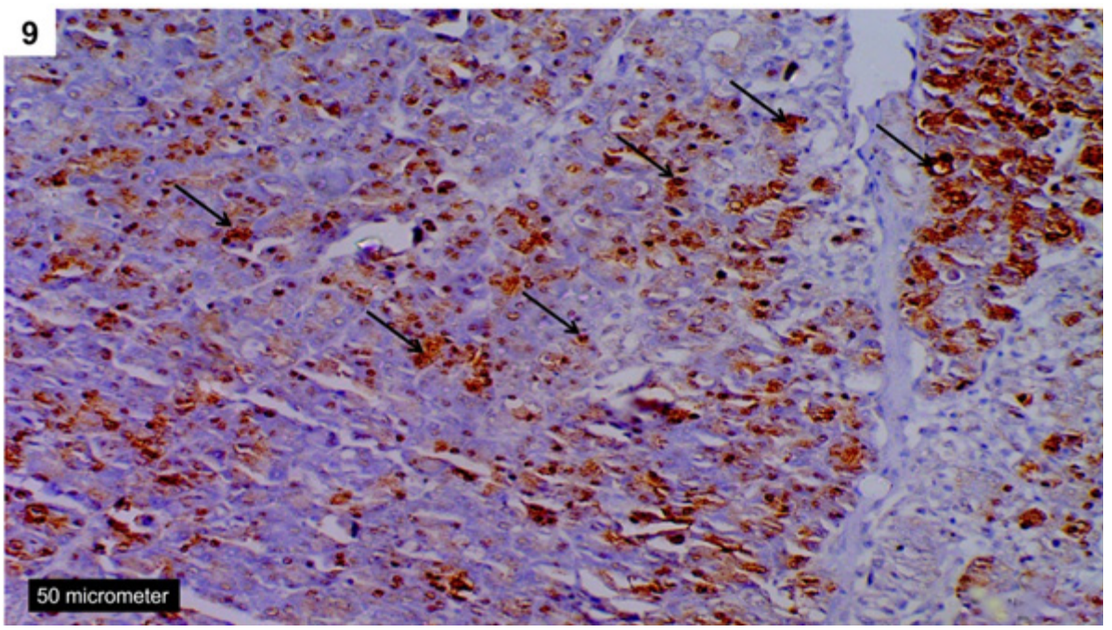

Figure 9: A photomicrograph of a section in the rat pancreas of ALA and AP group showing positive IE in the nucleus and cytoplasm of most pancreatic acini (arrows). (HSP70, 200x).

