

AlQalam Journal of Medical and Applied Sciences Special Issue for 6th International Conference in Basic Sciences and Their Applications (6th ICBSTA, 2023), <u>https://journal.utripoli.edu.ly/index.php/Algalam</u> eISSN 2707-7179

The Potential Protective Effect of Sidr Honey on Some Hematological Changes Caused by Exposure to Cigarette Smoke on Male Albino Rats

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The present study investigated the negative impact of smoking (CS) on some blood parameters in adult male rats and the protective effect of the sidr honey. Twenty four individual were divided into four groups: Normal control (NC), H Group: received Sidr honey orally (100 mg/kg b.w./d.) for 4 weeks, CS group: exposed to cigarette smoke (5 times/d.) for 4 weeks and protective (P) group: received Sidr honey orally (100 mg/kg b.w./d.) for 2 weeks then treated with cigarette smoke for 4 weeks. The results indicated that treatment with cigarette smoke caused significantly decreased (P<0.05) in red blood cells (RBCs), haemoglobin (HB), hematocrit (HCT), mean corpuscular volume (MCV) and platelets (PLT). While, it was a significant increase in white blood cells (WBCs) count compared to control and honey animals. On the other hand, p group showed a slight increase in the mean value of HG, HCT, MCV and PLT, but showed a significant positive decline in WBCs as compared with the CS group. This study indicates that treatment with Sidr honey caused somewhat of an improvement against CS-induced hematological changes in male albino rats.

Keywords: Cigarette smoke, Sidr honey, Hematological changes, rats.

Introduction:

Smoking is known to be a significant risk factor for cardiovascular disease, high blood pressure, infections, stroke, thrombosis, and respiratory disease (de Heens *et al.*, 2009). Moreover, many harmful substances, especially free radicals such as superoxide anions, hydroxyl radicals, H2O2, and HOCL, present in smoke can damage cellular components, leading to serious inflammation, high of white blood cells (WBC), these ROS can damage lipids, proteins and DNA, thus changing the structure and function of cells (Marnett *et al.*, 2003). Folic acid is an essential vitamin. Folate levels have been

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hypothesized to be important in the pathophysiology of many diseases, including neonatal neural tube defects (Butterworth and Bendich, 1996). Smoking addicts have lower levels of folic acid in their blood serum, red blood cells, and respiratory tract (Heimburger et al., 1992; Piyathilake et al., 1994; Giles et al., 1998). Another study also provides evidence that lower folate levels associated with smoke exposure may be important in studies linking smoking to diseases such as breast cancer, colon cancer, and birth defects (Chao et al., 2000). Honey is a natural product with very complex chemical composition, it contains more than 180 substances (Bogdanov et al., 2008; AL-Waili et al., 2012), including, proteins, phenolic, phytochemicals, peroxidase, flavonoids, ascorbic acid minerals, moisture; sugars; enzymes; trace essential elements; vitamins as well as some flavonoids and phenolic acid (Martos et al., 2000; Cooper et al., 2002). And because, there are no scientific reports on the effectiveness of Libyan sidr honey to validate its traditional use on the cure and control of physiological changes in general. Therefore, the current study examined the positive effect of Sidr honey against hematological changes and stressors caused by exposure to smoking in male rats.

Materials and Methods:

Chemicals:

- Libyan Sidr honey. -Karelia red cigarettes.

Experimental animals:

24 adult male albino rats, 10 weeks old weighing 180-200 g were used. Rats were obtained from the animal house of the Zoology Department, Faculty Science, University of Omar Al-Mukhtar, El-Beyda, Libya.

Experimental design:

Rats were randomly assigned into four groups of 6 animals as follows: Group 1: The normal control group (NC), nothing was exposed. Group 2: (H) group, rats were given Sidr honey (100 mg/kg b.w./d.) (Kolawole *et al.*, 2015) orally by gavage for 4 weeks. Group 3: (CS) group, were exposed cigarette smoke by a machine was designed locally in the Zoology Department, Faculty Science, University of Omar Al-Mukhtar, El-Beida, Libya (Figure1). As stated by Alshailabi *et al.* (2023). Group 4: (P) group, rats were given Sidr honey (100mg/kg b.w./d.) orally for 2 weeks then treated with cigarette smoke after taking the Sidr honey for 4 weeks.

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Figure 1: The glass box and smoking machine (Alshailabi et al., 2023).

Hematological analysis:

Blood samples will collect from the orbital sinus. EDTA will use an anticoagulant agent to determine the Red blood cells count (RBC), white blood cells count (WBC), platelet count (PLT), hematocrit value (HTC), hemoglobin level (HB) and mean corpuscular volume (MCV).

