STUDY OF THE GENDER RELATED COMPLICATIONS OF EXPERIMENTALLY INDUCED HYPERGLYCEMIA ON SOME VITAL PARAMETERS IN LOCAL RABBITS

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Abstract

The impacts of hyperglycemia on some hematological and serum lipid parameters in rabbits during the seventy days were explored. The parameters assessed incorporate serum lipids, red and white platelet files. Fifty two local breed rabbits, weighing 1.7-2.2 kg was utilized in this study. The rabbits were divided equally into, diabetic group (26 rabbits) and a control group (26 rabbits). Each group in turn was subdivided equally according to gender into, male and female groups. The hyperglycemia was induced by a single injection of alloxan monohydrate at dose 100 mg /kg via the marginal ear vein. The rabbits were divided equally into, diabetic group (26 rabbits) and a control group (26 rabbits). Each group in turn was subdivided equally according to gender into, male and female groups. The blood samples were collected to assert fasting serum glucose and serum lipids, red and white platelet files in periods of 10, 30, 50 and 70 days. The outcomes display that the hyperglycemia affects both genders as the difference was mild in some parameters, especially with regard to lipid variables. A fundamentally diminished packed cell volume, RBCs, and WBCs count in diabetic groups. However, the hyperglycemia had a less considerable influence on MCH, MCHC and MCV. Moreover, the hyperglycemia extensively elevated total cholesterol and serum triacylglycerol levels in the serum whilst it possessed a less significant effect on serum HDL-cholesterol concentration when compared with control groups. In conclusion, the hyperglycemia may also have an impact on serum cholesterol concentration, although it adversely affects some hematological indices.

Keywords: Hyperglycemia, Hematological parameters, Serum lipid, Rabbits

Introduction

Diabetes mellitus, a main non-transferable illness with different etiological agents, influences million of individuals worldwide and according to the World Health Organization (WHO) around 300 million person would encounter the ill effects of diabetes mellitus continuously 2025 and along these lines it considered as one of the five driving purposes behind death on the world (Zimmet 1999; Zheng et al., 2018; Organization, Canada, and Canada 2005; WHO, 2006). Diabetes mellitus is depicted by hyperglycemia in view of unsetting influence in carbohydrates metabolism and defective secretion of insulin, that outcomes in acute and long-term diabetic confusions, which are liable for premature death and disability (Paolisso et al., 1993; Southerland et al., 2006; Pavana et al., 2007).

The hyperglycemia impact the structure and function of all tissues, just as hemopoietic tissue. One of the most clear adverse effect of hyperglycemia is the defect in the ratio of low density lipoprotein LDL/LDL receptor. Author evident case of the functional disorder of the diabetes progresses is the chronic kidney disease (CKD), decreased production of erythropoietin, and anemia (Collaboration 2010; Inzucchi et al., 2011; Association 2014; Reiniger et al., 2010; Collaboration 2010; Webster et al., 2017; Loutradis et al., 2016).

In patients with diabetic nephropathy, the beginning of anemia can manifest from the get-go at some phase in CKD (Hassan 2012). As CKD progresses, anemia typically worsens (Webster et al., 2017). In diabetic patients, the serum creatinine, and albumin excretion rate are the controllers of hemoglobin concentration, proposing that the erythropoietin response in patients with diabetes may be due to early renal interstitial damage or the glycosylation mechanism (Artunc and Risler, 2007).

Anemia is one of the most common complications among individual with type 1 diabetes. People with diabetes who do not follow an appropriate diet are at risk of developing anemia, due to a lack of nutrition or eating inappropriate foods, as they are more likely to have iron and folate deficiency. The state of nephropathy associated with diabetes is one of the most important reasons that necessarily lead to anemia in patients (Shu et al., 2006; Mehdi and Toto 2009; Pyram et al., 2012). The current study aimed to investigate the effect of gender related complications in experimentally induced diabetes mellitus on some hematological parameters in the experimental rabbits.

Materials and Methods

1. The Experimental Animal:

Fifty two local breed rabbits, weighing 1.7-2.2 kg was utilized. Rabbits were housed in confines with suitable dimension (150×100×90), temperature 25±2 ºC and humidity 60%, with standard granulated nourishment, and freely accessible water. The experimental animals were left for two weeks for the purpose of adapting before starting the experiment.

