Al-Azhar Med. J. *Vol. 44(1), January, 2015, 23-40*
DOI : 10.12816/0015697

ROSUVASTATIN: A POTENT STATIN FOR MANAGEMENT OF INTESTINAL ISCHEMIA/REPERFUSION-INDUCED MULTIPLE ORGAN DYFUNCTION SYNDROME

By

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ABSTRACT

Background: Intestinal ischemia/reperfusion (I/R) is one of the factors entailed in multiple organ dysfunction syndrome (MODS) etiology, possibly via disrupting the redox status and/or triggering the inflammatory cascade in intestine, liver and kidney. The management of MODS is becoming more complicated with new medications and new treatment paradigms. **Objective:** The current study elucidated the possible protective mechanisms of rosuvastatin, known for its pleiotropic effects against intestinal I/R insult. **Material and Methods:** Animals in the current study were randomly assigned into three groups: the first group served as the sham group, the second was the positive control one (I/R) receiving saline, while the third presented the rosuvstatin- pretreated group. Oral rosuvastatin (10 mg/kg) was given for 3 days before I/R, which was carried out by clamping the superior mesenteric artery for 30 minutes followed by declamping for 60 minutes. At the end of the reperfusion period, blood samples were collected and the tissue homogenate from intestine, liver and kidney were used to determine the effect of rosuvastatin against intestinal I/R insult. **Results:** In the three organs studied, intestinal I/R elevated lipid peroxide formation and the activities of myeloperoxidase (MPO), proinflammatory cytokines (IL-1B, IL-6) and caspase-3. Also, the insult decreased the activities of both superoxide dismutase (SOD), and glutathione S transferase (GST). Intestinal I/R increased serum levels of alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatinine (Cr), and creatine kinase (CK). **Conclusion:** Rosuvastatin pretreatment protected against intestinal I/R-induced alteration in the previous assessed parameters. Apart from its lipid lowering capacity, our data suggested a therapeutic potential for rosuvastatin in attenuating redox status disturbance, triggered inflammation and modulating immune response as well as anti-apoptotic capacity. Also, the study proved the presence of a direct correlation between the extent of target organ damage with the activation of inflammation and induction of apoptosis in gut I/R model. This documented the interplay of several factors in the MODS model, which pointed to the use of certain agents that can control multiple pathways in this disorder.

Key words: Rosuvastatin, multiple organ dysfunction syndrome, intestinal ischemia/reperfusion

INTRODUCTION

Multiple organ dysfunction syndrome (MODS), the final common end point resulting from uncontrolled systemic inflammation, is a major cause of morbidity and mortality in surgical intensive care units (ICU). It is characterized by progressive failure of two or

more organs remote from the origin of injury **(Marshall, 2001 and Wang & Ma, 2008)**. Multiple etiologies of MODS, such as multiple trauma, acute respiratory distress syndrome and intra-abdominal sepsis, affect the intestine both directly and indirectly. In each case the insult is often insidious in nature **(Mizock, 2009)**. The prevalence of MODS has been very high, affecting one-third of all in-hospital patients and more than 50% of all ICU patients **(Vincent, 2013)**. Statins are a class of compounds that competitively inhibits the enzyme 3-hydroxy-3 methylglutaryl coenzyme A (HMG-CoA) reductase, the first committed step in cholesterol biosynthesis. Statins can be classified into three categories, i.e. naturally derived, semi-synthetic and synthetic **(Manzoni and Rollini, 2002)**. Statins may exert beneficial cardio-vascular effects independent of their lipidmodifying properties. These pleio-tropic properties include improvement of endothelial cell function, modification of inflammatory responses and enhancement of tissue plasminogen activator synthesis **(Liao, 2002)**. Rosuvastatin, a new HMG-CoA reductase inhibitor, has exhibited a more potent affinity for the active site of HMG-CoA reductase than other statins. In addition, rosuvastatin known for its antioxidant, anti-inflammatory, antiapop-totic capacity and immunomodulating effect that is urgently required to attenuate multiple organ injury. Thus the present study elucidated the possible protective mechanisms of rosuvastatin, known for its pleiotropic effects, against intestinal I/R insult.

MATERIAL AND METHODS

Animals: Adult male albino rats, each weighing 250-300 g, obtained from the animal house of National Research Center (Giza, Egypt), were used in this study. Rats were housed at controlled conditions: temperature of 25±2 ?C, humidity 60-70% and light cycles (12 h light/ 12 h dark). Rats were placed in stainless steel cages (40 x 30 x 25 cm) three animals per each cage. Animals were allowed a free access to water and pelleted standard rat chow diet. The animal experiments described later comply with the ethical principles and guidelines for the care and use of laboratory animals adopted by the National Egyptian Community. The animal experimental protocols were approved by Al-Azhar University Committee.

Drugs and Experimental design: Rosuvastatin was supplied by Astrazeneca (Cairo, Egypt). All the chemicals used in this study were of analytical grade and were purchased from Sigma-Aldrich (USA). Rosuvastatin was freshly prepared in 1% tween 80 and given orally using oral feeding tube for 3 days before operation and the dose was (10 mg / Kg) within the range reported in the literature **(Manucha *et al.*, 2011)**.

