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Morphological Investigation of Protective Effects of CDP-Choline on Liver and Small Intestine in an Experimental Sepsis Model in Rats

Investigación Morfológica de los Efectos Protectores de la CDP-Colina en el Hígado y el Intestino Delgado en un Modelo Experimental de Sepsis en Ratas

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SUMMARY: Fifty male Wistar albino rats were divided into 5 groups; Group 1 as a sham group. Group 2 as a control group, Group 3 as 100 mg/kg CDP-choline administered group, Group as 200 mg/kg CDP-choline administered group, and Group 5 as sepsis group. The sepsis model was performed by ligating and perforating the caecum of rats. Liver and small intestine tissues were assessed either histologically or quantitatively and qualitatively. There was a significant difference between the sepsis and CDP-choline groups for liver and intestinal damage evaluated in tissue samples. ($p < 0.001$). CDP-choline treatment partially improved dose-dependent the clinical parameters of sepsis and septic shock, reversed micro-anatomical damage caused by sepsis.

KEY WORDS: CDP-choline; Liver; Small intestine; Tissue damage.

INTRODUCTION

CDP choline is a mononucleotide that occurs as an endogenous intermediate product during phosphatidylcholine synthesis, one of the cell membrane phospholipids. It plays a key role in the speed-limiting step during the chemical process called "Kennedy Pathway". Following exogenous implementation, CDP-choline is degraded to cytidine and choline by the phosphodiesterase in the cell membrane (Weiss, 1995).

Sepsis is defined as a systemic inflammatory response syndrome (SIRS) arise due to an infectious agent that is proven by blood culture. Although the mortality rate due to sepsis has been decreasing dramatically, sepsis is still one of the most important causes of mortality among hospitalized patients (Dombrovskiy *et al*., 2005). Sepsis is the main cause of hospitalization in almost 11 % of patients hospitalized in intensive care units, especially in western societies. In developed countries, sepsis affects 3 to 10 out of 100 people each year, with mortality rates up to 35 % (Perner *et al*., 2016). The widespread use of chemotherapy, immunosuppression due to various causes, resistant nosocomial infections and rising frequency of bedside invasive procedures increase the risk of sepsis (Dombrovskiy *et al*., 2005). In severe Sepsis clinically characterized by multiple organ failure; functional failures may develop in all organs, especially in the lungs and kidneys (Bosmann & Ward 2013) Mortality rates increase up to 80 % of cases for severe sepsis with more than two organ system involvement (Bosmann & Ward 2013).

However, there is a consensus that cecal ligation and puncture (CLP) is one of the suitable experimental model of sepsis in rats (Remick *et al*., 2000). Although there are many studies performed to assess the neuroprotective, cardio protective and anti-ischemic effects of CDP-choline using experimental sepsis and septic shock models, there are few studies evaluating the effects of this molecule on macro- and micro-anatomical changes and cellular damage in the

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liver and small intestine during sepsis. This study aimed to investigate the potential effects of CDP-choline on adverse morphological changes in the liver and small intestine in sepsis by using an experimental intraperitoneal sepsis model with the CLP technique in rats.

MATERIAL AND METHOD

This study was approved by the Bursa Uludag University Animal Experiments Ethics Committee (Date: 16.09.2014, Number: 2014-13/01) and supported by Bursa Uludag University Research Fund.

Fifty male Wistar Albino rats 100-120 days old and weighing 300-450 grams were used in the study. All rats were kept in the experimental laboratory of the Department of Anatomy in Bursa Uludag University Experimental Animals Production and Research Center for one week before the experiment to ensure their acclimatization, where the temperature (25 ± 2 °C) and relative humidity (32 ± 7 %) were stable. Each rat was housed in a standard single cage under 12 hours of light and dark cycle environment. Free feed and water intake were provided up to 1 day before surgery and just one pellet of food on the last preoperative day. All rats were fasted for 12 hours to ensure to have similar faecal contents and to avoid aspiration after anaesthesia, but their access to water was allowed.

