EFFECT OF ROSIGLITAZONE, A PEROXISOME PROLIFERATOR ACTIVATED RECEPTOR-GAMMA, ON L-ARGININE-INDUCED ACUTE PANCREATITIS IN EXPERIMENTAL ALBINO RATS

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ABSTRACT

The treatment for acute pancreatitis (AP) is still based on supportive care. The search for a new drug is a continuing challenge for many researchers. Rosiglitazone, an oral anti-hyperglycemic agent used for non-insulin-dependent diabetes mellitus, is a high-affinity synthetic agonist for peroxisome proliferator-activated receptor-gamma (PPAR-γ) and was also discovered to have anti-inflammatory effect. This study was carried out to investigate the effect of different doses of rosiglitazone on the development of acute pancreatitis (AP) induced experimentally by L-arginine in albino rats. Sixty-four male rats were divided into four groups: group I (control), group II (rosiglitazone group) subdivided into 3 subgroups (IIa,b,c) and were received different doses of the drug (3,10,50 mg/kg as a single oral dose), group III (AP group) were received single injection of L-arginine (250 mg/100gm b.w., i.p.) and group IV (rosiglitazone pre-treated group) subdivided into 3 subgroups (IVa,b,c) and were administered different doses of the drug (3,10,50 mg/kg as a single oral dose) one hour prior to AP induction. The rats were sacrificed at 24 hours after AP induction, blood samples were collected for assessment of serum amylase, MDA and catalase enzyme activity, in addition, TNF-α and IL-6 were determined in the serum. Pancreatic tissues were also collected for histopathological examination. Our results revealed that rosiglitazone pre-treatment produced dose-dependent attenuation of pancreatic tissue damage as evidenced by reduction of serum amylase and improvement of pancreatic histology. Rosiglitazone pre-treated rats also showed significantly decreased levels of pro-inflammatory cytokines (TNF-α and IL-6) and the lipid peroxidation marker (MDA) while the serum catalase activity was increased in a dose-dependent manner. In the light of these findings, rosiglitazone confers significant dose-dependent protection against L-arginine-induced acute pancreatitis in rats and this beneficial effect is probably due to its anti-inflammatory and anti-oxidant activities. It could represent a new therapeutic target in the treatment of acute pancreatitis.
INTRODUCTION

Acute pancreatitis (AP) is a serious condition that carries a significant risk of morbidity and mortality. It is characterized by intra-acinar cell activation of digestive enzyme, pancreatic inflammation, cell damage, and a systemic inflammatory response governed by the release of proinflammatory cytokines (Pandol, 2006). The most common causes of acute pancreatitis in humans are alcoholism and gallstones. There are, however, several drugs or drug classes that have been implicated in the development of exocrine pancreatic injury as well. These include azathioprine, estrogens, furosemide, methyldopa, procainamide, sulfonamides, and thiazide diuretics (Vlodov and Tenner, 2001).

Various proinflammatory cytokines, such as interleukin-1β (IL-1β), IL-6 and tumor necrosis factor-α (TNF-α), appear to play a major role in the inflammation associated with acute pancreatitis (Norman, 1998). In addition, adhesion molecules such as intercellular adhesion molecule-1 (ICAM-1) have been reported to be upregulated, promoting the migration of neutrophils into the pancreatic parenchyma, which has been implicated as one of the major factors that promote the worsening of pancreatitis (Zaninovic et al., 2000). At the same time, it has been found that oxidative stress, mediated by short-lived intracellular oxygen free-radical species is one of the mediators of acinar cell and remote organ injury in experimental acute pancreatitis (Rau et al., 2000).

