

Assessment of Hematological and Biochemical Markers in the Early post-Burn Period for Predicting Recovery and Mortality in Animals with Burn Injuries

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Abstract

Here we investigate hematological and biochemical alterations in the burn animals. The values of these parameters are very important in allowing the veterinarians to know how the animal body is responding to the different therapies that being provided? And this will help the veterinary medical staff for proper management with less morbidity and mortality. Three burned animals were admitted to the University's Veterinary Hospital, Faculty of Veterinary Medicine, South Valley University. They were suffering from second to third degree burn injury. Blood samples were collected after 48 h post burn for hematological and biochemical analyses. For the hematological parameters, RBCs count, hemoglobin (Hb) concentration, packed cell volume (PCV%) and platelets count exhibited significant decreases in burnt animals in comparison to healthy control animals. Burns also induced increases in WBCs count, MCV and MCH in comparison to the controls. For the differential leukocytic count, lymphocytosis and neutrophilia were recorded in burned animals. For biochemical parameters, liver enzymes activities as well as serum creatinine and urea were significantly increased when compared with the control animals. The total serum protein and albumin of burn animals revealed significant decrease. The serum electrolytes, serum sodium showed a significant decrease while serum potassium showed significant increase.

Keywords: Burns; Biochemical; Hematological; Electrolytes; Health care team; Proper management

Introduction

The skin is one of the largest and most protective organs of the body. The animal skin has a surface area of 3.70 to 4 square meters and forms the major interface between the internal organs and the external environment. The skin is continuously exposed to many harmful materials, including liquids, gases, solid matters, sunlight, and microorganisms. Furthermore, skin acts as an immunological barrier where it contains the macrophage cells (Langerhans cells) which detect harmful antigens, presenting protective effect in allergic dermatitis and skin graft rejection [1,2]. Burn is considered as one of the most common and highly destructive forms of skin trauma [3]. It causes damages to the skin by destroying the skin's tissues. Coming to the skin in contact with the source of injury such as flames, electricity, chemicals and radiation resulted in the burns. Thermal burns are caused by external sources of heat, such as flames, scalding liquids, or steams [1]. The severity of any burn injury is related to the burn's size and deepness and related also to which part of the body that has been burned. Expecting the burn-related mortality is depending on the specialized care, and the type and probability of complications of the burn, as well as the burn proportion to the animal's total body surface area (TBSA) [3]. Burns classification depends on the depth of the wound as in the first-degree burns the epidermal layers of the skin only are affected. In the second degree burns the skin's superficial and deep partial-thickness are included. Third-degree burns all layers of the skin

are injured, while in the fourth-degree burns the full thicknesses of the skin with underlying structures are injured [4]. Decrease the morbidity and mortality from the major burn in the veterinary society was achieved by the good progress in trauma and burn management which resulted in improved the animal's survival. The improvement in the survived burnt animals caused by the advancements in the revival, well advanced surgical techniques, prevent and control the infection and nutritional support [1].

The most common effects of thermal injury may be both local and systemic. As response to the burn injury the body exhibited different responses includes hypermetabolism, prolonged catabolism, organ dysfunction, cellular protection mechanisms, inflammations and immunosuppression [5]. Burned animals commonly exhibit an inflammatory changes not only in the burn area but also involved in the entire organs (kidney, liver and other organs), which summarized in the term of systemic inflammatory response syndrome (SIRS). Burn sepsis with SIRS are major indicators for detecting the morbidity and mortality rate in thermally injured animals [4]. Larger surface body burns are characterized by marked hypermetabolic response and marked losses in the total body mass. The hypermetabolic responses induced by burns are related to a progressive decrease in the host defenses mechanisms through the immunological changes [6]. The state of immunosuppression induced by thermal injuries induced predisposes burn animals to infectious complications and risks of sepsis [3]. Many veterinary surgeons feel uncomfortable conditions in managing the animals with major thermal trauma, although burn injuries are frequent in our veterinary society. These conditions are because of the high morbidity and mortality rates. In this study we

estimate some hematological and biochemical alterations in the burn animals whom admitted to University Hospital. This will help the veterinary medical staff for proper management with less morbidity and mortality.

Materials and Methods

There 3 burned cattle were admitted to the University's Hospital, Faculty of Veterinary Medicine, South Valley University. The burnt animals were clinically investigated by specialist veterinarians and were classified between first and third degree burns. Blood samples were collected carefully after 48 h post burns. Five ml of blood was drawn from the three burnt animals and three other samples were collected from healthy control animals for hematological and biochemical study. The blood samples were divided in two portions; the first portion withdrawn on tubes contains EDTA as anti-coagulants to prevent clotting of blood to be used for hematological studies. The second portion was added in plain tubes without anti-coagulant, for obtaining serum for subsequent biochemical tests. The serum samples were frozen at -20°C for biochemical analysis [7].