Animals in the current study were randomly assigned into three experimental groups (six animals per each group), the groups were divided as follows:

Group I: Sham-operated group, in which the animals were subjected to all the surgical manipulation without occlusion of the superior mesenteric artery (SMA). The animals received the vehicle (1 % tween 80).

Group II: Ischemia control group in which rats were given the vehicle (1 %

tween 80).and subjected to clamping of the SMA ischemia for 30 min and then reperfusion was allowed for 60 min.

Group III: Animals in this group mimic the previous group, except that they received rosuvastatin (10 mg/kg) for 3 days before exposure to mesenteric ischemia/ reperfusion.

Induction of mesenteric ischemia/reperfusion (I/R) injury: Ischemia/reperfusion was performed according to modified method for that by **Murata *et al.* (2006)**. Rats were aniesthetized and placed supine, where a midline laparotomy was performed. The SMA was identified, and subjected to warm ischemia by occlusion of the SMA temporarily with a micro-vascular bulldog clamp. This clamping persisted for 30 min. Afterwards, reperfusion was allowed for 1 hour by gently declamping the artery. Abdomen was closed with two small clamps immediately after the termination of the 30 min of ischemia. At the end of the reperfusion period, blood samples were collected then the animals were sacrificed.

Blood and tissue sampling: The collected blood was centrifuged to separate serum which was used for the estimation of alanine aminotransferase (ALT), aspartate aminotransferase (AST), creatinine (Cr) and creatine kinase (CK). The organs (liver, kidney, and intestine) were harvested for each rat to be weighed separately and then homogenized in ice-cold saline to obtain 20% homogenate. The homogenates were centrifuged and the supernatants were used for oxidative stress markers (Redox parameters), inflammatory markers and apoptotic markers.

Measurement of Redox parameters: Lipid peroxidation (nmol/g tissue) was determined according to the method of **Mihara and Uchiyama (1978)**, where the thiobarbituric acid reactive substances (TBARS), measured as malondialdehyde (MDA), was used as an index of lipid peroxides. The cytosolic superoxide dismutase (SOD) activity was measured using the pyrogallol method according to **Marklund and Marklund (1974)**. The SOD activity was expressed as U/mg protein. The determination of glutathione S transferase (GST) activity was based on the method of **Habig *et al.* (1974)** and **Chen *et al.* (1993)**. The data were expressed as nmol/min/mg protein.

Measurement of inflammatory markers: The measured parameters were: myeloperoxidase (MPO), interleukin-1β (IL-1β) and interleukin-6 (IL-6) in the three organs (liver, kidney and intestine) of all groups of rats. Myeloperoxidase activity was determined using Rat ELISA kit, as suggested by the manufacturer (Ray Biotech, Inc., Norcross, USA), IL-1β production was determined using Rat ELISA kit (Ray Biotech, Inc., Norcross, USA), while the production of IL-6 was performed using Rat ELISA kit, (KOMA Biotech Inc, Seoul, Korea).

Measurement of Caspase-3 activity: Caspase-3 plays a central role in the execution phase of cell apoptosis (apoptotic marker). Assay of caspase-3 was carried out according to the manufacturer's instructions (Cusabio

Biotech Co., WUHAN, Hubei Province, P.R. China).

Liver function tests: The serum levels of ALT and AST were determined by standard commercial biochemical kits according to the method described by **Reitman and Frankel (1957)**.

Kidney function test: The serum level of Creatinine (Cr) was determined using a standard commercial kit according to the method described by **Fabiny and Ertingshausen (1971).**

Intestinal function test: Creatine kinase (CK) activity was determined by using Stanbio CK-NAC dignostic kit (San Antonio, TX, USA) according to a modification of the kinetic method of **Rosalki (1967)** and **Szasz (1976)** technique.

Statistical analysis: Data were expressed as mean ± S.E.M. Statistical comparisons between means were carried out using one-way analysis of variance (ANOVA), followed by Tukey's test. Correlation was calculated using linear regression analysis. The statistical significance of difference was considered at $P \leq 0.05$.

RESULTS

Effect of rosuvastatin on redox parameters in tissues of multiple organs:

Exposure of animals to SMA occlusion exhibited increment in their MDA content that was evaluated in segments excised from liver, kidney and peaking at intestine as comparable to baseline values for each one. The pre-administration of Rosuvastatin before the intestinal insult, diminished lipid peroxides in the three organs compared to I/R group (Figure 1). The SOD activity decreased significantly following I/R in the three organs in comparison to the sham control group. However, pre-administration of rosuvastatin before I/R insult, succeeded to restore SOD activity (Figure 2). Regarding liver, animals subjected to I/R showed a precipitous decrease in the GST activity as regarded to sham - operated animals. A similar pattern was detected in both the kidney and the intestine with significant lower GST levels. On the other hand, giving rosuvastatin, before the operation increased GST activity as compared to the sham-operated group (Figure 3).