The modulatory role of peroxisome proliferator-activated receptors (PPARs) has been proposed in the inflammatory response of different organs (Chinetti et al., 2003). The PPAR family consists of at least three different isoforms; PPAR-α, PPAR-δ, and PPAR-γ (Mangelsdorf et al., 1995). Peroxisome proliferator-activated receptor-gamma (PPAR-γ) is a member of the nuclear hormone receptor superfamily originally reported to be expressed at high levels in adipose tissue and to play a critical role
in adipocyte differentiation, glucose metabolism and lipid storage (Folch-Puy et al., 2006). PPAR-γ is expressed in both endocrine and exocrine pancreatic cell types (Welters et al., 2004). Its role in pancreas is not restricted to insulin signaling pathways, it has been shown to be imported in cell growth, metabolism (in particular response to altered energy homeostasis), apoptosis and inflammation (Semple et al., 2006). Activation of PPAR-γ receptors leads to anti-inflammatory and antiproliferative effects as well as increased insulin sensitivity, improvement in dyslipidemia and regression of atherosclerosis (Lehrke and Lazar, 2005). PPAR-γ has powerful synthetic ligands, the thiazolidinediones (TZDs), also called glitazones; torglitazone, ciglitazone, pioglitazone and rosiglitazone. TZDs are used to treat type 2 diabetes because of their efficacy in controlling blood glucose secondary to enhancing insulin action through a mechanism (s) that is yet to be completely elucidated (Owens, 2002).

Rosiglitazone, PPAR-γ agonist, is a widely used drug for the treatment of type 2 diabetes mellitus. It increases insulin sensitivity of peripheral tissues. In addition, there is evidence that rosiglitazone has anti-inflammatory effects (Delerive et al., 2001 and Mohanty et al., 2004).

Considerable evidence indicates that PPAR-γ agonists inhibit inflammatory responses during inflammatory diseases (Zhang and Young, 2002). Therefore, this study based on a model of acute pancreatitis in male adult albino rats aimed to determine the anti-inflammatory and antioxidant effects of different doses of rosiglitazone treatment on acute pancreatitis.
MATERIAL AND METHODS

Drugs:

- L-arginine (Sigma, USA), it was supplied as pure powder and was dissolved in sterile saline (0.9%).
- Rosiglitazone (RSG) (Avandia, GalxoSmithKlein), it was supplied as white tablets (each 4 mg) which was crushed, dissolved in distilled water and suspension was given orally to the rats.

Animals and Study Design:
The experiment was conducted on sixty-four male adult albino rats, approximately weighting 200-220 gms each. Animals were housed and fed under suitable environmental conditions. Food and water were allowed ad libitum. The animals were maintained one week before the experiment for acclimatization.

Animal Grouping:
The animals were randomly allocated into 4 groups as follows:

Group I: Control group (n = 8), rats were received sterile saline only intraperitoneally.

Group II: Rosiglitazone group (n = 24), this group was subdivided into three equal groups; rosiglitazone was given to the animals in a single oral dose of 3 mg/kg (group IIa), 10 mg/kg (group IIb), and 50 mg/kg (group IIc).

Group III: Acute pancreatitis (AP) group (n = 8), AP was induced by intraperitoneal injection of L-arginine (L-Arg) as previously described by Pozsar et al. (1997), 250 mg/100 g b.w. 20% L-arginine in sterile saline was given.

Group IV: Rosiglitazone pre-treated group (n = 24), this group was subdivided into three equal groups; rosiglitazone was given as a single oral dose 1 hour prior to induction of AP as follows:
3 mg/kg (group IVa) (Chung et al., 2005), 10 mg/kg (group IVb) (Cuzzocrea et al., 2004), and 50 mg/kg (group IVc) (Ivashchenko et al., 2007).

All animals were sacrificed 24 hours after induction of experimental acute pancreatitis.

**Experimental Parameters**

**Biochemical Assessment**

At the end of experimental period, the animals were anesthetized by ether. Blood samples were obtained from retro-orbital venous plexus using capillary pipette and then sacrificed. Each sample was centrifuged and the serum was separated for determination of amylase (U/L) according to the method of Winn-Deen et al. (1988), malondialdehyde (MDA) (nmol/ml) as a marker of lipid peroxidation following the method described by Draper and Hadley (1991) and catalase (CAT) activity (U/ml) was determined following the method described by Johansson and Borg (1988). Tumor necrosis factor-α (TNF-α) (pg/ml) and interleukin-6 (IL-6) concentration (pg/ml) were determined in serum using the commercially available rat ELISA kits according to the methods of Corti et al. (1992) and Klinik et al. (1998) respectively.

