RESEARCH ARTICLE

Protective Effect of Echinacea purpurea Plant Extract Against Pasterullosis in Rabbits: Hematological, Biochemical and Oxidative Stress Studies

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Abstract

Echinacea purpurea (E. Purpurea), as one of the most important plants, has been used in prevention and control of infectious diseases of respiratory systems. In this study, we investigated the effect of Echinacea andenrofloxacin on Pasteurella multocida type A infection in male white New Zealand rabbits through evaluation of the hematological, biochemical, phagocytic activities, and oxidative stress markers. Forty male white New Zealand rabbits were divided into four equal groups; group 1 was kept as a control, group 2 was intranasally infected with P. multocidatyp A (4x10⁷ CFU/mL/rabbit) at the 14th day from beginning of experiment (FBE), group 3 was administrated E. Purpure orally (130mg/kg body weight “B.W.”) day after day from 1st day to the 35th day and infected with P. multocida the 14th day FBE and group 4 was intranasally infected with P. multocida at the 14th day FBE, then treated with enrofloxacin (10 mg / kg B.W. in drinking water) after the appearance of the clinical signs from the 18th-20th day. The results revealed that P. multocida infection in rabbits produced macrocytic hypochromic anemia (RBCs = 5.26 x10⁶/μL, MCV = 76.06 Fl, MCHC = 24.99%), leukocytosis (14.09x10³/μL) with a reduction in the phagocytic percent (51.60 %) and phagocytic index (0.26). We observed decrease in serum levels of total protein, albumin, and globulin. Moreover, significant increase in serum alanine aminotransferase (ALT, 34.4 U/L) and alkaline phosphatase (ALP, 27.80 U/L) were recorded. Also, the serum bilirubin (total bilirubin = 1.03mg/dL, direct bilirubin = 0.61 mg/dL and indirect bilirubin = 0.42 mg/dL) levels were significantly elevated. Serum urea (47.32mg/dL) and creatinin levels (2.10 mg/dL) were significantly increased. Furthermore, P. multocida infection in rabbits induced oxidative stress, which was observed in a significant reduction in the serum catalase (CAT, 3.13 nmol/L) with elevation in the lipid peroxidation (MDA, 3.46 nmol/L). In conclusion, Echinacea treatment before and after rabbit infection with P. multocida type A is able to ameliorate the alterations in all studied parameters compared to enrofloxacin treated rabbits.

Key words: Rabbits, Echinaceapurpurea, Enrofloxacin, Pasteurellamultocida, Oxidative stress.

Introduction

Echinaceapurpurea is one of the most important medicinal herbs of the genus Echinacea[1]. It is a purple cone flower that is widely used as a medicinal plant in the United States and Europe. The main active principles are alkamides, glycoproteins, caffeic acid derivatives and polysaccharides. Echinacea purpurea has immunostimulatory, immunomodulator, antibacterial, anti-inflammatory, wound healing, antiseptic and antineoplastic properties and growth promoting effects. Moreover, it has some short term effects against herpes, vesicular stomatitis and influenza viruses [2- 7]. Echinacea
improves the immunity, decreases the blood sugar levels, anxiety, and inflammation. Also, it has anti-cancer and skin health improvement effects as well as it was documented to be effective in prevention and complex treatment of Corona virus.[8]. Echinacea purpurea (E. Purpurea) is one of the most promising herbal medicine for improving the immunity of various species of animals and humans [9].

Enrofloxacin is the first fluoroquinolone of second generation quinolones[10]. It acts by inhibiting the DNA gyrase, which is essential for the coiling and uncoiling of bacterial DNA and packing it within the cell [11]. Enrofloxacin is indicated for the treatment of bacterial infections of the alimentary and respiratory tracts; it is rapidly distributed through the tissues before being eliminated [12].