Rats were divided into 5 groups: Group 1 was the sham laparotomy group (n = 10). In this group, solely laparotomy was performed without CLP but caecum was gently palpated to force the faecal content to move into distal segments of the caecum. Group 2 was the control group (n = 10). The rats included in control group were just sacrificed without any additional procedure and tissue samples were taken. Group 3 was the CLP + 100 mg/kg of CDP-choline group (n = 10). All rats in this group were preoperatively administered intraperitoneal 100 mg/kg of CDP-choline before CLP. Group 4 was the CLP + 200 mg/kg of CDP-choline group (n = 10). The group was preoperatively administered intraperitoneal 200 mg/kg of CDP-choline before CLP. Group 5 was the only CLP (sepsis) group (n = 10). All rats were sacrificed after 8 hours of experimental procedures, and tissue samples were taken.

Monitorization of the vital parameters. During the experiment, vital parameters of the rats were recorded by using tail and rectum for measurements with non-invasive method. Graphs for vital values were used to evaluate sepsis findings. Heart rate (HR), mean arterial blood pressure (MAP) and rectal temperature (RT) were measured as the main follow-up parameters. In addition, the rats were monitored every 30 minutes in the first 2 hours postoperatively and at least every 2 hours in the follow-up period.

Administration of CDP-choline. CDP-choline (Sigma) was administered in two groups as 100 mg/kg and 200 mg/kg via intraperitoneal way 1 hour before the surgical procedure. All rats were weighed separately, an anaesthetic drug dose was calculated for each rat separately. All surgical procedures were performed under anaesthesia by ketamine (80-100 mg/kg, intraperitoneal injection single dose) and xylazine (5-15 mg/kg, intraperitoneal injection single dose). The depth of anaesthesia was followed by the rats' skin or finger pinching responses

Sham and CLP protocol. The skin was disinfected in the sham and CLP groups by wiping from top to bottom at least times using an antiseptic gauze. Laparotomy was performed with a 2,5 cm long midline incision. The caecum was explored, and mesentery was dissected along the caecum line. In sham group, faecal content of the caecum was forced to move into distal segments by gentle palpation. In groups where the CLP procedure was performed, the caecum was ligatured with 4/0 silk suture with attention to keep the ileocecal valve open. The caecum was then perforated from the mesenteric wall to the anti-mesenteric wall using an 18 G needle. After ensuring that needle perforation is adequate to leak faecal content, the caecum was placed again in its anatomic position without contaminating the abdominal wall layers. In rats undergoing laparotomy, the anterior abdominal wall was closed with 4/0 silk sutures in three layers (peritoneal, fascia, and skin). The skin was re-wiped with antiseptic, and a pomade containing fusidic acid was applied on the surgical incision line closed by dressing in sterile gauze. Buprenorphine (0.05 mg/kg subcutaneous injection) was administered for analgesia to prevent post-operative pain during 8 hours of the observation period. All rats were kept in separate cages under post-operative heat and light-controlled circumstances. Fluid support was provided postoperatively as 5 ml per 100 g by subcutaneous saline. The rats were fasted, but they were allowed to reach water freely during the post-operative period.

Histopathological analysis. At the end of the experiment, rats were sacrificed with a high dose of anaesthetic drug according to the ethics rules for animal experiments. Samples obtained from liver and small intestine tissues of rats were fixed in 10 % formalin solution. Tissue sections of 5µm in thickness were prepared from samples embedded in paraffin blocks and stained. Liver and small intestine tissue sections stained with Haematoxylin-Eosin were examined under light microscopy in terms of tissue damage and photographed (Olympus, BX50). The evaluations were done randomly

selected 10 separate fields for each organ obtained from each rat. The liver injury was evaluated in terms of portal and parenchymal inflammation, venous dilatation, congestion, haemorrhage, sinusoidal dilatation. Histological examination of small intestine damage was performed as following parameters; inflammation, villous epithelial degeneration and desquamation, venous dilatation, central lacteal dilatation, congestion, and haemorrhage. All histological changes were graded as follows; 0 for normal (no damage), 1 for light damage, 2 for moderate damage and 3 for heavy damage and the intestinal damage score and liver damage score of the groups were calculated.