2. Study Design

The rabbits were divided equally into two groups: diabetic group (26 rabbits) and control group (26 rabbits). Each group in turn was subdivided equally according to gender into two groups: male and female groups.
3. Experimental Induction of Diabetes Mellitus

- **Preparation of Alloxan for venous infusion**: Alloxan (Sigma Pharmaceuticals Company) was prepared by dissolving 100 mg of alloxan in 1 ml of 0.9% normal saline (Bhimji, Godin, and McNeil 1985).

- **Injection of Alloxan and post animal care**: A sole injection dose of alloxan monohydrate 100 mg /kg via the marginal ear vein was used for induction of diabetes mellitus in overnight fasting rabbits. Immediately, after injection, 10 ml/20% and 2 ml/5% glucose were given intravenously and orally, respectively, in order to beat the sudden hypoglycemia. While, the control groups were injected intravenously with 1 ml of 0.9 % of normal saline. Moreover, all experimental rabbits was prevented from eating for 12 h, and drinking water was substituted by 5% glucose for 24 h. The procedures of alloxan infusion and blood collection done under kitamine 44 mg /kg-xylazine 5 mg/kg sedation.

4. Collection of Blood

According to the schedule set by the study 10, 30, 50, and 70 days, the blood samples were collected and placed in test tube free of anticoagulant. The blood samples were allowed to clot and the serums were obtained by centrifuging at 4000 rpm for 5 minutes (Parasuraman, Raveendran, & Kesavan, 2010). The clear serum was removed by pipetting and part of the blood serum was used directly for a blood glucose level examination and the other part was preserved at a – 20 °C for the purpose of conducting the serum lipid variables at the end of the study.

5. Estimation of Fasting Serum Glucose

The evaluation of the fasting serum glucose (FSG) in the rabbits of the experiment was conducted three days after the introduction of hyperglycemia. The rabbits were starved for 12 hours before checking. Use the diagnostic kit supplied by the (Spinrecta, Spin), for this purpose. The evaluation of FSG was then carried out at different intervals 10, 30, 50 and 70 days after the introduction of hyperglycemia.

6. Determination of Serum Lipid Parameters

The total serum cholesterol and serum triacylglycerol concentration were estimated by using special kit prepared by (Spinrecta, Spin), while serum HDL -cholesterol concentration was determined by the method of (Assmann, Schriewer, Schmitz, & Hägele, 1983).

7. Determination of Hematological Parameters

The hematomatological parameters including the packed cell volume (PCV) checked by the method of Jain, (1986), red blood cell (RBC) and white blood cell (WBC) counts estimated by the routine haemocytometer method. The platelet count (PLC) was determined by the RBC/Platelet Ratio Method (International Council for Standardization in Haematology, 2001).

The red cell indices were calculated according to the method described by Jain (1986):

- Mean Corpuscular Volume (MCV) = PCV * 10/RBC(fl)
- Mean Corpuscular Haemoglobin (MCH) = Hb * 10/RBC (pg)
- Mean Corpuscular Haemoglobin concentration = Hbc = 100/PCV

**Statistical Analysis**

The outputs presented as mean ±SD were analyzed using the Duncan Multiple Range test and differences at P< 0.05 were considered significant (Montgomery 2017). The SPSS software version 22 was used for the purpose of conducting various statistical analyzes (SPSS, 2012).

**Results and Discussion**

The effect of hyperglycemia on some serum lipid indices is presented in (Figures: 1, 2 and 3). Hyperglycemia significantly (P<0.05) caused increased serum total cholesterol concentration and posed a significant effect (P<0.05) on serum HDL-cholesterol concentration compared to controls. Furthermore, serum triacylglycerol concentration was notably elevated (P<0.05) in diabetic groups.

As per various investigations, high blood cholesterol concentration is one of the significant hazard factors for cardiovascular disease (Jackson et al., 2005). Al-Karagoly (2007) uncovered that the most significant gross appearance at necropsy after the experimental induction of hyperglycemia in local rabbits was cardiac hypertrophy. Hence the rise in serum total cholesterol fixation instigated by diabetes mellitus expanded the danger of cardiovascular disease (Lorenzo et al., 2007). The increment in serum total cholesterol concentration is because of increment in serum HDL-cholesterol concentration. It accordingly infers that height in different fractions of serum total cholesterol concentration other than HDL-cholesterol might be liable for the rise of serum total cholesterol concentration watched.