**Histological assessment:**

After the animals were sacrificed, pancreatic tissue samples were collected and fixed in 10% formalin. Subsequently, it was embedded in paraffin blocks, cut and stained with hematoxylin and eosin according to Drury and Walington (1980).

**Statistical Analysis:**

The statistical analysis were carried out using the computer program SPSS (statistical program for social science). All data are expressed as mean ± SD. Differences between groups were compared by
student's t-test with \( P<0.05 \) selected as the level of statistical significance (Goldstone, 1983).

**Results**

**Biochemical Results:**

Table (1) shows no statistically significant \((P>0.05)\) difference between control group (I) and rosiglitazone groups (IIa, b, c) which received single oral dose of the drug (3, 10, 50 mg/kg) respectively regarding serum amylase, MDA and catalase activity.

Table (1): Comparison between control group (I) and rosiglitazone groups (IIa, b, c) as regards serum amylase, MDA and catalase activity.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control (I)</th>
<th>Rosiglitazone group (II)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>IIa (3 mg/kg)</td>
</tr>
<tr>
<td>amylase (U/L)</td>
<td>18.36± 0.79</td>
<td>17.65± 0.81</td>
</tr>
<tr>
<td>MDA (nmol/ml)</td>
<td>2.42± 0.29</td>
<td>2.38± 0.31</td>
</tr>
<tr>
<td>catalase (U/ml)</td>
<td>2.46± 0.16</td>
<td>2.53± 0.14</td>
</tr>
</tbody>
</table>

Data are expressed as mean ± SD \((n = 8\) for each group).

Table (2) shows that administration of L-arginine (group III) induced acute pancreatitis as evidenced by significant increase \((P<0.05)\) in serum amylase level. At the same time, there was significant increase \((P<0.05)\) in serum MDA level and significant decrease \((P<0.05)\) in serum catalase enzyme activity as compared to group I (control group). Pretreatment of group IVa with rosiglitazone in a dose of 3 mg/kg, 1 hour prior to induction of acute pancreatitis, induced insignificant decrease \((P>0.05)\) in serum amylase level while, rosiglitazone pretreatment in doses of 10, 50 mg/kg (group IV b, c) produced significant decrease \((P<0.05)\) in serum amylase as compared to AP group (group III). While,
there was a significant increase (P<0.05) of serum amylase in rosiglitazone pre-treated groups (IVa, b, c) when compared with control group (group I).

The measurement of serum MDA level showed no significant change (P>0.05) in rats of group IVa as compared to AP group (group III). In contrast, MDA was significantly decreased (P<0.05) in rats of groups IVb and IVc compared to AP group. On the other hand, there was significant increase (P<0.05) in serum catalase activity in groups IVb and IVc but there was insignificant change (P>0.05) in its level in group IVa when compared to AP group (group III). Regarding serum MDA level and catalase activity, there was a significant difference (P<0.05) between groups IVa, b, c when compared to control group (group I) (figs 1,2,3).

Table (2): Effect of rosiglitazone pre-treatment in different doses (3, 10, 50 mg/kg, p.o.) on serum amylase, MDA and catalase activity in rats with L-arginine- induced acute pancreatitis

<table>
<thead>
<tr>
<th>Groups</th>
<th>Control (I)</th>
<th>AP (III)</th>
<th>Rosiglitazone pre-treated AP groups (IV)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>amylase (U/L)</td>
<td>18.36± 0.79</td>
<td>863± 15.8*</td>
</tr>
<tr>
<td></td>
<td>MDA (nmol/ml)</td>
<td>2.42± 0.29</td>
<td>10.87± 0.52*</td>
</tr>
<tr>
<td></td>
<td>catalase (U/ml)</td>
<td>3.46± 0.16</td>
<td>0.38± 0.06*</td>
</tr>
</tbody>
</table>

Data are expressed as mean ± SD (n = 8 for each group)

* : Significant change (P < 0.05) compared to control group.
# : Significant change (P < 0.05) compared to acute pancreatitis (AP) group.
Fig. (1): Effect of rosiglitazone pre-treatment in different doses (3,10,50 mg/kg, p.o) on serum amylase level in L-arginine induced pancreatitis in rats.