Statistical Analysis. The normality of distributions for HR, MAP and RT were evaluated by Shapiro Wilkes test. Mean, standard deviation, minimum, maximum or median, minimum, maximum values were expressed according to normality of distributions. ANOVA test was used to compare the baseline values of the data with normal distribution among the study groups. In the case of statistical significance, Tukey´s test was used in the subgroup analyzes in which the groups were compared in pairs. To compare the 4th and 8th hour values of HR, MAP and RT measurements, changed values in percentage (PC) were calculated according to baseline measurement. PC values were calculated using the formula [(last measurement-first measurement) / (first measurement) x100] and ANOVA or Kruskal Wallis tests were used for comparisons. In statistical significance, the groups were compared in pairs after ANOVA test by using Tukey test and after Kruskal Wallis test by using Dunn test. Comparisons of sepsis-related variables were made using Fisher-Freeman-Halton test. In statistical analysis, Statistical Package for the Social Sciences (SPSS®, IBM Corp. SPSS for Windows, Version 21.0. Armonk, NY, USA) software was used and $p<0.05$ was considered statistically significant.

RESULTS

Vital Parameters. The vital parameters of rats in all groups were compared at various time intervals. The distribution of vital parameters was homogenous. The summary of statistical data for HR, MAP and RT according to groups are given in Tables I, II and III. There was no significant difference between HR in all groups at hour 0 (p=0.268).

Table I. HR according to groups on hours 0, 4 and 8.

Groups	n		HR- Hour 0	HR- Hour 4	HR- Hour 8	¥p
Control	10	Mean	384,70*	406,20*	398,60*	0,676
		SD	65,10	47,22	50,23	
Sham	10	Mean	342,40*	340,30*	336,00*	0,948
		SD	51,85	38,05	43,10	
100 mg/kg CDP-choline	10	Mean	384,10*	398,40*	400,50*	0,819
		SD	71,22	60,92	55,40	
200 mg/kg CDP-choline	10	Mean	343,40*	361,00*	355,70*	0,658
		SD	51,27	40,94	38,04	
Sepsis	10	Mean	373,70*	529,60**	566,60**	<0,001
		SD	46,31	63,28	53,32	
		¥p	0,268	<0,001	<0,001	

Abbreviations: HR according to groups on hours 0, 4 and 8. (SD: Standard Deviation), *p 0>0.05, **p <0.001, vs. all groups; with post-hoc tests. ¥Anova test

Table II. MAP according to groups on hours 0, 4 and 8.

Groups	n		MAP- Hour 0	MAP- Hour 4	MAP- Hour 8	¥p
Control	10	Mean	92,50*	96,31*	96,25*	0,794
		SD	13,72	14,44	14,76	
Sham	10	Mean	115,06*	116,02–*	125,26**	0,129
		SD	11,81	13,08	10,93	
100 mg/kg CDP-choline	10	Mean	108,81*	108,96*	100,01*	0,118
		SD	10,65	12,87	7,86	
200 mg/kg CDP-choline	10	Mean	107,70*	108,14*	101,22*	0,306
		SD	11,38	12,26	9,19	
Sepsis	10	Mean	105,52**¥	77,22**	72,88**	<0,001
		SD	9,48	10,99	9,97	
		¥p	0,002	<0,001	<0,001	

Abbreviations: MAP according to groups on hours 0, 4 and 8. (SD: Standard Deviation), *p 0>0.05, **p <0.001, vs. all group s; <p =0.001 vs. control group; with post-hoc tests. ¥Anova test

However, initial MAP and RT measurements showed a significant difference between all groups (p=0.002 and P=0.028). MAP at hour 0 was significantly lower in the control group. Initial RT measurements also revealed a significant difference between groups (p=0,028). However, groups were self-contained by post-hoc testing, and there was no significant difference between the groups in terms of RT at hour 0.

Table III. RT according to groups on hours 0, 4 and 8.

Groups	n		RT- Hour 0	RT- Hour 4	RT- Hour 8	¥p
Control	10	Mean	37,21*	37,42*	37,16*	0,393
		SD	0,30	0,50	0,50	
Sham	10	Mean	37,17*	37,26*	37,31*	0,632
		SD	0,39	0,34	0,23	
100 mg/kg CDP-choline	10	Mean	36,85*	37,34*	37,65*	0,001
		SD	0,44	0,45	0,35	
200 mg/kg CDP-choline	10	Mean	36,74*	37,06*	37,35*	0,003
		SD	0,29	0,37	0,41	
Sepsis	10	Mean	37,03*	38,50**	38,96**	<0,001
		SD	0,40	0,48	0,50	
		¥p	0,028	<0,001	<0,001	

Abbreviations: RT according to groups on hours 0, 4 and 8. (SD: Standard Deviation), *p 0>0.05, **p <0.001, vs. all groups; with post-hoc tests. ¥Anova test.