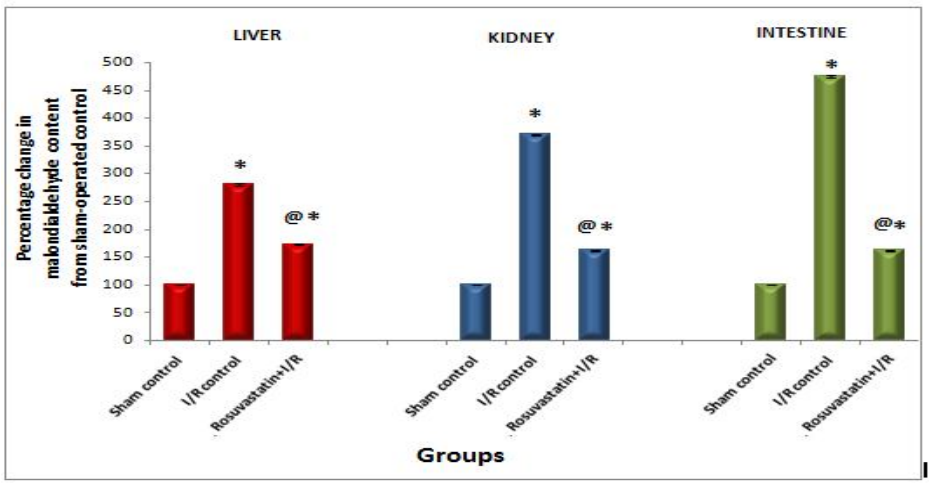

Figure (1): Effect of rosuvastatin on MDA content (nmol/g tissue) in tissues of different organs.

* Significantly different from sham group and @ significantly different from the corresponding I/R control group, ($p<0.05$).

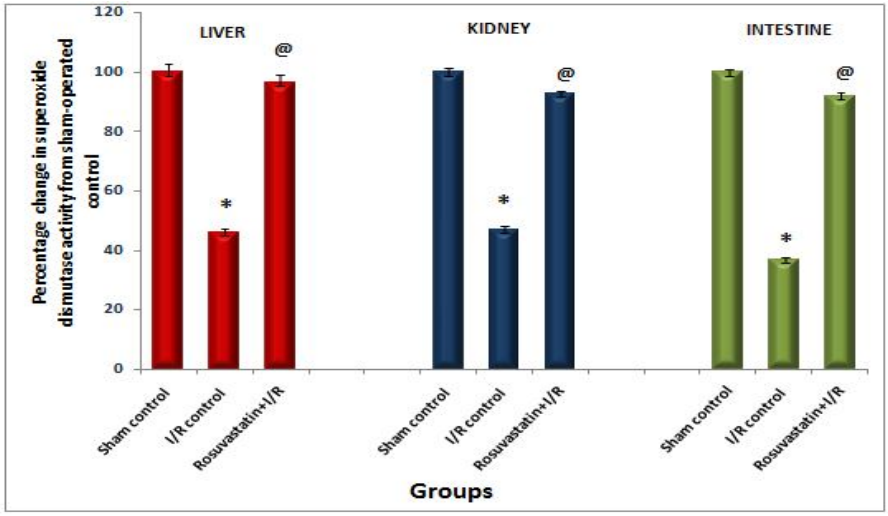

Figure (2): Effect of rosuvastatin on SOD activity (U/mg protein) in tissues of different organs.

* Significantly different from sham group and @ significantly different from the corresponding I/R control group, ($p<0.05$).

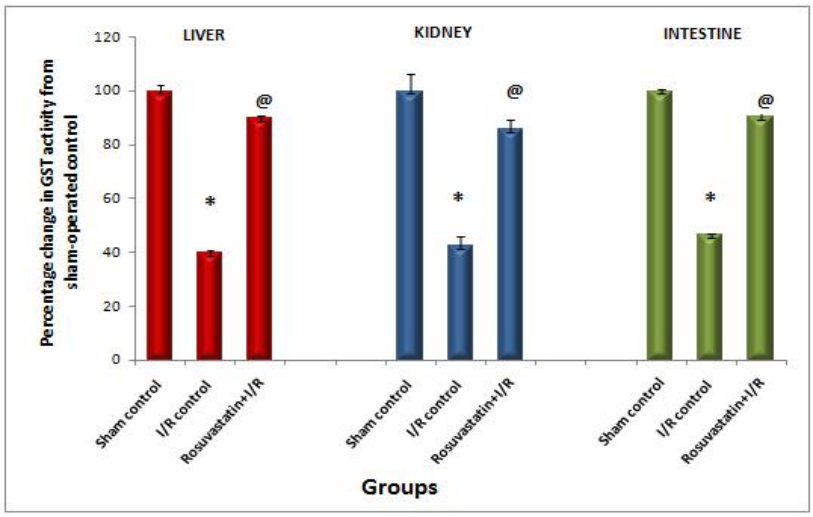

Figure (3): Effect of rosuvastatin on GST activity (nmol/min/mg protein) in tissues of different organs.

* Significantly different from sham group and @ significantly different from the corresponding I/R control group, ($p<0.05$).

Effect of rosuvastatin on inflammatory markers in tissues of multiple organs:

Ischemia/reoxygenation caused a significant increase in MPO activity in the liver in comparison to non-operated animals, an effect that was completely normalized by the pre-adminstraion of rosuvastatin. The same pattern was mirrored in the other two organs, viz., kidney and intestine (Figure 4). The systemic organs expressed arose in IL-1β production in correlation with sham-manipulated group as depicted in Figure (5). The I/R insult was prohibited by the

pre-adminstration of rosuvastatin which leveled off the IL-1β values in the corresponding organs as compared to I/R non-treated groups. Compared with findings in the sham group, the tissue levels of IL-6 were significantly increased in I/R group. This increase significantly decreased in rosuvastatin treated group compared with I/R group (Figure 6).