* : Significant change (P< 0.05) compared to control group.
# : Significant change (P< 0.05) compared to acute pancreatitis (AP) group.
Group I: Control group. Group III: Acute pancreatitis group.
Group IVa,b,c: rosiglitazone pre-treated groups (3,10,50 mg/kg).

Fig. (2): Effect of rosiglitazone pre-treatment in different doses (3,10,50 mg/kg, p.o) on serum MDA level in L-arginine-induced pancreatitis in rats.

* : Significant change (P< 0.05) compared to control group.
# : Significant change (P< 0.05) compared to acute pancreatitis (AP) group.
Group I: Control group. Group III: Acute pancreatitis group.
Group IVa,b,c: rosiglitazone pre-treated groups (3,10,50 mg/kg).
Fig.(3): Effect of rosiglitazone pre-treatment in different doses (3,10,50 mg/kg, p.o) on serum catalase level in L-arginine-induced pancreatitis in rats.

Table (3) shows no statistically significant difference (P>0.05) between control group (I) and rosiglitazone groups (IIa, b, c ) which received single oral dose of the drug (3, 10, 50 mg/kg) respectively regarding serum TNF-α and IL-6 levels.

Table (3): Comparison between control group (I) and rosiglitazone groups (IIa, b, c) as regards TNF-α and IL-6 levels.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control (I)</th>
<th>Rosiglitazone group (II)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>IIA (3 mg/kg )</td>
</tr>
<tr>
<td>TNF-α (pg/ml)</td>
<td>55.55 ± 2.62</td>
<td>53.80 ± 4.80</td>
</tr>
<tr>
<td>IL-6 (pg/ml)</td>
<td>36.4±2.3</td>
<td>37.2±2.9</td>
</tr>
</tbody>
</table>

Data are expressed as mean ± SD  
( n = 8 for each group)
Table (4) shows that L-arginine administration resulted in significant increase (P<0.05) in both TNF-\(\alpha\) and IL-6 serum levels in AP group (group III) compared to control group (group I). Pretreatment with rosiglitazone in different doses (3, 10, 50 mg/kg) in L-arginine-induced-AP (group IV a, b, c) was associated with a significant decrease (P<0.05) in serum level of TNF-\(\alpha\) and IL-6 compared to AP group (group III). Comparing groups IV a, b, c with control group (group I), there was a significant difference (P<0.05) in the same parameters (figs 4, 5).

The protective effect of rosiglitazone against L-arginine induced AP in rats was dose dependent and was more obvious in group IVc (50 mg/kg).

Table (4): Effect of rosiglitazone pre-treatment in different doses (3, 10, 50 mg/kg, p.o.) on serum TNF-\(\alpha\) and IL-6 levels in rats with L-arginine-induced acute pancreatitis.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Parameters</th>
<th>Control (I)</th>
<th>AP (III)</th>
<th>Rosiglitazone pre-treated AP groups (IV)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td>IVa (3mg/kg)</td>
<td>IVb (10mg/kg)</td>
</tr>
<tr>
<td></td>
<td>TNF-(\alpha) (pg/ml)</td>
<td>27.50±2.62</td>
<td>87.72±4.52*</td>
<td>72.56±3.24**</td>
</tr>
<tr>
<td></td>
<td>IL-6 (pg/ml)</td>
<td>31.4±2.3</td>
<td>68.90±3.9*</td>
<td>55.65±4.1**</td>
</tr>
</tbody>
</table>

Data are expressed as mean ± SD (n = 8 for each group)

*: Significant change (P< 0.05) compared to control group.