Pasteurellosis is a common and highly contagious disease in domestic rabbits that transmitted by direct contact and aerosol transmission. It is characterized by high morbidity and mortality rates [13]. About 30–90% of apparently healthy rabbits may be a symptomatic carrier[14]. It is one of the main causes of economic losses in rabbits [15]. Pasteurella infection is depending on age and stress factors [16]. It has three forms in rabbitaries in Egypt. The 1st one is, subacute form during first two days post infection (PI). This form manifested by drowsiness, sneezing, pyrexia and anorexia. In the third day, they became off food with lacrimation and mild diarrhea. In the later stages, infected rabbits showed dullness, emaciation, coughing and breathing with a sound "snuffling". Also nasal discharge became turbid and sticky [17]. The subclinical infection was established in the upper respiratory tract. After colonization, the infection extends to the rest of respiratory tract and can cause clinical rhinitis, conjunctivitis, pneumonia, tracheitis or otitis media[14]. The 2nd form is characterized by abscess formation at any part in the body and the case is terminated with septicemia[18]. Otitis media or interna form includes accumulation of pus or fluid in the middle or inner ear, so the rabbit twist its head e.g "wry neck" or torticollis. Also, subcutaneous and visceral abscesses may be clinically silent for long periods and spontaneously rupture[19]. The third form manifested by urogenital infection, mastitis, pyometra, orchitis, infertility, abortion and meningitis [20].

This study aimed to investigate the efficacy of E. Purpurea plant and enrofloxacin in controlling P. multocida experimental infection in rabbits.

Material and Methods

Experimental animals

A total number of forty, 30 days old, apparently healthy male white New Zealand rabbits (750-1000g) were purchased from the Faculty of Agriculture, Zagazig University, Egypt. These rabbits were kept under good hygienic conditions, housed in clean metal cages, kept at 24 ± 2 °C with a 50–60% humidity and a 12-hour light:dark cycle, fed on well balanced commercial ration and water was provided ad-libitum. Rabbits were kept for 2 weeks before starting the experiment. The management and treatment procedures were done according to the Guidelines for the Care and Use of Laboratory Animals of the National Institutes of Health.

Chemical treatment agents

Enrofloxacin (Opitryl) was manufactured by El-Obour Modern Pharmaceutical Industries Co, Egypt. Each 100 mL contains 10g enrofloxacin, it was given as a 10% oral solution. Echinacea purpurea dry extract capsules (each one contains 175mg) were purchased from Arab Company for
Pharmaceuticals and Medicinal Plants, Purpureaaco-Egypt. All the biochemical tests were performed using test kits of Diamond Diagnostics, Egypt.

**Bacterial strain**

*Pasteurullamultocida* type strain C51-17 (capsular type A) was obtained from the Animal Health Research Institute, Dokki, Giza, Egypt. Rabbits were infected intranasally by a dose of 1mL with a final concentration of 4x10^7 colony forming unit (CFU).

**Experimental design**

Forty apparently healthy male, white New Zealand rabbits, were randomly classified into four equal groups (each group contains 10 rabbits), Group 1 was given 1mL phosphate buffer saline (PBS) intranasal (IN) and kept as a negative control. Group 2 was infected with *P. multocida* type A (4x10^7 CFU/mL/rabbit) at the 14th day [21], from the beginning of the experiment (FBE) (a positive control). Group 3 was orally administrated *E. purpurea* (130 mg/kg B.W.) day after day [22] from 1st day to the 35th day and infected with the above mentioned dose of *P. multocida* type A at 14th day FBE. Group 4 was infected IN with *P. multocida* type A at the 14th day, and treated with enrofloxacin (10mg/kg B.W.), orally by gavag for 3 days after the appearance of the clinical signs [23].

**Samples collection and preparation**

Blood samples were collected from 3 rabbits in each group under anesthetiaon the 35th day FBE from the ear vein. Two mL of the blood was collected in a heparinized test tube for evaluation of phagocytic activities, 0.5mL of the blood collected in a test tube with ethylene diamine tetra acetic acid (EDTA) for hematological examination, and 5mL was collected in serum separation glass tube without anticoagulant to measure biochemical parameters.

**Hematological assay**

Collected blood samples on EDTA (10%) were used for evaluation of total erythrocyte count, packed cell volume (PCV), hemoglobin (Hb), white blood cells (WBC) and differential leukocyte counts (DLC) according to Coles[24]. Mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) values were calculated.

**Phagocytic percent and index**

The heparinized blood samples were used for leukocytes separation. *Candida albicans* (*C. albicans*) was prepared at Animal Health Research Institute Zagazig and used for determination of the phagocytic activity according to Wilkinson[25]. Macrophages (monocytes and lymphocytes) number containing *C. albicans* (phagocytic %) that were engulfed by 100 phagocytes in each individual preparation was examined under light microscope under oil immersion lens. The phagocytic index was calculated through evaluating the average number of attached and ingested *C. albicans* multiplied by the phagocytic % [26].