Vital parameters were re-evaluated at 4th and 8th hours. HR measurements were significantly higher in group 5 at the 4th and 8th hours than in the other groups (at 4th hour, p<0.001 and at 8th hour, p<0.001). It was determined that HR measurements within each group according to experimental time intervals increased significantly only in group 5 (p<0.001). No significant difference was observed between hour zero, hour 4, and hour 8 in the other groups (Table I).

MAP measurements were significantly different between the groups at the 4th and 8th hours (p <0.001 at the 4th hour and p <0.001 at the 8th hour). MAP values of Group 1 on the 4th hour were significantly higher than those of group 2 (p = 0.01). However, MAP values at 8th hour were considerably higher in group 1 when compared to all other groups (p <0.001). MAP measurements of Group 5 were significantly lower than those of all groups (p <0.001). There was no significant difference between other groups in terms of MAP values (p> 0.05). When each group was analyzed separately according to time intervals, MAP measurements at 4th and 8th hours were significantly lower in Group 5 compared to hour 0 measurements (p <0.001). There was no significant difference in MAP measurements according to time interval in other groups (Table II). RT measurements at the 4th and 8th hours showed significant differences between the groups (p <0.001 for the 4th hour and p <0.001 for the 8th hour). In Group 5, the mean RT value at the 4th and 8th hours was significantly higher than the other groups (p <0.001 and p <0.001, respectively). When we evaluated the groups separately; in group 3, group 4 and group 5, the RT values measured at the 4th and 8th hours differed significantly from initial (hour zero) RT values (p = 0.001, p = 0.003 and p <0.001, respectively). There was no significant difference in RT measurements according to time intervals for groups 1 and 2 (p = 0.393 and p = 0.632, respectively) (Table III).

Effects of CDP-Choline on Hepatic Tissue Damage in Sepsis and Septic Shock. Evaluated parameters are; portal inflammation, parenchymal inflammation, congestion, venous dilatation, sinusoidal dilatation, haemorrhage in hepatic tissues, and evaluating in terms of tissue damage.

Table IV A. Statistical evaluations of histopathological criteria in liver tissue.

HEPATIC	Group 1	Group 2	Group 3	Group 4	Group 5
Portal inflammation	0	0	2^{*+}	$1^{*+\delta}$	$3^{*+\delta x}$
Parenchymal inflammation	0	0	0	0	$2^{*+\delta x}$
Congestion	0	0	2^{*+}	0^{δ}	$3^{*+\delta x}$
Venous dilatation	0	0	1^{*+}	0^{δ}	$3^{*+\delta x}$
Sinusoidal dilatation	0	0	2^{*+}	0^{δ}	$3^{*+\delta x}$
Haemorrhage	0	0	0	0	$2^{*+\delta x}$

Abbreviations: *p <0.001 vs. Group 1, + p <0.001 vs. Group 2, δ p <0.001 vs. Group 3, xp <0.001 vs. Group 4 with post-hoc tests.

Table IV B. Statistical evaluations of histopathological criteria in intestinal tissue.

INTESTINAL	Group 1	Group 2	Group 3	Group 4	Group 5
Inflammation	0	0	2^{*+}	0^{-}	3^{*+-x}
Epithelial degeneration	0	0	2^{*+}	0^{-}	3^{*+-x}
Congestion	0	0	2^{*+}	0^{-}	3^{*+-x}
Venous dilatation	0	0	2^{*+}	0^{-}	3^{*+-x}
Central lacteal dilatation	0	0	2^{*+}	0^{-}	3^{*+-x}
Haemorrhage	0	0	2^{*+}	0^{-}	3^{*+-x}

Abbreviations: *p <0.001 vs. Group 1, + p <0.001 vs. Group 2, δ p <0.001 vs. Group 3, xp <0.001 vs. Group 4 with post-hoc tests.