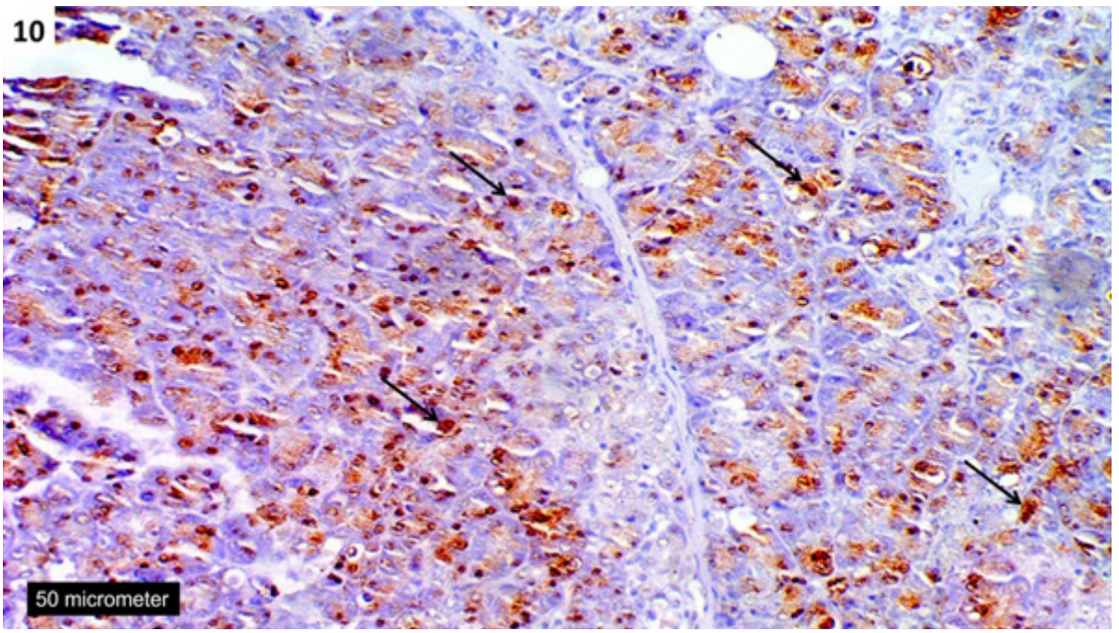

Figure 10: Photomicrograph of a section of a rat pancreas group coenzyme Q10 and AP showing positive IE in the nucleus and cytoplasm of multiple pancreatic acini (arrows). (HSP70, 200x).

NF-κB Immunostaining:

The control Gp showed negative nuclear and mild positive cytoplasmic IE in the pancreatic acinar cells (Figure 11). AP Gp showed marked positive IE in the nucleus and cytoplasm of widespread pancreatic acinar cells (Figure12). On the other hand, ALA and AP Gp showed positive IE in the nucleus and cytoplasm of a few pancreatic acinar cells (Figure13). Coenzyme Q10 and AP Gp showed positive IE in the nucleus and cytoplasm of some pancreatic acinar cells (Figure14).

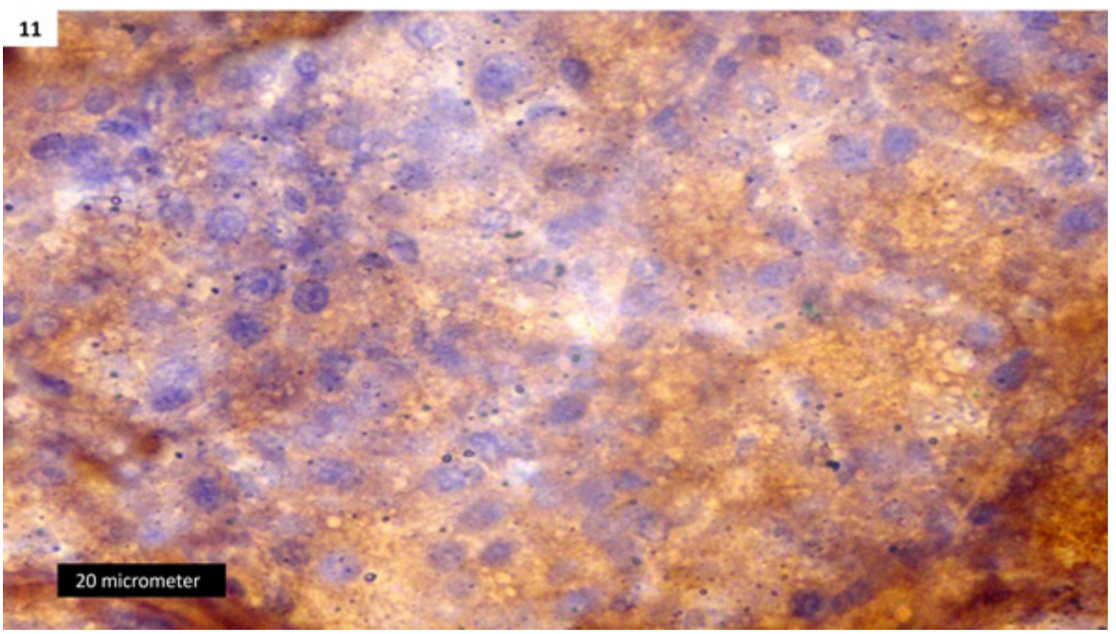

Figure 11: A photomicrograph of a section in the rat pancreas of the control group showing negative nuclear and mild positive cytoplasmic IE in the pancreatic acinar cells. (NF-kB, 400x).