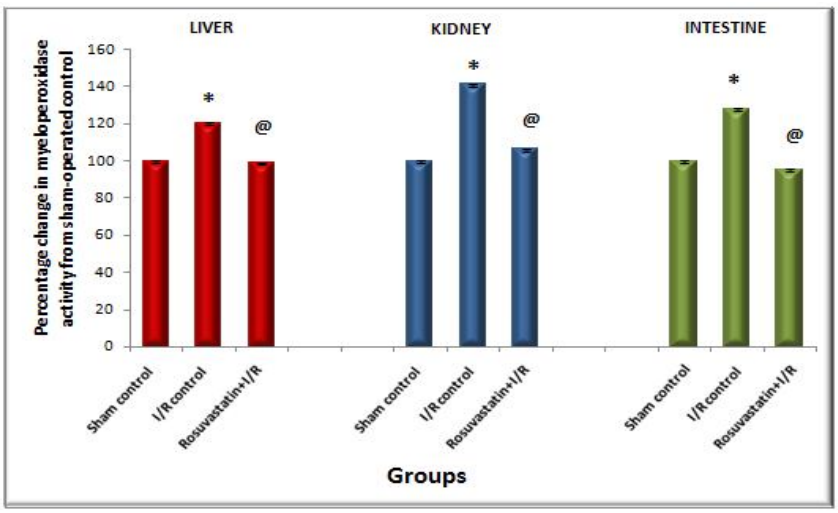

Figure (4): Effect of rosuvastatin on MPO activity (ng/100mg) in tissues of different organs.

* Significantly different from sham group and @ significantly different from the corresponding I/R control group, ($p<0.05$).

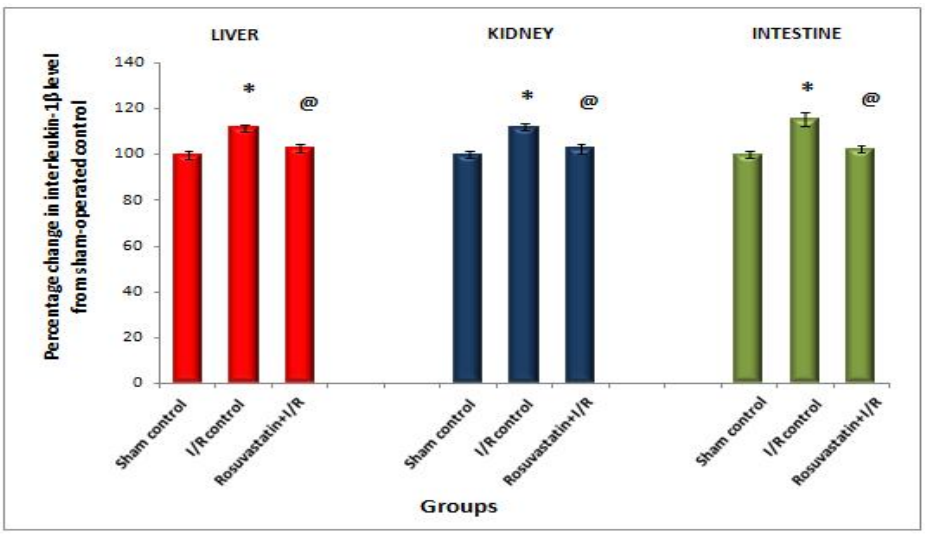

Figure (5): Effect of rosuvastatin on IL-1β levels (pg/ml) in tissues of different organs.

* Significantly different from sham group and @ significantly different from the corresponding I/R control group, ($p<0.05$).

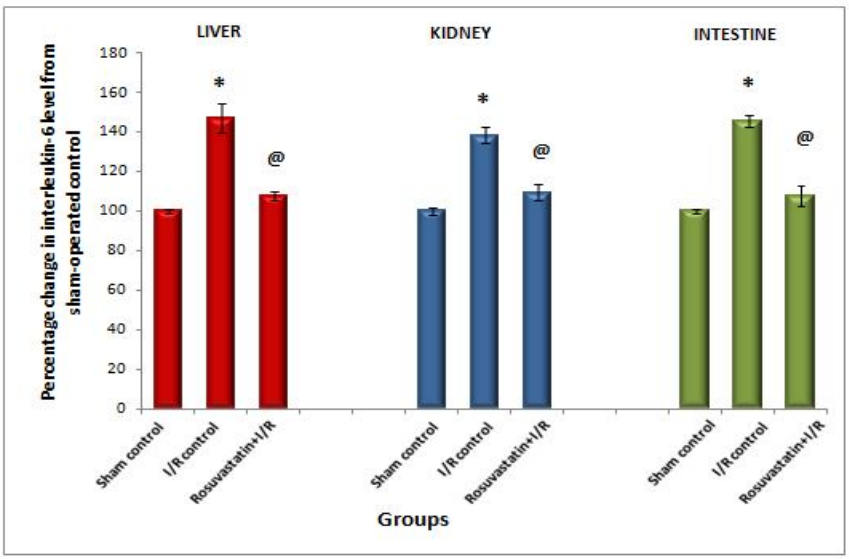

Figure (6): Effect of rosuvastatin on IL-6 levels (pg/ml) in tissues of different organs.