The critical result is compared to CDP-sepsis group (Group 5) with CDP- choline groups (Groups 3 and 4) separately; found a statistically significant difference between the groups in terms of all parameters ($p<0.001$) on hepatic tissues. A statistically significant difference was found between Groups 3 and 4 regarding other parameters of tissue damage as portal inflammation, congestion, venous dilatation, and sinusoidal dilatation ($p<0.001$) (Tables IVA and B).

Effects of CDP-Choline on Intestinal Tissue Damage in Sepsis and Septic Shock. Inflammation, epithelial degeneration, congestion, venous dilatation, central lacteal dilatation, haemorrhage parameters were evaluated. When CLP-sepsis group (Group 5) and both CDP-choline groups (Groups 3 and 4) were compared separately, a statistically significant difference was found between the groups in terms of all parameters ($p<0.001$) on intestinal tissues. When Group 3 and Group 4 were compared, a statistically significant difference was found between the two groups in all parameters ($p<0.001$) (Table IV). Based on dose according to the compared results, the tissues of the rats treated with 100 mg/kg CDP -choline for both tissues seem to be positively affected. In addition, damage in the tissues of rats administered 200 mg/kg is much less. Taking into our results, we can say that CDP choline has a dose-dependent protective effect on both liver and small intestine tissue. The 200 mg/kg dose has a better protective effect than the 100 mg/kg dose in which can be seen in the tissues clearly, and these remained much better preserved. Morphological differences between the groups are observed in Figures 1 and 2.

DISCUSSION

Sepsis is a life-threatening condition that arises from the host's inappropriate response to infection that occurs physiological, pathological, and biochemical abnormalities (Singer *et al.*, 2016). Sepsis-associated organ dysfunction includes multiple responses to inflammation, including endothelial and microvascular dysfunction (Pool *et al*., 2018). The most common causative agent of sepsis is gram-negative coliform bacilli. CLP model is the most preferred for the experimental sepsis model because it aimed to contaminate the peritoneum of the colonic gram-negative bacilli by perforating the cecum and creating intraperitoneal sepsis.

Various pharmaceuticals have been tested both in clinical studies on patients and experimental sepsis models on rats (Doig *et al*., 2003; Mathias *et al*., 2015; Feng *et al*., 2017; Schulz *et al*., 2019). Since the systemic inflammatory response is a highly complex process at the cellular and molecular level, the precise mechanism of action to reduce inflammation by some agents still needs further investigation. There is evidence that many of these substances have positive effects on uncontrolled inflammatory response and can prevent tissue damage to certain amounts (Rhodes *et al*., 2017). Many clinical and experimental studies attract the attention of CDP-choline (Cansev *et al*., 2008; Yilmaz *et al*., 2008; Coskun *et al*., 2014). Most of the studies performed with CDP-choline examine the neuroprotective effects of the molecule. The number of studies investigating the effects on ischemia-reperfusion injury in visceral organs or the protective effects on visceral organ damage in sepsis appears very limited.

Parrish *et al*. (2008) also suggested that the use of intraperitoneal citicoline in the experimental sepsis model led to suppression of TNF-a and HMGB1 levels and increased life expectancy in endotoxin-induced septic shock. Another interesting study has shown that myocardial infarction size and the number of apoptotic cells can be significantly reduced by applying CDP-choline in an experimental myocardial infarction model created in rats (Coskun *et al*., 2014). Therefore, it is seen that CDP-choline has various physiological effects besides its cytoprotective properties. These positive effects of citicoline will perhaps enable this molecule to take place in the supplementary/ complementary treatment of obesity in the future. In a more