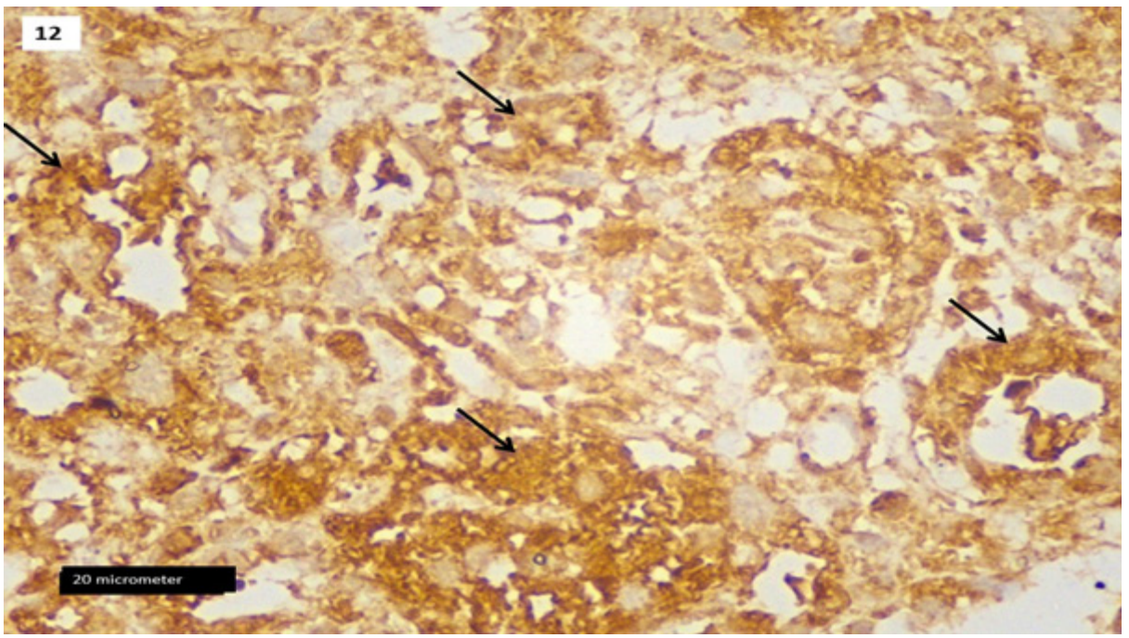

Figure 12: A photomicrograph of a section in the rat pancreas of the AP group showing marked positive IE in the nucleus and cytoplasm of widespread pancreatic acinar cells (arrows). (NF-kB, 200x).