**Biochemical assay**

Serum samples were analyzed for determination of alanine amino transferase (ALT) by the method of Reitman and Frankel[27] and alkaline phosphatase (ALP) according to a previously published protocol[28]. Total protein and albumin levels were measured described elsewhere[29,30]. The globulin level was calculated by subtracting the obtained albumin level from obtaining total protein level according to Doumas and Biggs[31]. Serum bilirubin (total and direct) was determined according to Colombo [32] and the indirect bilirubin level was calculated as the difference between total and direct bilirubin. Urea and creatinine were determined documented previously [33, 34].

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**Oxidative stress markers assay**

Serum samples were used to determine the catalase (CAT) activity which was measured by an enzymatic colorimetric method using ready-made kits provided by Biodiagnostic Egypt. CAT No. CAT 25 17 according to Aebi [35], and lipid peroxidation (MDA) was determined depending on the thiobarbituric acid reactivity of Satoh [36], CAT No. 25 29.

**Statistical Analysis**

The obtained data was statistically analyzed using "F" test one way ANOVA according to Tamhans and Dunlop (37) using "MSTAT-C" computer program. Means in the same column followed by different letters were statistically significant at P < 0.05 and the highest values were represented with the letter (a).

**Results**

**Clinical signs and mortality rate**

Negative control rabbits appeared healthy without any clinical signs of illness and no mortalities were recorded. On the other hand, rabbits in positive control group showed ruffled fur, sneezing, nasal discharge, purulent exudates from nose, abnormal respiratory sound and mortality rate reached 60%. Both rabbits treated with **Echinacea purpurea** and enrofloxacin then infected with **P. multocida** showed mild clinical signs with mortality rates of 33.33% and 20% in groups 3 and 4, respectively.

**Hematological findings**

Rabbits infected with **P. multocida** showed a significant decrease (P < 0.05) in RBCs count, Hb concentration, PCV and MCHC, with a significant increase in MCV (macrocytic hypochromic anemia) compared with the control. While rabbits infected and treated with **E. purpurea** and enrofloxacin showed improvement in RBCs count, Hb concentration, PCV and MCHC with significant decrease (P < 0.05) in MCV in comparison with infected rabbits (Table 1).

In comparison to negative control rabbits, infected rabbits showed a significant increase (P < 0.05) in the counts of leukocytes, granulocytes and monocytes with non-significant change in lymphocytic count. While **E. purpurea** and enrofloxacin treated rabbits showed a significant decrease (P < 0.05) in total leukocytes count, granulocytic count and monocyte count with non-significant change in lymphocyte count when compared to the infected group (Table 2). Moreover, **P. multocida** infected rabbits showed a significant decrease (P < 0.05) in phagocytic percent and phagocytic index compared to control negative rabbits, while **E. purpurea** and enrofloxacin treated rabbits showed a significant increase (P < 0.05) in phagocytic percent and phagocytic index comparing with infected rabbits (Table 2).

**Biochemical findings**

Serum activities of ALT, ALP and bilirubin (total, direct and indirect) levels showed a significant increase (P < 0.05) in rabbits infected with **P. multocida** compared to the negative control, while treated groups 3 and 4 showed a significant decrease (P < 0.05) in serum ALT, ALP activities and bilirubin levels when compared to infected rabbits (Table 3). Serum urea and creatinine levels showed a significant increase (P < 0.05) in group 2 compared with the negative control, while groups 3 and 4 showed a significant decrease (P < 0.05) compared with the infected group (Table 4).

**Antioxidant and oxidative stress findings**

Rabbits infected with **P. multocida** showed a significant decrease (P < 0.05) in serum CAT activity and significant decrease (P < 0.05) in MDA level compared to the negative control.
While rabbits treated with *E.purpurae* and enrofloxacin (groups 3 and 4) showed a significant increase ($P < 0.05$) in MDA level compared to infected rabbits (Table 5).