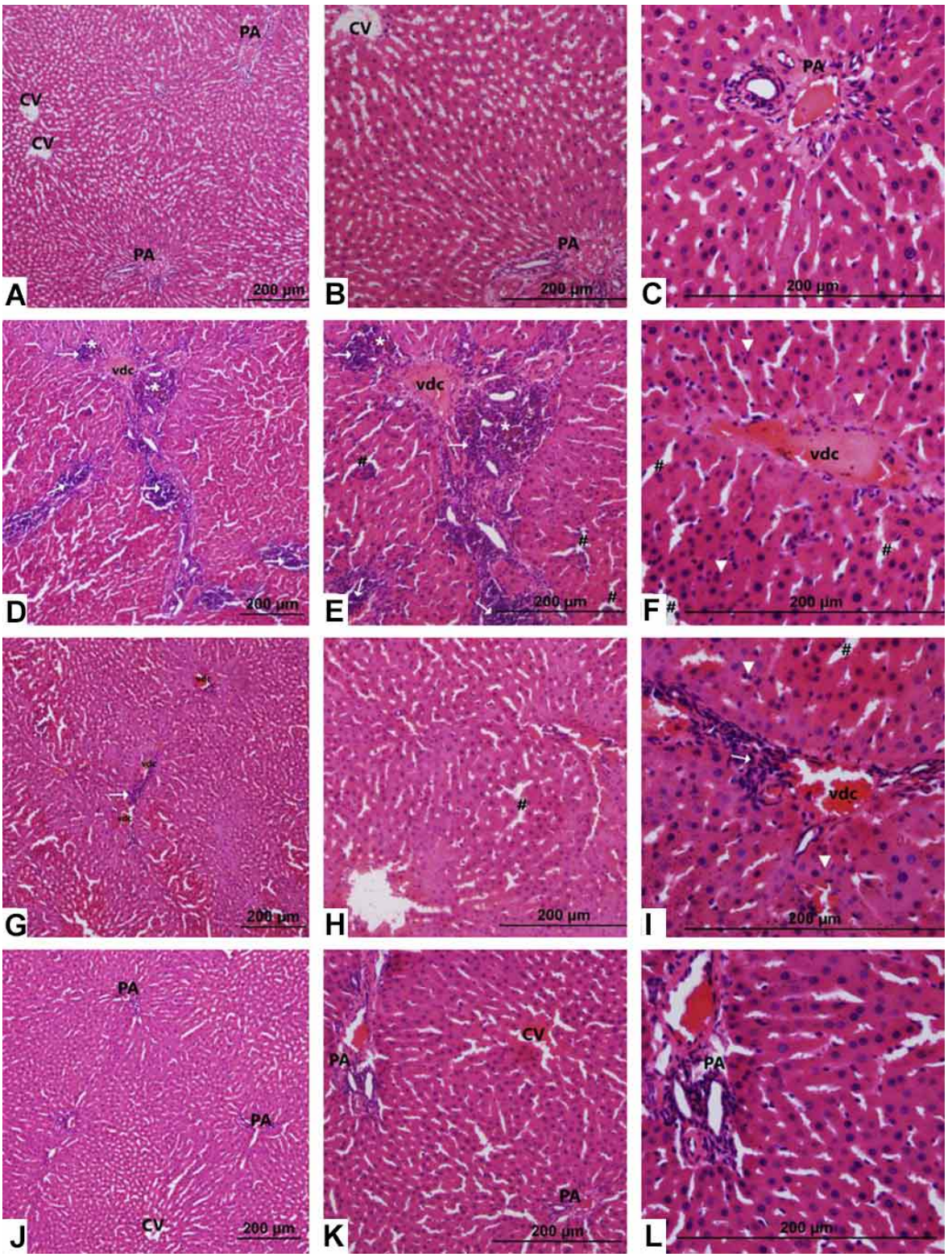

Fig. 1. Histological appearance of liver tissue. Control group (Group 2) (a-c). CLP-Sepsis group (Group 5) (d-f); portal and parenchymal inflammation (d, e), venous dilatation and congestion (d-f), sinusoidal dilatation (e, f), haemorrhage (d, e). CLP+100mg/kg CDP-choline group (Group 3) (g-i); portal inflammation (g, i), venous dilatation and congestion (g, i), sinusoidal dilatation (h, i). CLP+200mg/kg CDP-choline group (Group 4) (j-l). (PA: Portal area, CV: Central vein, ›: Inflammation, *: Haemorrhage, vdc: Venous dilatation and congestion, #: Sinusoidal dilatation. H&E