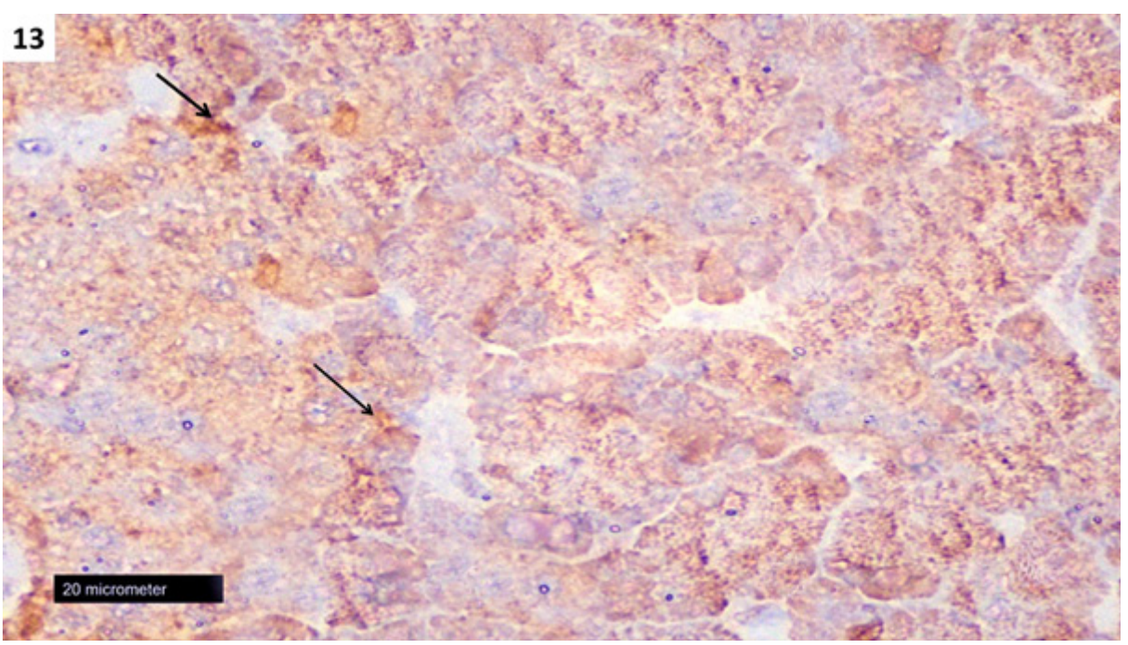

Figure 13: A photomicrograph of a section in the rat pancreas of the ALA and AP group showing positive IE in the nucleus and cytoplasm of a few pancreatic acini (arrows). (NF-kB, 400x).

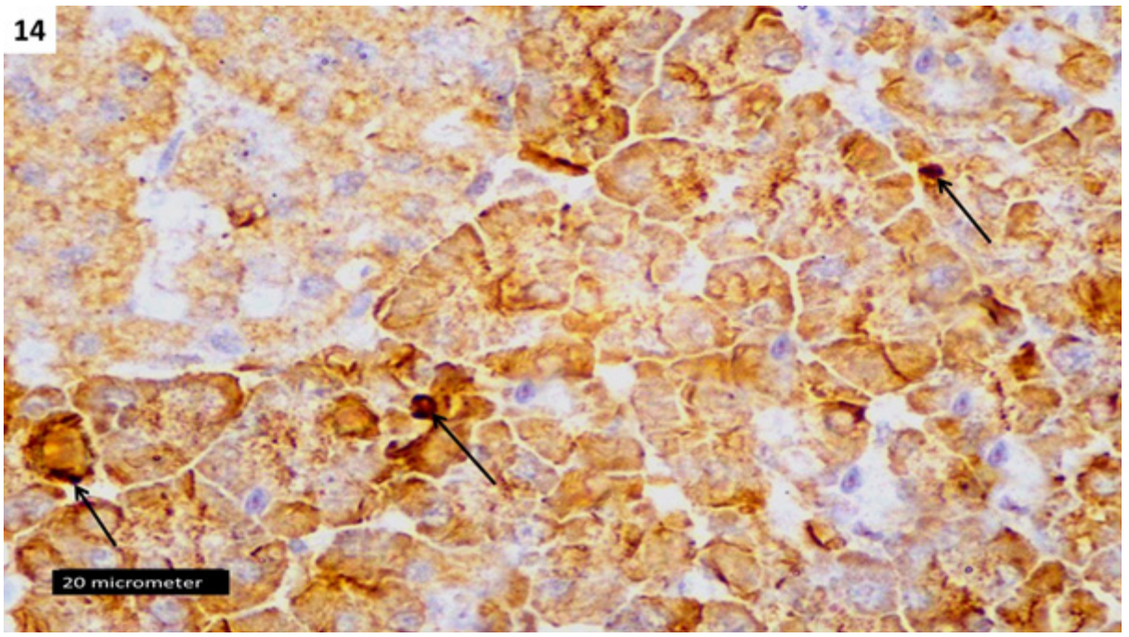

Figure 14: A photomicrograph of a section in the rat pancreas of coenzyme Q10 and AP group showing positive IE in the nucleus and cytoplasm of some pancreatic acini (arrows). (NF-kB, 400x).