#: Significant change (P< 0.05) compared to acute pancreatitis (AP) group.
Fig.(4): Effect of rosiglitazone pre-treatment in different doses (3,10,50 mg/kg, p.o) on serum TNF-α level in L-arginine-induced pancreatitis in rats

Fig.(5): Effect of rosiglitazone pre-treatment in different doses (3,10,50 mg/kg, p.o) on serum IL-6 level in L-arginine-induced pancreatitis in rats.

* : Significant change (P< 0.05) compared to control group.
# : Significant change (P< 0.05) compared to acute pancreatitis (AP) group.
Group I: Control group.
Group III: Acute pancreatitis group.
Group IVa,b,c: rosiglitazone pre-treated groups (3,10,50 mg/kg).
Histopathological Results:

Histopathological examination of the sections of pancreatic tissue of the control rats (group I) showed normal pancreatic architecture. It was formed of closely packed acini separated from each other by minimal connective tissue septa containing ducts and blood vessels. The acinar cells were pyramidal in shape with very narrow acinar lumen, their cytoplasm showed an intense basal basophilia and apical acidophilia. The nuclei were basal in position, round in shape. The same histopathological picture were observed in the sections of pancreatic tissue of rats treated with rosiglitazone in different doses (groups IIa,b,c) (fig 6-A,B,C,D). 

The animals treated with L-arginine (group III) displayed obvious pancreatic injury which was characterized by massive edema with inflammatory cellular infiltration. These changes were associated with cytoplasmic vacuoles of most pancreatic acinar cells and apparently congested dilated blood vessels. The structure of the islets of Langerhans and the pancreatic ducts were preserved (fig 7-A,B). While sections of pancreas of rats pre-treated with 3 mg/kg rosiglitazone (group IVa) and sacrificed after 24 hours from induction of acute pancreatitis revealed mild edema. Pancreatic acini showed cytoplasmic vacuoles, congested dilated blood vessels with mild cellular inflammatory infiltration (fig 8). While the sections of pancreas from L-arginine-induced pancreatitis rats pretreated with 10 mg/kg rosiglitazone (group IVb) showed improvement of most of changes including edema, vacuolization with preservation of the normal pancreatic architecture. There was mild congestion of the blood vessels with mild cellular inflammatory infiltration (fig 9). At the same time, it was found that the light microscopic study of group IVc which was pre-treated with 50 mg/kg rosiglitazone showed marked decrease in edema, disappearance of vacuolization and preservation of normal pancreatic acini.(fig 10).
Figure (6): Photomicrograph of pancreas sections stained with hematoxylin-eosin

(A) A section taken from the control rat (group I) (B-D) Sections taken from the rats treated with 3,10,50 mg/kg rosiglitazone (groups IIa, b, c) shows normal pancreatic architecture, acini, blood vessels and normal islets of Langerhans (original magnification X 400).
Figure (7-A,B,C) : Photomicrograph of pancreas sections stained with hematoxylin-eosin
A section taken from the L-arginine-induced acute pancreatitis (group II) showing severe interstitial edema (E), congested dilated vessels (D), vacuolization of acinar cytoplasm (C) and cellular inflammatory infiltration (I) (original magnification X 400).
Figure (8) : Photomicrograph of pancreas sections stained with hematoxylin-eosin
A section taken from rosiglitazone pretreated rats (3 mg/kg) (group IVa) showing edema, congested dilated vessels, vacuolization of acinar cytoplasm and cellular inflammatory infiltration.(original magnification X 400).

Figure (9) : Photomicrograph of pancreas sections stained with hematoxylin-eosin
A section taken from rosiglitazone pretreated rats (10 mg/kg) (group IVb) showing less interstitial edema and less vacuolization of acinar cytoplasm with a few cellular inflammatory infiltration.(original magnification X 400).
DISCUSSION

Several models of acute pancreatitis have been developed in rats for the study of its pathophysiology and its treatment. In this work, intraperitoneal injection of L-arginine was used to induce acute pancreatitis as this model is non-invasive, highly reproducible and produces selective, dose-dependent acinar cell necrosis. Moreover, L-arginine causes severe acute pancreatitis that is manifested by hyperamylasemia and histological findings which is very similar to the human disease (Hegyi et al., 2004).