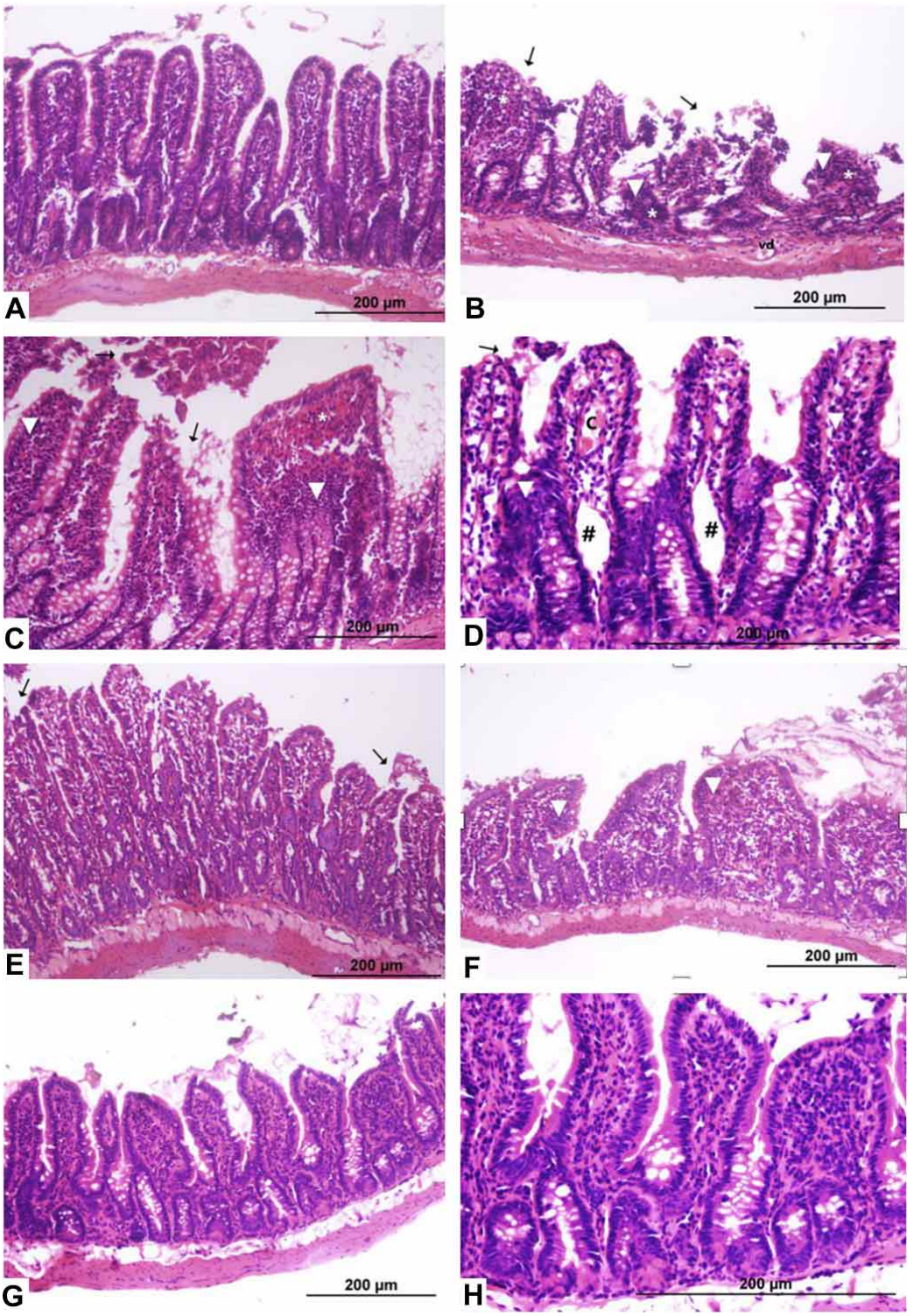

Fig. 2. Histological appearance of small intestine. Control group (Group 2) (a). CLP-Sepsis group (b-d); villous epithelial degeneration and desquamation (b-d) , inflammation (b, c), haemorrhage and disintegration in lamina propria (b, c), venous dilatation (b), congestion (c), central lacteal dilatation (d). CLP+100mg/kg CDP- choline group (Group 3)(e, f); villous epithelial degeneration and desquamation (e), inflammation (f). CLP+200mg/kg CDP-choline group (Group 4) (g, h). (t: Inflammation, *: Haemorrhage, ›: Villous epithelial degeneration and desquamation, vd: Venous dilatation, C: Congestion, #: Central lacteal dilatation). H&E