Complete blood count

The whole blood samples were analyzed using an automatized blood analyzer (Urite, China) for red blood cell count (RBCs), hemoglobin (Hb) concentration, packed cell volume (PCV) percentage, white blood cell count (WBC), thrombocytes count and blood indices, mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), and mean corpuscular volume (MCV). The Peripheral blood films smear was examined. Differential white cell counts were performed on blood films stained with Giemsa stain.

Serum biochemical analyses

Liver function test: Determination of alanine aminotransferase (ALT), Aspartate aminotransferase (AST), alkaline phosphatase (ALP), total bilirubin, total protein and albumin was performed according to the manufacturer's protocol of reagent kits purchased from bio diagnostics, Egypt.

Kidney function test: Determination of serum creatinine, blood urea nitrogen (BUN) and uric acid were performed according to the manufacturer's protocol of reagent kits purchased from Bio diagnostics, Egypt.

Electrolytes detection: Determination of serum sodium and potassium according to the manufacturer's protocol of reagent kits purchased from Human Company, Germany.

Results

Hematological studies

Peripheral blood film smear: Examined blood films exhibited poikilocytosis. The common cells observed were spherocytes, echinocytes, keratocytes and elongated cells. Also there were many cells with vesicle formation as well as the presence of fragmented erythrocytes as shown in Figure 1.

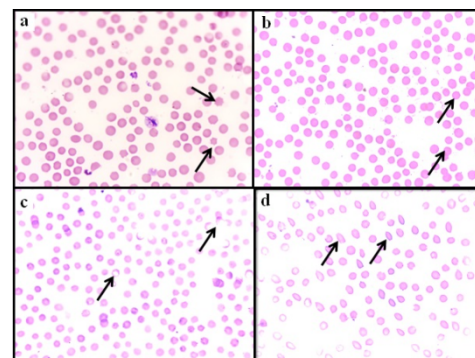


Figure 1: Blood film smear (A, B, C & D) showing red blood cells morphological changes in burn animals where A shown echinocytes Figure 1A, spherocytes Figure 1B, keratocytes Figure 1C and elongated cells Figure 1D (Giemsa stainX1000).

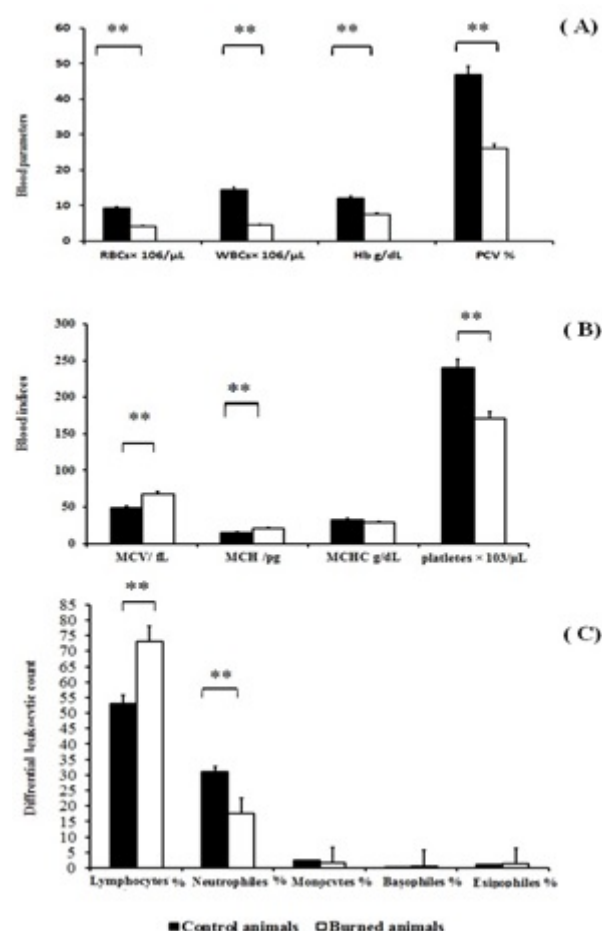


Figure 2: Hematological changes (A), blood indices (B) and differential leukocytic count (C) in burn animals 48 h post thermal injury 48 h post thermal injury in comparison to healthy animals. Mean ± SE **P<0.05.