Morphometric and statistical results:

HSP70 IE:

The area percentage of HSP70 determined a significant increase in ALA and AP Gp compared to all other groups. Coenzyme Q10 and AP Gp showed a significant increase when compared to AP Gp and the control group while AP Gp showed a significant increase when compared with the control group Table (2).

NF-kB IE:

The area percentage of NF-kB determined a significant increase in AP Gp compared to all other groups. On other hand, group Coenzyme Q10 and AP Gp showed a significant increase when compared to ALA and AP Gp and the control group. ALA and AP Gp showed no significance in the control group. Table (2).

Table 2: Area % of HSP70 and NFKB in pancreatic tissue IE (± SD):

Groups	Area % HSP70 (IE)	Area % NFKB (IE)
Group I (Control Gp)	0.124 ± 0.038	35.508 ± 5.37
Group II (AP Gp)	17.41 ± 4.65 ^	55.434 ± 7.49 *
Group III (ALA +AP Gp)	30.038 ± 5.699 *	38.682 ± 4.05@
Group IV (Coenzyme Q10 and AP Gp)	23.268 ± 2.14 #	45.78 ± 2.6 $

P significant increase < 0.05.
(^) Significant compared to group I.
(*) Significant compared to all other groups.
(#) Significant compared to group I and group II.
(@) Nonsignificant compared to group I.
($) Significant compared to group I and group III.

DISCUSSION

For improved management of AP, more efficient therapy choices need to be created because until now there has been no established treatment approach for AP; instead, only supportive care is provided. Therefore, in this work, we looked at how ALA and Q10 affected a rat model of AP brought on by LA.

Our study demonstrated that LA administration displayed distortion of pancreatic acinar cells, pyknotic and karyolysis in multiple nuclei. Congestion of blood vessels and infiltration of mononuclear inflammatory cells were observed. Dilated intralobular pancreatic ducts were noted. These histological findings concur with the findings of other earlier studies that documented comparable changes[26, 27]. Eşrefolu *et al.*[28] explained the cellular damage by stating that lipids are one of the key targets for free radical harm after acute pancreatitis. By removing one hydrogen atom from polyunsaturated fatty acids, free oxygen radicals cause lipid peroxidation, which leads to the generation of hydroperoxides. According to the author, these reactions also damage the fluidity and integrity of cell membranes, which results in cell death. Therefore, these extracellular environment events set off several inflammatory cascades that cause more harm.

By avoiding pancreatic dysfunction brought on by free radicals, ALA significantly reduces morbidity and death[14]. In the current study, treatment with ALA revealed apparently normal pancreatic acini architecture. According to earlier research, ALA protects vessels from oxidative damage and has an anti-inflammatory effect on the nervous system and collagen tissue[29]. According to Bulut *et al.*[14], ALA lessens inflammation by reducing neutrophil activation and infiltration, which promotes tissue damage. Therefore, ALA protects the pancreatic tissue from the toxicity brought on by free radicals.

In our work, some of the pancreatic acini and intralobular ducts of the Coenzyme Q10 and AP Gp were improved. A few pancreatic acinar cells still exhibited vacuolation. Also, there were some degenerated acini that contained eosinophilic material. These findings could be explained by the findings of Mirmalek *et al.*[30] who found that therapy with coenzyme Q10 reduces inflammation by reducing inflammatory mediators and oxidative stress by increasing the expression of the main antioxidant enzymes in the experimental AP model. Therefore, by regulating cytokines and oxidative indicators as well as lowering pancreatic enzyme levels in the plasma, tissue damage can be mitigated. In a recent study, Q10 was also shown to reduce pancreatic acinar cell death, parenchymal edema, and inflammatory cell infiltration in the rat model of AP[31].