Our results revealed that administration of single dose of L-arginine (250 mg/100 gm b.w., i.p.) induced acute pancreatitis as evidenced by significant increase in serum level of amylase and MDA with significant decrease in serum catalase enzyme activity when compared to control group. Furthermore, serum levels of TNF-α and IL-6
were significantly increased. Acute pancreatitis was confirmed by the histopathological examination which showed massive edema, cytoplasmic vacuoles of most of pancreatic acinar cells, congested, apparently dilated blood vessels and inflammatory cellular infiltration.

These results are in agreement with that obtained by Takacs et al. (1996), Varga et al. (1997), Czako et al. (2000) and Takacs et al. (2002). They attributed this significant increase of serum amylase and MDA levels and significant decrease in serum catalase enzyme activity with the histopathological changes to generation of nitric oxide, oxygen free radicals and inflammatory mediators that result in the development of acute pancreatitis.

The results in the present work coincides with Hegyi et al. (2004) who reported that serum amylase level is significantly increased after 24 hrs from induction of pancreatitis by L-arginine and begin to decrease from 2 to 7 days, also the histopathological signs of acute pancreatic inflammation become prominent after one day.

Generation of free radicals is an early and perhaps pivotal mechanism in the pathogenesis of arginine-induced acute pancreatitis (Hegyi et al., 2004). Free radicals can act as a molecular trigger of various inflammatory processes leading to increasing production of prostaglandins, thromboxane, leukotriens and trigger the accumulation of neutrophils, their adherence to the capillary wall with occurrence of edema in the pancreas (Schulz et al., 1999).

Czako et al. (1998) demonstrated that the pancreatic MDA level was significantly elevated at 24 hrs, and peaked at 48 hrs after administration of L-arginine. In addition, the endogenous scavengers, superoxide dismutase (SOD) and catalase activities decreased significantly throughout the entire study versus the control.
The current study confirms and extends previous findings (Takacs et al., 1996; Czako et al., 2000) by showing that inflammation of pancreatic tissue caused by L-arginine is accompanied by a significant increase in both serum TNF-α and IL-6 levels. Rakonczay et al. (2003) found that the increased serum TNF-α, IL-6 and IL-1 levels were demonstrated at 24 hrs after the induction of pancreatitis with L-arginine in rat. Moreover, Czako et al. (2000) reported that increase serum level of TNF-α and IL-6 were detected 12 hrs after administration of L-arginine and remained elevated at 48 hrs in treated animals versus control.

Several studies have demonstrated that the use of PPAR-γ ligands inhibits the intensity of the inflammatory response in different processes including colitis (Sanchez-Hidalgo et al., 2005), adjuvant-induced arthritis (Shiojiri et al., 2002), and cerulien-induced pancreatitis (Cuzzocrea et al., 2004). In vitro, the expression of inflammatory mediators such as TNF-α, IL-1β and IL-6 could be inhibited by PPAR-γ ligands (Ricote et al., 1998; Zafiriou et al., 2005) and this effect seems to be dependent on the inhibition of nuclear factor-κB (NF-κB) pathway (Su et al., 1999). These findings have raised the possibility that these agents could be useful for the treatment of the inflammatory disorders.