* Significantly different from sham group and @ significantly different from the corresponding I/R control group, ($p<0.05$).

Effect of rosuvastatin on Caspase-3 activity in tissues of multiple organs:

The intestine showed high susceptibility to the apoptotic paradigm offered by the studied model indicated by increased caspase-3 enzyme activity. Kidney and liver also revealed an augmented activity, but to a lesser extent. However, *rosuvastatin* pre-administration antagonized the effect of I/R and decreased caspase-3 level in the liver, kidney and intestine as shown in Figure (7).

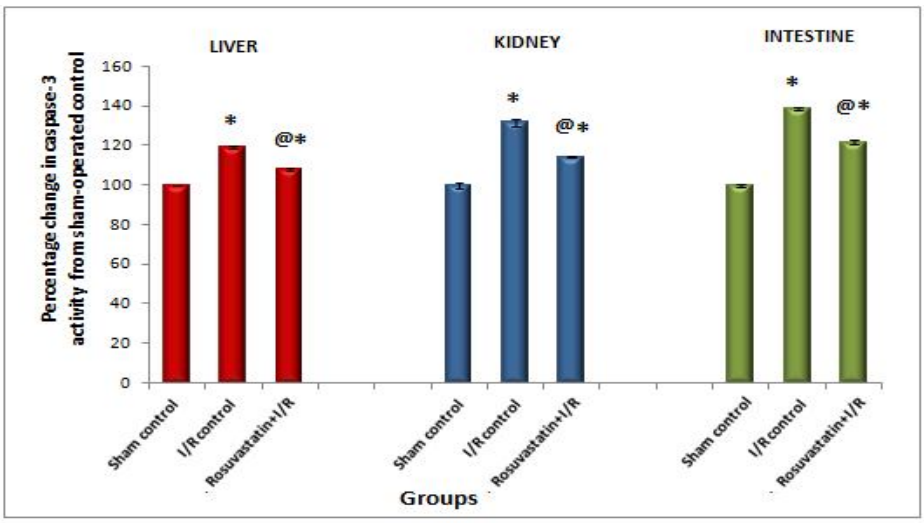

Figure (7): Effect of rosuvastatin on caspase-3 activity (ng/ml) in tissues of different organs.

* Significantly different from sham group and @ significantly different from the corresponding I/R control group, ($p<0.05$).

Effect of rosuvastatin on functions of multiple organs:

Serum ALT and AST measurements showed a vast increase in their levels mounted to more than two folds in respect to sham control samples. A similar pattern was detected in Cr and CK levels. I/R-mediated alteration in serum levels of ALT, AST, Cr and CK significantly hindered by pretreatment regimen used with rosuvastatin (Table 1).

Table (1): Effect of rosuvastatin (10 mg/kg) on alanine amino transferase (ALT), aspartate amino transferase (AST), creatinine (Cr) and creatine kinase (CK) activities in serum of rats after intestinal ischemia/reperfusion

Groups \ Enzymes	ALT (U/L)	AST (U/L)	Cr (mg/dl)	CK (U/L)
Sham control	22.5 ± 1.05	24 ± 1.6	0.5 ± 0.037	83.17 ± 2.57
I/R control	52.9 ± 1.9 *	64.37 ± 1.9 *	1.77 ± 0.049 *	247.67 ± 2.64 *
Rosuvastatin+ I/R	28.3 ± 1.7 @	30.5 ± 1.7 @	1.06 ± 0.035 @*	128 ± 2.62 @ *

* Significantly different from sham group and @ significantly different from the corresponding I/R control group, ($p<0.05$).

Correlation coefficient (r) between hepatic caspase-3, MDA, ALT and AST with IL-1β, IL-6, MPO, MDA and caspase-3 in rats after intestinal ischemia/ reperfusion:

There was a positive correlation between the apoptosis marker caspase-3 and the immunomodulatory cytokines (IL-1β, IL-6), MPO and MDA. This association was also reflected on MDA, which was positively correlated with IL-1β, IL-6 and MPO. The hepatic parameters, ALT and AST correlated also significantly with IL-1β, IL-6, MPO, caspase-3 and MDA (Table 2).

Table (2): Correlation coefficient (r) between hepatic caspase-3, MDA, ALT and AST with IL-1β, IL-6, MPO, MDA and caspase-3 in rats after intestinal ischemia/ reperfusion.

Parameters	IL-1β	IL-6	MPO	MDA	Caspase-3
Caspase-3	0.8893 P<0.0001	0.9 P<0.0001	0.5909 P<0.01	0.8571 P<0.0001	-
MDA	0.9143 P<0.0001	0.8929 P<0.0001	0.6338 P<0.05	-	0.8571 P<0.0001
ALT	0.8756 P<0.0001	0.7413 P<0.001	0.5081 P<0.05	0.8791 P<0.0001	0.7520 P<0.001
AST	0.9123 P<0.0001	0.8408 P<0.0001	0.5363 P<0.05	0.8801 P<0.0001	0.8229 P<0.0001

Correlation was carried out in untreated and treated ischemic/ reperfused rats using linear regression analysis.