**Table (1):** Erythrogram (mean value ±SE) of rabbits’ groups at the 35th day from the beginning of the experiment.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Parameters</th>
<th>RBCs Count x10⁶/µL</th>
<th>Hb g/dL</th>
<th>PCV %</th>
<th>MCV Fl</th>
<th>MCH pg</th>
<th>MCHC (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>GP1 (Control)</td>
<td></td>
<td>6.48±0.30</td>
<td>12.50±0.37</td>
<td>43.15±0.98</td>
<td>66.92±1.04</td>
<td>19.38±0.13</td>
<td>28.96±0.27</td>
</tr>
<tr>
<td>GP2 (P.multocida)</td>
<td></td>
<td>5.26±0.14</td>
<td>10.00±0.33</td>
<td>40.00±0.85</td>
<td>76.06±1.83</td>
<td>19.00±0.46</td>
<td>24.99±0.14</td>
</tr>
<tr>
<td>GP3 (Echinacea+P.multocida)</td>
<td></td>
<td>6.10±0.18</td>
<td>12.00±0.48</td>
<td>42.00±1.00</td>
<td>68.85±0.46</td>
<td>19.67±0.15</td>
<td>28.57±0.14</td>
</tr>
<tr>
<td>GP4 (P.multocida+Enrofloxacin)</td>
<td></td>
<td>6.21±0.22</td>
<td>12.20±0.23</td>
<td>42.70±0.24</td>
<td>68.76±0.60</td>
<td>19.46±0.37</td>
<td>28.57±0.92</td>
</tr>
</tbody>
</table>

Means within the same column carrying different letters are significantly different at $P < 0.05$

RBCs Count: Red blood cell count; MCV: Mean corpuscular volume; Hb: Hemoglobin; MCH: Mean Corpuscular Hemoglobin; MCHC: Mean Corpuscular Hemoglobin Concentration; Fl: femtoliters (one thousand trillionths of a liter) per/cell; PGV: Packed Cell Volume; pg: picograms/cell (one trillionth of a gram); x10⁶/µL: millions of cells per microliter; g/dL: grams per deciliter.

**Table (2):** Leukogram and phagocytic activity (mean value ±SE) of rabbits' groups at the 35th day from the beginning of the experiment.

<table>
<thead>
<tr>
<th>Groups</th>
<th>TLC (x10⁶/µL)</th>
<th>Differential leukocytic count</th>
<th>Phagocytic</th>
<th>Phagocytic</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Granulocytes (x10⁶/µL)</td>
<td>Lymphocytes (x10⁶/µL)</td>
<td>Monocytes (x10⁶/µL)</td>
</tr>
<tr>
<td>GP1 (Control)</td>
<td>12.74±0.12</td>
<td>5.63±0.11</td>
<td>6.56±0.29</td>
<td>0.55±0.02</td>
</tr>
<tr>
<td>GP2 (P.multocida)</td>
<td>14.09±0.17</td>
<td>6.80±0.03</td>
<td>6.43±0.21</td>
<td>0.85±0.06</td>
</tr>
<tr>
<td>GP3 (Echinacea+P.multocida)</td>
<td>13.14±0.14</td>
<td>6.00±0.09</td>
<td>6.56±0.11</td>
<td>0.58±0.03</td>
</tr>
<tr>
<td>GP4 (P.multocida+Enrofloxacin)</td>
<td>12.89±0.16</td>
<td>5.80±0.09</td>
<td>6.54±0.13</td>
<td>0.55±0.05</td>
</tr>
</tbody>
</table>
Means within the same column carrying different letters are significantly different at $P < 0.05$, TLC : Total Leukocytic Count, $x10^3/\mu L$ : Thousand per microliter

**Table (3):** Some serum biochemical parameters (mean value ±SE) of rabbits' groups at the 35th day from the beginning of the experiment.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Groups</th>
<th>ALT U/L</th>
<th>ALP U/L</th>
<th>Total bilirubin mg/dL</th>
<th>Direct bilirubin mg/dL</th>
<th>Indirect bilirubin mg/dL</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>GP1 (Control)</td>
<td>23.60c</td>
<td>18.80c</td>
<td>0.56c</td>
<td>0.31cd</td>
<td>0.25c</td>
</tr>
<tr>
<td></td>
<td>GP2 (P. multocida)</td>
<td>±0.45</td>
<td>±0.25</td>
<td>±0.02</td>
<td>±0.01</td>
<td>±0.01</td>
</tr>
<tr>
<td></td>
<td>GP3 (Echinacea + P. multocida)</td>
<td>±0.87</td>
<td>±0.72</td>
<td>±0.06</td>
<td>±0.04</td>
<td>±0.03</td>
</tr>
<tr>
<td></td>
<td>GP4 (P. multocida + Enrofloxacin)</td>
<td>24.10c</td>
<td>20.00bc</td>
<td>0.62c</td>
<td>0.35bc</td>
<td>0.27c</td>
</tr>
</tbody>
</table>

Means within the same column carrying different letters are significantly different at $P < 0.05$

ALT: Alanine transaminase, mg/dL: milligram per deciliter, ALP: Alkaline Phosphatase U/L: international units/liter.