Blood parameters: Figures 1 and 2 exhibited the blood parameters of burned cattle 48 h after exposure to thermal injury. We noticed significance decrease in RBC's count, HB concentration and PCV% and thrombocytes count. Furthermore, burns induced significant increase in the WBC's count, MCV and MCH levels while the increase in MCHC level was proved non-significant in comparison to healthy controls. For differential leucocytic count, thermal injury induced lymphocytosis and neutrophilia. On the other hand, no changes were seen in the count of both basophiles and Eosinophils in comparison with the control group according data in Figure 3.

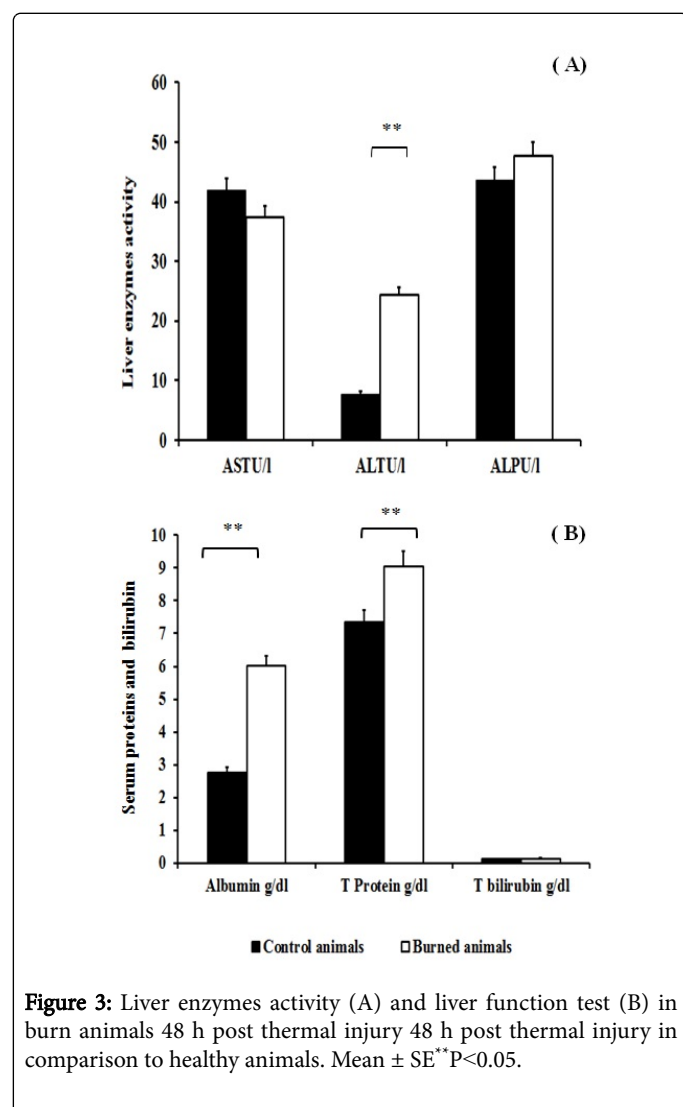


Figure 3: Liver enzymes activity (A) and liver function test (B) in burn animals 48 h post thermal injury in comparison to healthy animals. Mean $\pm$ SE **P<0.05.

Biochemical analyses

Liver function analysis: Liver enzymes activity were analyzed where there was increase in AST, ALT, ALP enzyme activities which proved highly significant in ALT enzyme when compared with control animals. In addition, burns induced significant decrease in serum total protein and albumin and total bilirubin which, proved non-significant when compared with normal control animals Figure 4.

Kidney function analysis: As shown in Figure 5, burns induced significance increases in serum creatinine and blood urea nitrogen

while non-significance increase was recorded in uric acid in comparison to control group.

Serum electrolytes: Burns also lead to significant increase in potassium levels, while sodium level was significantly decreased.