Correlation coefficient (r) between renal caspase-3, MDA and creatinine (Cr) with IL-1β, IL-6, MPO, MDA and caspase-3 in rats after intestinal ischemia/ reperfusion:

Caspase-3 and Cr were found to correlate positively with IL-1β, IL-6, MPO, MDA and Cr. furthermore, renal lipid peroxidation was found to correlate with MPO, IL-1β, IL-6, and apoptosis (caspase-3) (Table 3).

Table (3): Correlation coefficient (r) between renal caspase-3, MDA and creatinine (Cr) with IL-1β, IL-6, MPO, MDA and caspase-3 in rats after intestinal ischemia/ reperfusion**.**

Parameters	IL-1β	IL-6	MPO	MDA	Caspase-3
Caspase-3	**0.9246 P<0.0001**	**0.9102 P<0.0001**	**0.7525 P<0.001**	**0.9012 P<0.0001**	**-**
MDA	**0.9212 P<0.0001**	**0.8996 P<0.0001**	**0.6792 P<0.01**	**-**	**0.9012 P<0.0001**
Cr	**0.9023 P<0.0001**	**0.9166 P<0.0001**	**0.6880 P<0.01**	**0.8875 P<0.0001**	**0.9468 P<0.0001**

Correlation was carried out in untreated and treated ischemic/ reperfused rats using linear regression analysis

Correlation coefficient (r) between intestinal caspase-3, MDA and creatine kinase (CK) with IL-1β, IL-6, MPO, MDA and caspase-3 in rats after intestinal ischemia/ reperfusion:

Caspase-3 and CK were positively correlated with IL-1β, IL-6, MPO and MDA. Moreover, MDA also correlated with IL-1β, IL-6, MPO and caspase-3 (Table 4).

Table (4): Correlation coefficient (r) between intestinal caspase-3, MDA and creatine kinase (CK) with IL-1β, IL-6, MPO, MDA and caspase-3 in rats after intestinal ischemia/ reperfusion.

Parameters	IL-1β	IL-6	MPO	MDA	Caspase-3
Caspase-3	**0.8766 P<0.0001**	**0.8676 P<0.0001**	**0.6188 P<0.01**	**0.8523 P<0.0001**	**-**
MDA	**0.9169 P<0.0001**	**0.8776 P<0.0001**	**0.4821 P<0.05**	**-**	**0.8523 P<0.0001**
CK	**0.9036 P<0.0001**	**0.9429 P<0.0001**	**0.5658 P<0.05**	**0.8972 P<0.0001**	**0.8980 P<0.001**

Correlation was carried out in untreated and treated ischemic/ reperfused rats using linear regression analysis.

DISCUSSION

Intestinal ischemia/reperfusion (I/R), resulting in high mortality rate up to 70%, has been linked to peripheral organ damage, systemic inflammatory response syndrome (SIRS), as well as multiple organ dysfunction syndrome (MODS) **(Fink & Delude, 2005 and Fukatsu *et al.*, 2006)**. In MODS, oxidative stress plays a role in disease initiation and progression **(Shi *et al.*, 2005)**, where increased formation and release of reactive oxygen species (ROS) and free radicals attack the cell macromolecules and overwhelm the defense system. These insults result in loss of epithelial cell integrity and impair mucsal recovery **(Biffl and Moore, 1996)**.

As shown in the current study, the intestinal I/R insult reduced SOD level in the three studied organs. Hence, leaving them vulnerable to the increased shower of ROS, where the exhausted SOD permits an unopposed elevation of the superoxide anion ($O_2^{\cdot-}$). This finding collaborates with previous findings **(Barut *et al.*, 2007 and Sizlan *et al.*, 2009).** Also, I/R decreased the GST activity, a finding that goes in line with **Liu *et al.* (2005).** Malonaldehyde (MDA) concentration is directly proportional to the cell damage caused by free radicals. As a result to the I/R-related ROS and free radicals formation, lipid peroxidation level significantly incrased in intestinal, hepatic and renal tissues in the present study, a finding that was documented in previous works **(Wang *et al.*, 2008 and Sun *et al.*, 2012)**. Administration of rosuvastatin offered a significant protection against oxidative stress caused by intestinal I/R via increasing the SOD and GST activities in the three organs measured. The anti-oxidant effect of *rosuvastatin* is supported by the study of **Awad and Kamel (2010**) in hepatic I/R model and by **Ansari *et al.* (2012)** in an obesity-induced cardiac oxidative stress model in rats. The positive effect of *rosuvastatin* is attributed to its ability to induce the expression and activitation of Cu/Zn-SOD **(Verreth *et al.*, 2007 and Colucci *et al.*, 2013).** In addition, *rosuvastatin* reduced activation of the transcription factor nuclear factor-kappaB (NF-κB) that controls oxidant stress -sensitive and -infalmmatory signaling pathways **(Dichtl *et al.*, 2003 and Maack *et al.*, 2003)**. Furthermore, rosuvasatin reduces oxidative damage by controlling nitric oxide (NO) production **(Stoll *et al.*, 2004)**. Regarding the effect of *rosuvastatin* on mesenteric I/R-mediated lipid peroxidation, a significant decrease in the elevated MDA level was observed. In support to these results, pretreatment with rosuvastatin minimized lipid peroxidation repeatedly in murine hepatic and renal tissues of different animal models **(Awad and Kamel, 2010; Abe *et al.*, 2011 and Ansari *et al.*, 2012)**. The present investigation was undertaken to delineate the role of apoptosis in MODS after intestinal I/R, a role that was indicated by the increased caspase-3 activity (main executor of apoptosis) in the affected organs. **Zhang *et al.* (2008**) **and Cui *et al.* (2010)** showed increased caspase-3 expression and presence of apoptotic cell in the lung, kidney, liver and intestine following hypoxia/reoxyge-nation. Rosuvastatin significantly delved the I/R-induced elevation the intestinal, renal and hepatic caspase-3 activity. The pretreatment with rosuvastatin protects