**Table (4):** Some renal function tests (mean value ±SE) of rabbits' groups at the 35th day from the beginning of the experiment.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Groups</th>
<th>Urea mg/dL</th>
<th>Creatinine mg/dL</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>GP1 (Control)</td>
<td>38.60c</td>
<td>1.10c</td>
</tr>
<tr>
<td></td>
<td>GP2 (P. multocida)</td>
<td>±0.32</td>
<td>±0.05</td>
</tr>
<tr>
<td></td>
<td>GP3 (Echinacea + P. multocida)</td>
<td>47.32a</td>
<td>2.10a</td>
</tr>
<tr>
<td></td>
<td>GP4 (P. multocida + Enrofloxacin)</td>
<td>41.50b</td>
<td>2.04b</td>
</tr>
</tbody>
</table>

Means within the same column carrying different letters are significantly different at $P < 0.05$

mg/dL: milligram /deciliter

**Table (5):** Serum catalase and malondialdehyde (mean value ±SE) of rabbits' groups at the 35th day from the beginning of the experiment.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Groups</th>
<th>CAT (nmol/L)</th>
<th>MDA (nmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>GP1 (Control)</td>
<td>4.56a</td>
<td>1.86b</td>
</tr>
<tr>
<td></td>
<td>GP2 (P. multocida)</td>
<td>±0.32</td>
<td>±0.19</td>
</tr>
<tr>
<td></td>
<td>GP3 (Echinacea + P. multocida)</td>
<td>3.13b</td>
<td>3.46a</td>
</tr>
<tr>
<td></td>
<td>GP4</td>
<td>4.36a</td>
<td>2.00b</td>
</tr>
</tbody>
</table>
Discussion

*Pasteurella multocida* is the most common bacterial pathogen in rabbits. Stress, such as shipping, mating and handling, may cause replication of many serotypes of *P. multocida*. Depression, inappetence, rhinitis, sneezing, coughing, nasal discharge, conjunctivitis, breathing with a snuffling sound, pneumonia and torticollis were recorded in *P. multocida* infected rabbits, that attributed to the infection with *P. multocida* respiratory route causing inflammation of mucous membranes before spreading through blood [14, 16, 19, 20 and 38]. The high mortality rate was recorded in the infected rabbits that resulted from the side effect of *P. multocida* endotoxin [15, 17]. Severity of clinical signs and mortality rates was improved in the treated groups (3, 4), which mainly attributed to the presence of alkaloids, tannins and sterols or total phenols and flavonoids in the *E. purpurea* that have antimicrobial activity [39] and the antibacterial activities of enrofloxacin [40].

Concerning the erythrogram, *P. multocida* infected rabbits exhibited a significant reduction in RBCs count, Hb concentration and PCV% with the development of macrocytic hypochromic anemia that attributed to the hemorrhagic effect of the bacteria and/or its endotoxin on RBCs, which led to reduce their life span which stimulate active erythropoiesis that led to reticulocytosis. Our data supported the previously obtained results by Praveena and coauthors [41]. Rabbits protected and treated with *E. purpurea* before and after *P. multocida* infection (group 3) showed an improvement in the erythrogram. This indicates that using *E. purpurea* as prophylactic and treatment is more effective in protecting the cells [42, 43]. Enrofloxacin treated rabbits (group 4) also showed an improvement in the erythrogram due to the effectiveness of this antibiotic against *P. multocida*.

Leukocytosis was observed in rabbits infected with *P. multocida* due to heterophilia and monocytosis. Such elevation in heterophils and monocytes counts could be due to the infection and inflammation [24]. Heterophiles and monocytes counts were reduced in rabbits treated with *E. purpurea* before and after infection with *P. multocida* due to anti-inflammatory and antibacterial effect of *E. purpurea* [44]. Also, there was an improvement in the total leukocytic count, heterophils and monocytes in enrofloxacin treated rabbits.