Immunohistochemical staining with HSP70 and NF-κB supported our histological findings. In both normal and stressed circumstances, the primary protein of the HSP family, HSP70, maintains the structure and functionality of cell homeostasis[32]. In our results, HSP70 was expressed normally in the nucleus and cytoplasm of scattered pancreatic acinar cells in the control Gp, and this is consistent with another study that reported HSP70 expression normally in all areas of renal tissue[33]. In the AP group, there was positive HSP70 IE in some distorted pancreatic acinar cells. Concomitantly, quantities of Hsp70 were produced by pancreatic acinar cells in AP experimental model rats and were suggested to play a protective role. They prevent intra-acinar activation of trypsinogen and acinar cell death and reduce the activation of NF-κB[34]. It was reported that extracellular HSP70 might induce a similar response to the systemic inflammatory response syndrome by activating toll-like receptor 4 (TLR4) in a murine model of AP induced by cerulein[35]. ALA and AP Gp showed an increased positive immunoexpression in most pancreatic acini, while Coenzyme Q 10 and AP Gp showed an increased positive IE in multiple pancreatic acini. In support, Feng *et al.*[36] stated that the expression of HSP70 protein was remarkably raised in renal tissues after resveratrol treatment, which has anti-inflammatory and antioxidant effects. An increase in HSP70 expression functions as a protection against cells that prevents oxidative damage and repairs damaged proteins by balancing the state of ischemia and increasing the production of free radicals[37]. An increase in the expression of Hsp70 is associated with an improvement in survival, less tissue damage, and a decreased inflammatory response[38]. In the case of AP induced by sodium taurocholate in rats, the administration of glutamine favors the expression of HSP70, conferring protection from the organic damage induced by the illness[23]. HSP70 regulates the inflammation response through various mechanisms, one of which is the inhibition of the secretion of HMGB1[39]. On the contrary, it has been shown that vitamin E seems to diminish the rise in HSP70 expression. Whether the reduction of HSP70 by antioxidants is beneficial or harmful is still controversial and needs to be considered[40].

NF-κB is a central mediator of pro-inflammatory gene induction and functions in both innate and adaptive immune cells. Hence, a well-recognized

function of it is the regulation of inflammatory response. Deregulated inflammatory responses can cause excessive or long-lasting tissue damage, contributing to the development of acute or chronic inflammatory diseases. The function of NF-κB is also required for maintaining normal immune responses and cell survival. Thus, in normal tissue, NF-κB is usually expressed in the cytoplasm of the cells[41]. In the present study, the control Gp showed a negative nuclear and mild positive cytoplasmic IE in the pancreatic acinar cells. It was documented that after cellular injury, nuclear translocation occurs to induce the expression of various pro-inflammatory genes, including those encoding cytokines and chemokines. Its expression changes from cytoplasmic to nuclear and cytoplasmic[41]. AP Gp showed marked positive IE in the nucleus and cytoplasm of many pancreatic acinar cells. NF-B is quickly activated in the early stages of AP and causes several inflammatory reactions[42]. During AP, the NF-κB pathway is crucial in controlling the transcription and synthesis of inflammatory cytokines[43]. It has been demonstrated that pharmacological inhibitors of NF-kB reduce the severity of AP[44]. On the other hand, ALA and AP Gp showed positive IE in the nucleus and cytoplasm of a few pancreatic acinar cells, while the coenzyme Q10 and AP Gp showed positive IE in the nucleus and cytoplasm of some pancreatic acinar cells. In accordance, immunofluorescence labeling was used to show that betulinic acid inhibits the NF-κB signaling pathway, which has both preventative and therapeutic effects on AP[45]. Melatonin reduces the harmful effects of oxidative stress on ovarian damage caused by diabetes mellitus. The diabetic rats treated with melatonin had considerably reduced immunoexpression of NF-κB[46].

In our work, morphometric and statistical analysis of HSP70 IE demonstrated that ALA+AP Gp and coenzyme Q10+AP Gp were significantly higher than AP and control groups, indicating improvement. However, there was a significant difference between the ALA+AP Gp and coenzyme Q10+AP group, denoting that the treatment by ALA improves acute pancreatitis better than treatment by coenzyme Q10. The area percentage of NF-κB determined a significant increase in AP Gp compared to all other groups, indicating significant inflammation of pancreatic acini. On the other hand, coenzyme Q10+AP Gp showed a significant increase when compared to ALA+AP Gp, and the control group indicated that inflammation of pancreatic acini is still found. ALA+AP Gp showed no significance compared to the control group, indicating that inflammation of pancreatic acini is resolved.