recent article published by Schmidt *et al*. (2015), they suggested that CDP-choline has a protective effect on microvascular barrier function during endotoxemia and can be used as a therapeutic agent to treat of capillary leakage in sepsis, given the excellent pharmacological safety profile of the molecule. In their experimental model, the authors underlined that bacterial lipopolysaccharide triggers microvascular permeability and leukocyte adhesion, which are the main pathophysiological mechanism of mortality and morbidity associated with sepsis. It showed that CDP-choline administered at 100 mg/kg could significantly prevent these adverse effects. As a matter of fact, in our study, it was seen that CDP-choline prevented venous dilatation due to endotoxemia in hepatic and intestinal tissue samples. We think that this effect occurs through the mechanisms mentioned in the studies of Jambou *et al*. (2009) and Schmidt *et al*. (2015). Hernekamp *et al*. (2015) published an exciting study and investigated the effects of CDP-choline on edema that develops in rats exposed to thermal damage. The authors injected a 100 mg/kg bolus of CDP-choline into rats burned to 30 % of the body surface with hot water and examined the results. They investigated capillary leakage by fluorescein isothiocyanate albumin extravasation method and showed that edema and albumin leakage due to burning could be reduced significantly with citicoline application. This study is essential in demonstrating that the anti-inflammatory effects of CDP-choline can be used in burn patients. The body's metabolic and hormonal response to trauma-burn, sepsis, or mechanical trauma-are common. Therefore, whether it is the findings obtained from our study and other studies in the literature where different traumatic processes are evaluated, all these data prove that CDP-choline has a potent anti-inflammatory effect.

It is known that the CDP-choline pathway is suppressed during apoptosis (Morton *et al*. 2013). Therefore, the availability of phosphatidylcholine required for membrane stabilization and intracellular signaling is limited. Thus, the administration of CDP-choline can both prevent apoptosis and reduce the CDP-choline deficit that occurs during apoptosis. Although cell death was not examined with detailed histological tests regarding the distinction between necrosis and apoptosis in our study, since no cell death was observed in the 200 mg CDP-choline group, the data we obtained support previous information the application of CDP-choline is protective against apoptosis. Our research shows that the application of CDP-choline ameliorates inflammation in both liver and small intestine either with 100 mg/kg dose or 200 mg/kg dose. Moreover, it has been determined that the molecule has a dose-dependent effect and CDP-choline administered at 200 mg/kg gives much better results than the dose CDP-choline administered at 100 mg/kg dose.

In conclusion, CDP-choline has essential functions in all mammalian cells. Although our study hopes that this molecule can be used to prevent gastrointestinal system complications in patients with sepsis, intensive clinical studies are required. It might be proposed that indications of this safe and effective molecule will expand with new studies conducted with CDP-choline. There are available data about anti-inflammatory, antioxidant, and cytoprotective effects and mechanism of action of this molecule. We believe that the benefits of the therapeutic use of citicoline in sepsis and septic shock may be more accurately evaluated in further clinical trials with a prospective randomized fashion involving large series of patients.

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RESUMEN: Cincuenta ratas albinas Wistar macho se dividieron en 5 grupos; Grupo 1 como grupo control simulador, el grupo 2 como grupo de control, el grupo 3 como grupo al que se administró 100 mg/kg de CDP-colina, el grupo 4 como grupo al que se administró 200 mg/kg de CDP-colina y el grupo 5 como grupo con sepsis. El modelo de sepsis se realizó ligando y perforando el intestino ciego de las ratas. Los tejidos del hígado y del intestino delgado se evaluaron histológicamente o cuantitativa y cualitativamente. Hubo una diferencia significativa entre los grupos de sepsis y CDP-colina para el daño hepático e intestinal evaluado en muestras de tejido ($p<0,001$). El tratamiento con CDP-colina mejoró parcialmente, según la dosis, los parámetros clínicos de sepsis y shock séptico y revirtió el daño micro anatómico causado por la sepsis.

PALABRAS CLAVE: CDP-colina; Hígado; Intestino delgado; Daño al tejido.

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