The present examination shows that the two genders are influenced by the hyperglycemia, the most elevated impact stays in females contrasted with the male, because of the system of lipid metabolism in females. Where, increasingly checked collection of fat in the intra- abdominal visceral fat depots of men. Besides, circling blood lipid concentrations likewise show gender-related contrasts on the grounds that the physiological properties of the female that identified with ovulation and pregnancy, though complete cholesterol, LDL-cholesterol and triacylglycerol focuses are lower and HDL-cholesterol focus is higher than in male (Williams, 2004).

As saw in (Figures: 4, and 5), hyperglycemia fundamentally (P<0.05) diminished RBC includes and PCV in diabetic gatherings, when compared to control groups. Be that as it may, MCH, MCHC and MCV were not fundamentally modified by diabetes mellitus when matched to control groups (Figures 6, 7, and 8).

Since MCHC, MCH and MCV relate to individual red blood cells, while RBC and PCV relate to the total population of red blood cells in the blood, in this manner infer that the diabetes mellitus may neither influence the incorporation of hemoglobin into red platelets nor the morphology and osmotic fragility of red platelets produced (Blaslov et al., 2019). In any case, the decrease in RBC and PCV suggests that the hyperglycemia diminishes the number of population of red blood cells from the bone marrow (US, 2019). The hyperglycemia might impact the oxygen binding capacity of individual red blood cell, but may reduce the oxygen- carrying capacity of the whole blood due to his influenced on RBCs indicies, as well as population of red blood cells and anemia occur (Seeley et al., 2005; US, 2019; Barrett et al., 2010; Ali and Hassan 2019).
Hyperglycemia significantly reduced (P<0.05) WBC counts when compared with control (Figure 9). This indicates that the hyperglycemia may prompt development of some bioactive agents that could cause decimation or impaired production of white platelets (Barrett et al., 2010 and Mishra 2013). It can be concluded that the direct effect of diabetes on white blood cells comes from its action on the granulocyte-macrophage colony stimulating factor, macrophage colony stimulating factor, interleukins IL-2, IL-4 and IL-5 that regulate responsible for the proliferation, differentiation of white blood cells (Grossmann et al., 1996, Pennig et al., 2019; Banerjee and Saxena 2012; Hall 2015). Thus hyperglycemia may incline to reduce immune response and thus, enhances the possibility of developing various infections (Weekers et al., 2003).

The reduction of WBC and at all periods of hyperglycemia and the RBC at the late period of experiment without corresponding reduction in platelet count, suggest that the hyperglycemia may possess the potential of causing a progressive but selective bone marrow depression with increasing time (Marles and Farnsworth 1995, Kornicka et al., 2018). Likewise the production of these components of blood might be vulnerable to regulation by the hyperglycemia in an order having white blood cell production as the most susceptible and platelet production as the least susceptible (Malomo et al., 2002; Carrizzo et al., 2018).

Conclusion

It was inferred that hyperglycemia, antagonistically influence some hematological indices, particularly those identifying with red platelets and white platelets, as well as special effect on serum cholesterol concentration.

Ethical Approval

All applicable international, national, and/or institutional guidelines for the care and use of animals were followed. All procedures performed in studies involving animals were in accordance with the ethical standards of the institution or practice at which the studies were conducted.

Disclosure of potential conflicts of interest and current submission

This manuscript has not been previously published and is not under consideration in the same or substantially similar form in any other peer-reviewed media.
Fig. 6: Effect of the induced hyperglycemia on MHC (Pg) in the control and treated groups of the female and male rabbits. (n=52)

Fig. 7: Effect of the induced hyperglycemia on MCHC in the control and treated groups of the female and male rabbits. (n=52)

Fig. 8: Effect of the induced hyperglycemia on MCV (fL) in the control and treated groups of the female and male rabbits. (n=52)

Fig. 9: Effect of the induced hyperglycemia on WBCs (x10/L) in the control and treated groups of the female and male rabbits. (n=52)

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References


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