CONCLUSION

In In conclusion, both ALA and coenzyme Q10 had protective benefits in preventing AP in male albino rats, but ALA's effect was more pronounced due to its activation of HSP70. More AP research in the future will focus on using HSP70 as a treatment.

CONFLICT OF INTEREST

There is no potential conflict of interest among the authors.

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الملخص العربي

الآثار المفيدة لحمض الليبويك مقابل الإنزيم المساعد كيو ١٠ على الالتهاب الحاد للبنكرياس المستحث بالأرجينين في الجرذان البيضاء: دراسة هيستولوجية وهيستوكيميائية مناعية

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المقدمة: يتراوح التهاب البنكرياس من حالة معتدلة ومحدودة ذاتيًا إلى فشل في العديد من الأعضاء ويمكن أن يؤدي إلى الوفاة. ويمتلك حمض ألفا ليبويك والإنزيم المساعد كيو ١٠ تأثيرقوي كمضاد للأكسدة ومضاد للالتهابات. وهذا التأثير قد يخفف الضرر التأكسدي في التهاب البنكرياس الحاد عن طريق آليات مضادات الأكسدة المباشرة.

الهدف: مقارنة التأثير الوقائي لحمض الليبويك مقابل الإنزيم المساعد كيو ١٠في نموذج الجرذ التجريبي لالتهاب البنكرياس الحاد المستحث بالأرجينين.

المواد والأساليب: تم تقسيم أربعين من ذكور الجرذان البيضاء البالغه بشكل عشوائي إلى 4 مجموعات، 10 فئران في كل مجموعة: أعطيت المجموعة الضابطة حقنتين داخل الغشاء البريتوني من محلول ملحي عادي، بفارق ساعة واحدة. تم حقن مجموعة التهاب للبنكرياس الحاد بالأرجينين على جرعتين كل منهما 2 جم / كجم من وزن الجسم، على بعد ساعة واحدة. تم إذابة الأرجينين في محلول ملحي عادي. تم حقن مجموعة حمض اليبويك + التهاب البنكرياس الحاد بالأرجينين + حمض ليبويك المذاب في محلول ملحي عادي في العضل في جرعتين 60 مجم / كجم من وزن الجسم قبل ساعة واحدة من الجرعة الأولى للأرجينين و 24 ساعة بعد الجرعة الثانية للأرجينين. تم حقن مجموعة الإنزيم المساعد كيو ١٠+ التهاب للبنكرياس الحاد بالأرجينين +الإنزيم المساعد كيو ١٠ المذاب في محلول ملحي عادي في جرعتين من 30 مجم / كجم من وزن الجسم قبل ساعة واحدة من الجرعة الأولى للأرجينين و 24 ساعة بعد الجرعة الثانية من الأرجينين. أجريت الدراسات النسيجية والكيميائية المناعية والمورفومترية متبوعة بالتحليل الإحصائي.

النتائج: أظهرت مجموعة التهاب للبنكرياس الحاد تشوهًا كبيرًا لخلايا البنكرياس بينما أظهرت تلك التي عولجت بواسطة حمض اليبويك ومجموعة الإنزيم المساعد كيو ١٠ وخاصة حمض اليبويك تحسنًا ملحوظًا في أنسجة البنكرياس وتم تأكيد ذلك باستخدام الصبغات المناعيه HSP70 و NF-κB.

الخلاصة: أثبت علاج حمض اليبويك والإنزيم المساعد كيو ١٠تأثيرًا وقائيًا واضحًا في قمع التهاب للبنكرياس الحاد المتولد تجريبياً في ذكور الجرذان البيضاء، لكنه كان أكثر وضوحًا مع حمض اليبويك.

الكلمات المفتاحية: التهاب البنكرياس الحاد، إل-أرجينين، حمض اليبويك، الإنزيم المساعد كيو ١٠.