against apoptosis **(Enomoto *et al.*, 2009 and Sharma *et al.*, 2011)** which may occur due to inhibition of Ras by *rosuvastatin* **(Cormack-Aboud *et al.*, 2009).** Generally, myeloperoxidase is considered as a surrogate marker of inflammation and neutrophils infiltration in several tissues, including the intestine and is also considered an index for oxidative damage **(Huang *et al.*, 2008)**. The increased MPO activity was illustrated in the examined tissues resulted as a consequence to I/R insult **(Wang *et al.*, 2008 and Yao *et al.*, 2009)**. Rosuvastatin provides protection from the development of systemic inflammation and multiorgan dysfunction caused by neutrophil infiltration. Previous study corroborated with the present anti-inflammatory effect of rosuvastatin **(Naito *et al.*, 2006 and Monetti *et al*. 2007)**. The reduction in MPO may also be referred to the rosuvastatin-induced inhibition of pro-inflammatory cytokines, such as TNF-α, IL-1β and IL-6 in the light of its immunomodulatory effect **(Link *et al.*, 2006)**. This was documented in this work by the direct significant correlation between these parameters and MPO. Intestinal epithelial cells were recognized as a source of inflammatory cytokines and chemokines. IL-1β is one of the crucial cytokines that has pathological implication for the development of IR injury by inducing the cascade activation of immune cells **(Sanchez-Hidalgo *et al*., 2007 and Zwolinska-Wcislo *et al*., 2011)**. IL-6 also is involved in most types of inflammation and appears to amplify and perpetuate the ongoing inflammatory response, which have been reported to be involved in the pathophysiology of gut IR episode (**Horton and Walker, 1993)**. This may explain the present increments in both IL-1β and IL-6 levels offered by this model in each of the studied organs. The current findings are also consistent with **(**../../Eman/AppData/Local/Temp/Rar $DI00.638/discussion.docx - _ENREF _ 91 **Nezu *et al.*, 2008)**. Also, the role of cytokines, such as IL-1β and IL-6 in ischemia/reperfusion pathophysiology, had been implicated by authors previously as mediators for inflammation and apoptosis **(Tian *et al.*, 2002)**. This fact points to the correlation between increased caspase-3 activity and the level of IL-1β and 6 as proven in the current study. Administration of rosuvastatin controlled the intestinal I/R outcome. The inhibitory effect of rosuvastatin on inflammatory cytokines' levels was documented in different models of inflammation **(Link *et al.*, 2006; Kim *et al.*, 2007; Lazzerini *et al.*, 2013)**. As a more mechanistic insight, rosuvastatin by combating ROS can inhibit NF-κB, a transcription factor that is essential for the transcription of several inflammatory molecules. Moreover, statins-mediated inhibition of NF-κB resulted in inactivation of iNOS that produces the deleterious NO type (Huang et al., 2003). The effect of rosuvastatin demonstrates a direct correlation between the inhibition of apoptosis after mesenteric I/R and the reduction of the subsequent inflammation, which apparently shows apoptosis to be critical for the induction of inflammation in the studied model. Liver is the first distant affected organ by gut I/R, other than the gut per se **(Towfigh *et al.*, 2000)**, the degenerative insult increases membrane permeability leading to leakage of enzymes (ALT and AST) from hepatocytes into the circulation **(Zimmerman *et***

***al.*, 1990)**. Similar results were reported by another investigators using gut I/R **(Wang *et al.*, 2008; Yao *et al.*, 2009 and Zhao *et al.*, 2010)**. Rosuvastatin protective action could be mediated via its anti-oxidant, anti-inflammatory, immuno-modulatory and anti-apoptotic effect. The effect of rosuvastatin may also be mediated through increasing NO availability, which is capable of attenuating the ALT leakage from hepatocytes that occurs at 1 hour after reperfusion (**Horie *et al*., 1997).**

Mesenteric I/R have been shown to impair also renal tubular function that was reflected herein by the increased creatinine level above the normal one. The obtained result is in harmony with previous recorded data by **Zhang *et al.* (2008) and Sun *et al.* (2012).** The usage of rosuvastatin produced a subtle, yet significant, renal functional refinement as indicated by decreasing the serum creatinine level. Statins have been reported to ameliorate golmerular injury and preserve renal function **(Gianella *et al*., 2007 and Zhou *et al.*, 2008)**. ThIs, may be acquired by its anti-apoptotic effect, as proven in this work, through decreasing caspase-3 and MPO activities, which participate in maintaining the normal renal function. The intestine is considered extremely sensitive to I/R injury and the level of creatine kinase (CK) provided a valuable information about the related intestinal tissue injury **(Caglayan *et al*., 2002)**. In the current investigation, alike previous findings **(Caglayan *et al.*, 2002 and Sirmali *et al.*, 2007)**, increments in CK level was detected in I/R group comparable to the non-operated one. This was paralleled herein with the induction of apoptosis and lipid peroxidation that favors organ dysfunction by impairing several physiological mechanisms **(Del Rio *et al.*, 2005)**. Rosuvastatin restored the lost intestinal function which based on its anti-oxidant property combined with its prosurvival effect. Also, rosuvastatin resulted in a significant diminution in the immunomodultory cytokines (IL-1β and IL-6) and the neutrophil infiltration. These results comply with those previously observed in various studies **(Di Napoli *et al.*, 2005; Farnier *et al.*, 2009 and Erbs *et al.*, 2011)**.