Statistical analysis:

Statistical analysis was performed using a computer run package (Graph Pad Prism 7). One way ANOVA followed by Tukey's HSD test was performed to show the statistical significance among the means of the groups. Results were expressed as mean \pm standard error of the mean (SEM). P-value below 0.05 was considered to be statistically significant.

Results:

Red blood cells (RBCs) count:

RBC counts were obtainable in table (1) and figure (2). Statistically, a significant decrease (P<0.05) occurred in the mean value of RBCs in CS group (8.2 \pm 0.22) compare with NC group (9.09 \pm 0.17) and H group (9.98 \pm 0.18). On the other hand, no a significant changes between the mean value of p group (7.89 \pm 0.1) and CS group (8.2 \pm 0.22).

Haemoglobin (HB) level:

The mean values of the HB level were obtainable in table (1) and figure (3). A significant decrease (P < 0.05) occurred in HB level of CS group (19.71 ± 0.56) compare with NC group (22.29 ± 0.85) and H group (25 ± 0.69). While, there is

a slight improvement in p group (20±0.53) compared with CS group Alq J Med App Sci., Special Issue for 6th International Conference in Basic Sciences and Their Applications (6th ICBSTA, 2023), P:588-597, 2/12/2023

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 (19.71 ± 0.56) with CS group in a percentage of increase (1.47%). As in all results, there were not significant changes between the mean values of H group (25 ± 0.69) with NC group (22.29 ± 0.85) .

Hematocrit (HCT) level:

HCT levels were obtainable in table (1) and figure (4). Statistically, a significant decrease (P < 0.05) occurred in HCT level of CS group (28.16 \pm 1.14) compare with NC group (44.6 \pm 0.89) and H group (40.98 \pm 1.02).

No significant differences between P group (30.26 ± 0.46) and CS group (28.16 ± 1.14) . However, slight improvement was observed on the mean value of p group compared to CS group with a percentage of increase (7.46%).

Mean corpuscular volume (MCV) level:

The mean values of the MCV level were obtainable in table (1) and figure (5). There was a significant decrease (P < 0.05) occurred in MCV level of CS group (72.97±1.18) compare with NC group (94.31±0.79) in a percentage of decrease (-22.62%). On the other hand, a significant increase was recorded in P group (79.80±0.45) compared with CS group in a percentage of increase (8.56%).

White blood cells (WBCs) count:

WBC levels were obtainable in table (1) and graphically represented by the figure (6). It has shown very high (P < 0.05) in the CS group (16.8±0.73). Whilst, the P group showed a significant positive decline (P<0.05) in the mean value (10.89±0.28) as compared with CS group, and it was nonsignificant (P<0.05) between the H group (5.42±0.53) and the NC group (6.73±0.53).

Platelets (PLT) count:

The mean values of the PLT level were obtainable in table (1) and figure (7). Statistically, a significant decrease (P<0.05) occurred in PLT level of CS group (98.71 \pm 2.85) compared with NC group (147.43 \pm 4.17), While, P groups showed noticeable improvement in PLT count with percentage of increase (28.80%) compared with CS group.

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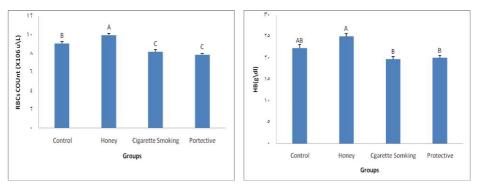


Figure 2: Averages of the mean value of **Figure 3**: Averages of the mean value of the RBCs count (X10⁶ μ l/L). the HG (g/d) level.

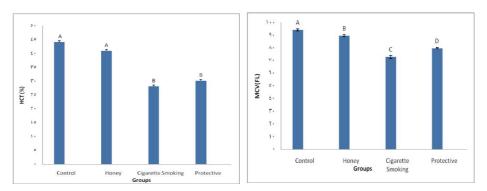


Figure 4: Averages of the mean value of **Figure 5:** MCV (FL) level. Averages of the mean value of HCT level (%).

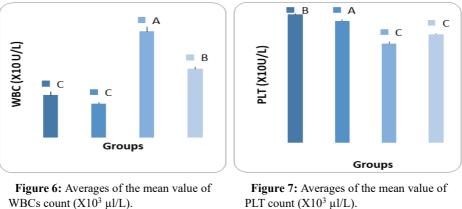


Table 1: Average the mean values of RBC, HB, HCT, MCV, WBC and Platelets levels in

control and experimental groups.