The mechanisms of TZDs anti-inflammatory effects are not completely deciphered. Ricote et al. (1998) demonstrated that PPAR-γ ligands inhibit macrophage activation. Furthermore, Ivashchenko et al. (2007) concluded that the anti-inflammatory effects of PPAR-γ ligands may be due to the induction of a potent anti-inflammatory mediator or to its antagonism of the pro-inflammatory transcription factors. To date, no direct anti-inflammatory genes have been found to be activated by PPAR-γ in pancreas.
The protective effects of TZDs in acute pancreatitis were evaluated in several animal models. Konturek et al. (2005) investigated the effects of pioglitazone, a PPAR-γ-specific ligand, in a cerulien model of acute pancreatitis. They concluded that pioglitazone administration ameliorates significantly the pancreatic damage associated with the cerulien-induced pancreatitis confirmed by the improvement of pancreas histology, the reduction in plasma lipase activity and the inhibition of IL-1β production and release.

Our results indicate that administration of rosiglitazone in different doses before induction of acute pancreatitis with L-arginine significantly and dose-dependently reduced serum amylase level and inflammatory response in the pancreas reflected in reduced serum level of TNF-α and IL-6 with improvement of histopathological manifestations of acute pancreatitis. There was marked reduction in the extent of inflammatory cellular infiltration, acinar cell vacuolization and edema of the pancreatic tissues specially with the highest dose used in this work.

These results are in agreement with that of Cuzzocrea et al. (2004) who found statistically significantly decreased activity of amylase, lipase and pancreatic inflammation in male mice treated with rosiglitazone in the course of edematous cerulein-induced acute pancreatitis. A study by Folch-Puy et al. (2006) also elicited complete prevention of the inflammatory response in the pancreas by rosiglitazone when given as a single dose before an experimental model of post-endoscopic retrograde cholangio-pancreatography acute pancreatitis (post-ERCP). Their findings was confirmed by decreased plasma lipase activity and histological findings which showed a clear reduction on pancreatic inter-lobular edema and absence of leukocyte cellular infiltration.
In addition, the work of Ivashchenko et al. (2007) revealed that rosiglitazone acts directly through PPAR-γ in acini, duct and islets to suppress markers of the inflammatory response developed during cerulein-induced acute pancreatitis as cell-type-specific knockout of PPAR-γ in epithelial cells of the pancreas removes the anti-inflammatory effects of rosiglitazone on pro-inflammatory cytokine production, pancreatic edema and infiltration of the tissues with the leukocytes.

To our knowledge, there are few reports about the anti-oxidant effect of rosiglitazone. In the present study, we have evaluated the anti-oxidant effect of rosiglitazone in experimental acute pancreatitis, our data indicate that it exhibits an antioxidant effect as evidenced by significant decrease in serum MDA with significant increase in catalase enzyme activity.

Thiazolidinedione (TZD) class exert anti-oxidant and anti-inflammatory effects in humans in vivo. This effect of TZDs was first observed with troglitazone which reduces reactive oxygen species generation by leukocytes and lipid peroxidation (Garg et al., 2000) and has been confirmed with rosiglitazone (Mohanty et al., 2004). Pioglitazone was shown to abolish ROS production in adipocytes (Houstis et al., 2006) whereas rosiglitazone reduced superoxide production in aortas from diabetic mice (Calkin et al., 2005). However, the molecular mechanism by which TZDs attenuate oxidative stress is not clear.

Ghanim et al. (2006) stated that rosiglitazone may exert antioxidant effect through suppression of reactive oxygen species and nicotinamide adenine dinucleotide phosphate reduced oxidase expression. Moreover, Ceolotto et al. (2007) demonstrated that rosiglitazone significantly decreases glucose-induced oxidative stress and suggested
that most of the antioxidative activity of the drug is determined by its ability to activate 5'-AMP-activated protein kinase (AMPK). Their data indicated that the antioxidative property of rosiglitazone is independent of its ability to activate PPAR-γ and is not a "class effect" because the PPAR-γ agonist GW1929 was unable to prevent ROS induction by high glucose. In contrast, Tao et al. (2003) found that treatment with rosiglitazone exerted a significant vascular protective effect in hypercholesterolemic rabbits, most likely by attenuation of oxidative and nitrative stresses as it enhanced PPAR-γ expression, improved endothelium-dependent vasodilatation, suppressed inducible nitric oxide synthase (iNOS) expression, reduced superoxide production, and inhibited nitrotyrosine formation. The same results were reported by Li et al. (2007).