Rabbits infected with *P. multocida* revealed a reduction in the phagocytic percent and phagocytic index, which attributed to the effect of oxidative stress that exhaust the body immune response resulted from infection and/or *P. multocida* endotoxin [45]. Phagocytosis was inhibited by *P. multocida* capsular polysaccharides [46]. While, rabbits treated with *E. purpurea* revealed a significant improvement in the phagocytic percent and index attributed to the glycoprotein rich fractions of *E. purpurea* that enhance the phagocytic activity [47, 48], also the immune-stimulatory effects of lipophilic alkylamides, polar caffeic acid derivative and cichoric acid of *E. purpurea* [49, 50]. The improvement of phagocytic percent and index was recorded in enrofloxacin treated rabbits due to the effectiveness of this antibiotic against *P. multocida* so reduces its negative impact on phagocytic percent and index.

Regarding to the biochemical results, rabbits infected with *P. multocida* showed reduction in the total protein and albumin with an elevation of globulin levels, which
may be due to a reduction in feed intake by diseased animal and / or protein synthesis by damaged liver. Also, it may be due to increase protein loss nephropathy [14]. While the hyperglobulinemia indicated the immune defense of rabbits due to respiratory infection and increase the level of antibodies to combat this infection[51, 52]. Hepatic enzymes (ALT and ALP) significantly elevated in the infected rabbits[53], such elevation in ALT activity due to the hepatocellular damage resulted from the infection and subsequent liberation of these enzymes from damaged hepatocytes into the circulation, also serum ALP elevation is associated with cholestasis [54].

Rabbits received *E.purpurea* before and after *P.multocida* infection showed an elevation in the protein level with reduction in serum activities of ALT and ALP that attributed to the hepato-protective effect of *Echinacea* [7 and 55], also *Echinacea* species stimulate the immune response of the body [56]. Moreover, rabbits treated with enrofloxacin showed a significant improvement in the total protein, albumin, globulin and hepatic enzymes. Fluoroquinolones act by interfering with bacterial DNA synthesis so preventing the growth and replication of *P.multocida*[57] reducing the damaging effect of it. Furthermore, serum total, direct and indirect bilirubin were elevated in infected rabbits, that attributed to hepatocellular damage resulted from the effect of *P.multocida* and/or its toxin on hepatocytes [17]. Thus, all major excretory steps of bilirubin may be distributed and resulted in an elevation in total bilirubin [54], these findings agree with Gilavits and Magyer[58]. Administration of *E.Purpurea* before and after *P.multocida* infection induced reduction in total, direct and indirect bilirubin, which attributed to the presence of large amount of chicoric acid and caftaric acid in *E.Purpurea*, which had a role in inhibition of hyaluronidase enzyme, that secreted by bacteria to facilitate penetration into tissue [59], so treatment with *E.Purpurea* protect hepatic cell and reduce the elevated bilirubin[55]. Moreover, using enrofloxacin could counteract the effect of *P. multocida* and reduce the level of bilirubin.

Concerning the kidney function tests, *P.multocida* infected rabbits exhibited a significant elevation in the levels of serum urea and creatinine levels, which attributed to the renal damage induced by *P. multocida* and/or its endotoxin on renal tissue. Also, the circulating toxin may lead to reduce the renal blood flow and glomerular filtration rate leading to accumulation of serum nitrogenous end products[60], our results run on the same ground with those reported by Gaber[15].

Rabbits administered *E.purpurea* before and after infection revealed amelioration of renal function, which confirms the ability of *E.purpurea* to protect the renal tissue against infection and toxins due to its high content of natural metabolites that have antibacterial activity [61], also the alkamides content of *E.Purpurea* down regulates inflammatory responses in the tissue [62]. Our results are in accordance with previously published researches[54, 63]. Moreover, enrofloxacin administered rabbits showed improvement in the serum urea and creatinin levels, which attributed to its antibacterial activity, which suppress bacterial infection and decreasing the damaging effect on the kidney[64].

Concerning the oxidative stress marker evaluation, infected rabbits showed a significant reduction in serum activity of CAT with elevation MDA level. From our opinion, this may be due to endotoxin effect of *P.multocida* which increased the oxidative stress and exhausting the antioxidant defense system, leading to decrease of CAT enzyme and increase of lipid peroxidation [37 and 65]. Rabbits treated with *E.purpurea* showed a significant elevation in the serum CAT level with the reduction MDA level,
which may be due to the natural components found in *E. purpurea* (echinacoside, caffeic acid, total phenols, and flavonoid), which have antioxidant activities by elimination of free radicals and subsequently prevent destruction of cell [7, 39, 66, 67, 68] and the alkalamides contents decreases the proinflammatory cytokines and TNF-alpha [62]. Enrofloxacin improved the antioxidant status of infected rabbits through its ability to reduce bacterial replication, as it is effective against *p. multocida* infection, so reduces the oxidative stress resulted from infective microorganism and its toxin.