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Parameter	NC	Н	CS	Р
RBC (X10 ⁶ µl/L)	9.09±0.172 ^b	9.98±0.18 ^a	8.2±0.22°	7.89±0.10°
%Of change 1		9.79%	-9.79%	-13.20%
%Of change2				-37.8%
HB (g/dL)	22.29±0.85 ^{ab}	25±0.69ª	19.71±0.56 ^b	20±0.53b
Of change1%		12.16%	-11.57%	-10.27%
Of change 2%				1.47%
HCT (%)	44.6±0.89 ^a	40.98±1.02 ^a	28.16±1.14 ^b	30.26±0.46 ^b
%Of change1		-8.11%	-36.9%	-32.26 %
%Of change 2				7.46%
MCV (FL)	94.31±0.79 ^a	89.69±0.81 ^b	72.97±1.18°	79.80±0.45 ^d
%Of change1		-4.9%	-22.62%	-15.38%
%Of change2				8.56%
WBC (X10 ³ µl/L)	6.73±0.53 °	5.42±0.19°	16.8±0.73ª	10.89±0.28 ^b
%Of change1		-19.5%	149.6%	61.81%
%Of change 2				-35.18%
Platelets(X10 ³ µl/L)	147.43±4.17 ^b	170.57±3.97ª	98.71±2.85°	127.14±2.48°
%Of change1		15.69%	-33.04%	-13.8%
%Of change 2				28.80%

%of change1 = Percentage of change between NC and other groups. %of change2 = Percentage of change between p group and CS group.

Discussion:

In the present study, results showed that cigarette smoke caused changes that vary between significant and non-significant (P < 0.05) in levels of RBC, HB, HCT, MCV PLT and WBC) compared to control and honey animals. This study demonstrates the rats exposed to CS for 4 weeks showed significant disturbances in the levels of hematological parameters which included significant decrease in RBCs, HB, HCT, MCV and PLT. These results were supported by Sherwin and Gastwirth (1990); Siana et al. (1992); Sharif et al. (2014); Alfourti et al. (2021) who presented that CS showed a significant decrease in RBC, HB, HCT, MCV and PLT. Also, this study showed that CS caused a significant increase (P<0.05) in WBC count compared to control and honey animals. These results are in agreement with Noble and Penny (1975); Schwartz and Weiss (1994); Freedman et al. (1996); Blann et al. (1998); AlAwadhi et al. (2008); Aula and Qadir (2013); El- Sawi et al. (2020); Alfourti et al. (2021). The reason for this may be due to Cigarette smoke has a toxic effect on the bone marrow, and therefore it will have immune responses as a result of inflammation after smoking for many years, which can damage all

blood cells leading decreases of RBCs and Hb (Salamzadeh, 2004). Cigarette Alq J Med App Sci., Special Issue for 6th International Conference in Basic Sciences and Their Applications (6th ICBSTA, 2023), P:588-597, 2/12/2023

smoke has 4000 substances among which CO and tars are the main toxic substances. CO can diffuse rapidly across alveolar capillaries, bind firmly to Hb, which can cause a high risk factor for cardiovascular diseases, increase the risk of intravascular clotting, coronary vascular resistance, and decreased coronary blood flow in RBC, PLT, and a predisposition to thrombosis (Richter et al., 2008; Zhong et al., 2008; Ravala and Paula, 2010; Soldin et al., 2011), thus the increasing risk of cardiac disease in smokers may be associated with high fibrinogen levels through arterial wall infiltration and effects on blood viscosity, platelet aggregation, and fibrin formation (Wannamethee et al., 2005). On the other hand, this study demonstrates the rats in the p group showed a slight increase in the mean value of HG, HCT, MCV and PLT, but showed a significant positive decline in WBCs as compared with the CS group, which is in agreement with other studies (Yao et al., 2004; Michalkiewicz et al., 2008) who indicated that the sider honey which contains moisture, sugars such as glucose and fructose, enzymes such as catalase and glutathione reductase, trace essential elements such as iron, copper, zinc, and calcium, vitamins such as vitamin A, C, and E, and some flavonoids and phenolic acids, which leading increases RBC, HB And HCT. However, Sidr Honey has been suggested to protect against lipid peroxidation by reducing the production of lipid hydroperoxides, which leading decreases to inflammation, WBC (Alvarez-Suarez et al., 2012; Hegazi et al., 2017). Folic acid is an essential B vitamin. It is found naturally in Sidr honey and is important in DNA repair. Blood folate levels reflect short-term exposure, while red blood cell levels reflect long-term exposure (Snow, 1999).

Conclusion:

In conclusion, these present findings identify that exposure to cigarette smoke leads to imbalances in the normal range of blood parameters. Moreover, treatment with Sidr honey caused somewhat of an improvement hematological changes in male albino rats.

Acknowledgment:

The authors wish to thank those responsible for the magazine and publishing procedures.

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