Summarizing, our data provide the evidence that rosiglitazone, a specific PPAR-γ ligand, exerts significant dose-dependent protection against L-arginine-induced acute pancreatitis in rats. This protective effect involves several mechanisms including prevention of inflammation and oxidative stress which may have synergistically contributed to the beneficial actions of rosiglitazone. These observations indicate that the treatment with rosiglitazone may offer a new therapeutic option in the treatment or prophylaxis of acute pancreatitis.

**Acknowledgment:-**

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تأثير دواء الروزيجليتازون على التهاب البنكرياس الحاد المستحدث تجريبياً بواسطة

ل- أرجينين في فئران التجارب البيضاء

أميمة محمد عبد الله و عبير عبد الوهاب شرف الدين
قسم الأدوية والمزاح. قسم الطب الشرعي والسموم الإلكترونية
كلية الطب. جامعة بنها

الملخص العربي

يعتبر مرض التهاب البنكرياس الحاد من الأمراض التي لا يوجد له علاج فعال حتى الآن. وقد وجد أن دواء الروزيجليتازون الذي يستخدم في علاج النوع الثاني من مرض البوول السكري له تأثير مضاد للالتهاب، وعلى ذلك أجري هذا البحث بهدف دراسة تأثيره على التهاب البنكرياس الحاد المستحدث تجريبيا بواسطة L-أرجينين في فئران التجارب البيضاء. وقد تم إجراء هذه الدراسة على 24 فأر ذكر وقد قسمت إلى أربعة مجموعات رئيسية على النحو التالي: المجموعة الأولى (مجموعة ضابطة) وتتكون من ثمانية فئران، المجموعة الثانية وتحتوي على أربعة وعشرين فأراً وقد قسمت إلى ثلاث مجموعات فرعية متساوية وقد استخدمت كمجمعتها ضابطة لدواء الروزيجليتازون وجرعات 50، 100، 300 مجم/كجم عن طريق الفم، المجموعة الثالثة وهي نموذج لالتهاب البنكرياس الحاد المستحدث تجريبياً وذلك عن طريق إعطاء الفئران ل-أرجينين بجرعة 50 مجم/كجم و 100 مجم كجرعة واحدة و ذلك عن طريق الحقن في التجويف البروتوني أما المجموعة الرابعة فقد تم علاجها بدواء الروزيجليتازون بجرعات مختلفة (1، 100 و 300 مجم/كجم) وذلك قبل ساعة من استخدام التهاب البنكرياس الحاد كما سبق في المجموعة الثالثة. وبعد 24 ساعة من استحداث المرض تجريبياً تم تجميع عينات الدم و ذلك لإجراء قياس مستوى إنزيم الأميليز والمالانونمالك و الكاتليا بالإضافة إلى قياس دلالات الالتهاب: الأنترلوكين-٨ و عامل تنكز الورم- ألفا و علاوة على ذلك أجريت دراسة هستولوجية على أنسجة البنكرياس لكل مجموعة من دراسات. وقد أظهرت نتائج الدراسة أن دواء الروزيجليتازون أحدث نقصًا ذو دلالات إحصائية في مستوى إنزيم الأميليز وقد ظهر أيضاً تحسن ملحوظ في أنسجة البنكرياس و بالإضافة إلى ذلك قد لوحظ نقص ذو دلالات إحصائية في دلالات الالتهاب: الأنترلوكين-٨ و عامل تنكز الورم- ألفا و كذلك مستوى المالانونمالك و (أهم نواتج الأكسدة الفوقية للدهون) مع حدوث زيادة ذو دلالات إحصائية في إنزيم الكاتليا (إنزيم مضاد الأكسدة). و مما سبق نستخلص أن دواء الروزيجليتازون له تأثير مضاد للالتهاب والأكسدة معتمد على جرعة الدواء المعطاة في حالات التهاب البنكرياس الحاد.