In conclusion: Although the prevailing concept has linked mortality in MODS with the unbridled hyper-inflammatory cytokine-mediated response, yet the failure of many clinical trials to treat MODS by controlling this cytokine response requires a 'rethink'. This disease arises from interplay between several molecular paradigms involving apoptosis, inflammation, immunomodulatory imbalance, as well as increased oxidative stress. As the usage of rosuvastatin leads to rectification of the collaborated factors, hence the current study adds new therapeutic approach for the management of MODS in this stringent animal model.

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خلفيه البحث: إن نقص الدم و إعاده تدفقه الى الأمعاء الدقيقة هو أحد العوامل المتضمنة فى حدوث متلازمة الخلل الوظيفى متعدد الأعضاء. و من الممكن أن يكون ذلك عن طريق إحداث خلل فى منظومة الأكسدة والإختزال والتهاب فى بعض الأعضاء مثل الأمعاء والكبد والكلى. وعلى الرغم من ذلك فإن الفهم الكامل لأسباب حدوث هذه المتلازمة و علاجها أصبح أكثر تعقيدا مع الأدوية الجديدة ونماذج علاجية جديدة.

الهدف من البحث: أوضحت الدراسة الحالية آليات الحماية الممكنة لعقار الروسفاستاتين والذى يمتلك تأثيرات طبية عديدة تجعله ذو تأثير هام فى تخفيف متلازمة الخلل الوظيفى فى عدة أعضاء.

طرق و مواد البحث: قسمت الجرذان المستخدمة فى هذا البحث إلى ثلاث مجموعات: المجموعة الأولى وهى المجموعة الضابطة ، والمجموعة الثانية تم تخديرها و ربط الشريان المساريقى العلوى مع قطع الإمداد عن الأمعاء لمدة نصف ساعة ثم إعادة التروية لمدة ساعة بدون علاج، والمجموعة الثالثة هى التى تم علاجها مسبقا عن طريق الفم بعقار الروسفاستاتين (10 ملغ / كلغ) لمدة 3 أيام قبل ربط الشريان المساريقى العلوى مع قطع الإمداد عن الأمعاء لمدة نصف ساعة ثم إعادة التروية لمدة ساعة، ثم تم ذبح الجرذان وإستخراج الأمعاء و الكبد و الكلى لتقييم الضرر الذى لحق بهم 0كما تم سحب عينات دم لإجراء الفحوصات المتعلقة بوظائف الأعضاء المذكورة.

النتائج: و قد كانت النتائج الرئيسية للدراسة كالتالى:

1- نتج عن نقص الدم بالأمعاء وإعادة تدفقه تغيير فى التوازن الطبيعى بين جهدى الأكسدة والإختزال فى عدة أعضاء ممثلة فى زيادة معدلات الدهون فوق المؤكسده مع نقص فى نشاط إنزيمى السوبر أوكسيد ديسميوتيز وجلوتاثيون ترانسفيراز 0 وقد كان لدواء روسفاستاتين تأثيرا واقيا لحماية الأمعاء و الكبد والكلى بسبب خصائصه المضادة للأكسدة.

2- تم التغلب على الإرتفاع فى وسائط الإلتهاب بواسطة الروسفاستاتين حيث أدى العلاج المسبق إلى الحد من نشاط الميالبير أوكسيديز و تقليل كل من مستوى إنترليوكين واحد- بيتا وإنترليوكين- ستة فى مصل الدم.

3- أدى إستخدام عقار الروسفاستاتين إلى تقليل موت الخلايا المبرمج فى كل من الأمعاء والكبد والكلى نتيجة لنقص الدم بالأمعاء كما يتضح من نقص نشاط إنزيم الكاسباس-3.

4- روسفاستاتين فعال فى حماية الأعضاء المختلفة ضد الخلل الوظيفى المتعدد المحدث فى هذه الدراسة كما يتضح من إستعادة المستويات الطبيعية لكل من إنزيم الألانين امينوترانسفيرز وانزيم أسبرتات امينوترانسفيرز، وكذلك الكرياتينين والكرياتين كيناز.

5- هناك علاقة فى هذا النموذج بين درجة التلف النسيجى العضوى وحدوث الإلتهاب والموت المبرمج للخلايا.

الاستنتاج: مما سبق يتضح أن هناك مشاركة فعالة للعديد من العوامل فى حدوث متلازمة الخلل الوظيفى متعدد الأعضاء مما يتطلب إستخدام طرق علاجية للسيطرة على هذه العوامل مجتمعة. وقد كان لإستخدام عقار الروسفاستاتين تأثيراً هاماً فى تخفيف متلازمة الخلل الوظيفى فى عدة أعضاء.