**Conclusion**

Our results indicate that using of *Echinacea* for prophylactic and treatment of *Pasteurella* infection returned the altered hematological, biochemical and oxidative stress markers nearly to the normal.

**Conflict of Interest**

The authors have no conflict to declare.

**Reference**


الملخص العربي
التأثير الوقائي لنبات الأشنسي على الإشنسا على الخلايا الدم وكيمايا الدم والقدرة على الإلتهام ومضادات الأكسدة في الأرانب المعدية تجريبياً بالباستيريلال مالتوسيدا.

نصر عبد الوهاب محمد رشا ثابت متولى إبتسام محمد جمال الدين ونها جمال أمين المتولىصقر

1- قسم الباثولوجيا الإكلينيكية، كلية الطب البيطري، جامعة الزقازيق، الزقازيق، الشرقية، مصر.
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الأشنسا هي واحدة من أهم النباتات، وقد استخدمت في الوقاية والعلاج من الأمراض المعدية التي تصيب الجهاز التنفسي. في دراستنا، قمنا بدراسة تأثير الأشنسي على الأرانب المعدية تجريبياً بالباستيريلال مالتوسيدا خلال تقييم خلايا الدم وكمية الدم والقدرة على الإلتهام ومضادات الأكسدة. تم تقسيم أربعين أرنب نيزلندى أبيض من الذكور بالغ متوسط وزنهم (750-1000 جرام) ومتوسط عمرهم شهريًا إلى 4 مجموعات متساوية، وتم الإختلاف بالحمرة الأولي كمجموعة ضابطة، ومجموعة الثانية تلقى عدوى تجريبي للأرناب بالباستيريلال مالتوسيدا (1مللي تحتوي على 710x4 ميكروبك حي) عن طريق الأنف في اليوم الرابع عشر من بدأ التجربة (مجمعا ضابطة معدية) والمجموعة الثالثة تم إعطاء نبات الأشنسي (130ملليجرام/كيلو جرام وزن حي) للأرناب عن طريق الفم يوم بعد يوم من اليوم الأول وحتى نهاية التجربة وتم إجراء عدوى تجريبي بالباستيريلال مالتوسيدا (1مللي تحتوي على 4x710 ميكروبك حي) عن طريق الأنف في اليوم الرابع عشر من بدأ التجربة والمجموعة الرابعة تلقى عدوى تجريبي للأرناب بالباستيريلال مالتوسيدا (1مللي تحتوي على 4x710 ميكروبك حي) عن طريق الأنف في اليوم الرابع عشر من بدأ التجربة وبعد ظهور الأعراض تم العلاج بالإنروفلوكساسين (10ملليجرام/كيلو جرام وزن حي) في ماء الشرب لمدة 3 أيام.

أوضحنا النتائج أن عدوى باستيريلال مالتوسيدا في الأرانب أدت إلى نقص معنوي في عدد كرات الدم الحمراء والهيموجلوبين وحجم الخلايا المضغوطة وحوت الفر من النوع كبير الخلايا الناقصة السمنة وزيادة عدد الكريات البيضاء مع انخفاضًا في مؤشرات الإصابة. لاحظنا انخفاضًا في مستويات البروتين الكلي والألبومين والجلوبينين في الدم. وعلاوة على ذلك، تم تسجيل زيادة كبيرة في إنزيمات الازيمات 아یین ًلا وエステرهايزان والآكاليكالينوفستيوزيز. كماعترف مستوي الصفراء الكلية والباشرة والغير مباشرة في الدم بشكل ملحوظ وتم زيادة مستويات اليوريا والكرياتينين بشكل ملحوظ. علاوة على ذلك، تسجيل انخفاض كبير في إنزيم الكتاليزات معنوية في الملل داي أدليديوجوج الضرور من علاج الإشنسا قبل وبعد عدوى باستيريلال مالتوسيدا في الأرانب قادره على تحسين قياسات جميع العوامل المختبرة مقارنة بالأرانب المعالجة بالإنروفلوكساسين.