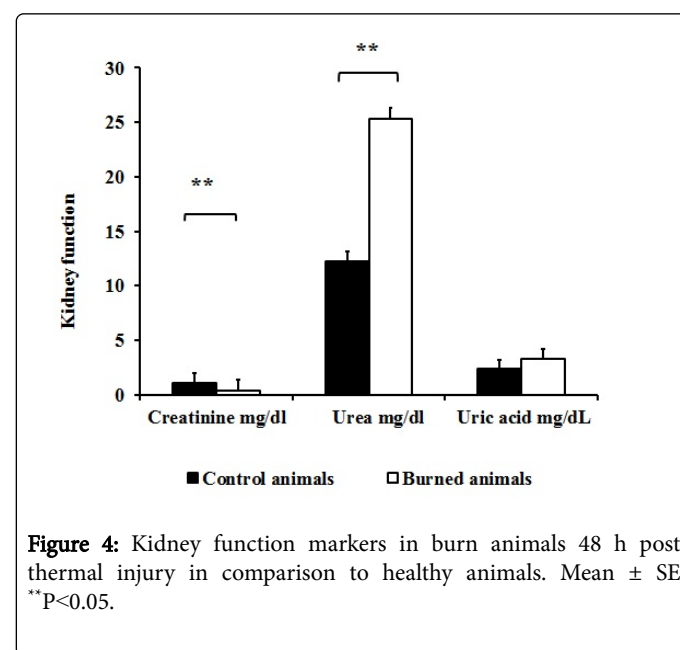


Figure 4: Kidney function markers in burn animals 48 h post thermal injury in comparison to healthy animals. Mean $\pm$ SE **P<0.05.

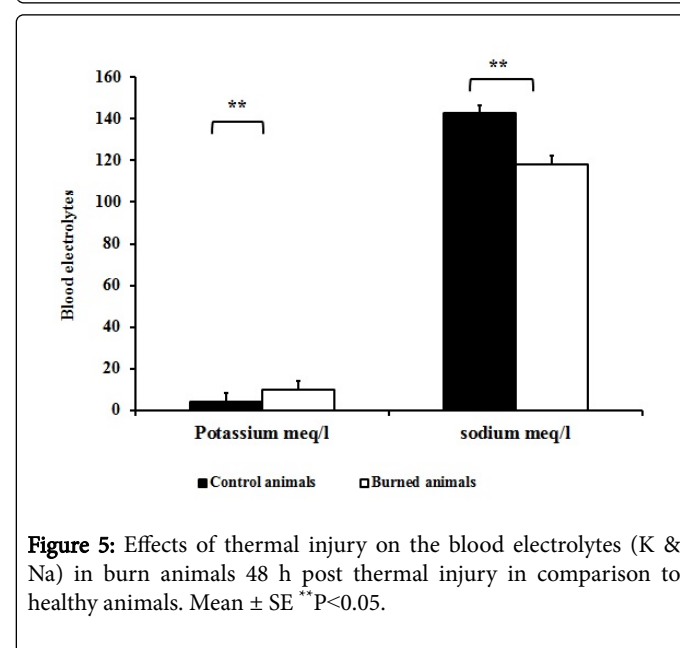


Figure 5: Effects of thermal injury on the blood electrolytes (K & Na) in burn animals 48 h post thermal injury in comparison to healthy animals. Mean $\pm$ SE **P<0.05.

Discussion

This study is aimed to estimate some hematological and biochemical changes in the burn animals, The levels of these parameters are very important in letting the veterinarian care team to know how the body is responding to the thermal injury and the response to the different therapies that being provided. For the hematological changes in the peripheral blood film smear many misshaping cells were observed. Echinocytes, Keratocytes and spherocytes were the dominant cells observed. The abnormal red cell morphology (paikilocytosis)

resembles echinocytes, a form which is known to be due to depletion of ATP concentration in the erythrocytes [8]. RBC's count, HB concentration and PCV % values were significantly decreased in association with significantly increased in WBC's count as shown in Figure 1. Our results come in agree with EI-Sonbaty & EI-otiefy [8]. Anemia has been assumed in injured animals being due to the accelerated erythrocytes decomposition. The decreasing of hematocrit is to be expected with inadequate fluid resuscitation, and may also be a hallmark of occult bleeding [9].

Significant leukocytosis was noticed in burned animals in this study. These findings are consistent with many reports. Gruber and Farese [10] reported that a peripheral leukocytosis lasted for 35 days or more in murine granulopoiesis after inducement of a standardized sublethal third-degree burn. A similar result was reported by D'Alesandro and Gruber [11] in an experimental study on rats, where after a 30% thermal injury leukocyte quantities were three to five times normal values. Burns induced significant increase in the MCV and MCH values. The increase in MCV and MCH values was attributed to hemolytic anemia and liver dysfunction [12]. In addition, there was significant decrease (thrombocytopenia) in PLT count when compared with control animals confirm data obtain by Fiebach et al. [13].

Thrombocytopenia was due to increase the platelet consumption for occult bleeding, dehydration and reduced thrombopoietin production due to liver failure. For differential leukocytic count, the data in Figure 3 exhibited thermal injury induced lymphocytosis and neutrophilia. These findings are agreed with those of Marina and Lara [14]. The increase in the lymphocytes attributed to increase the histamine release, which play role in increase the vascular permeability post burning [15], while increase in the neutrophils % was due to sepsis occurred after burns [16]. To better understand, the alteration expected in the internal organs biochemical analysis was done for detection liver and kidney function markers. We have obtained elevated liver enzymes AST, ALT, ALP. Those findings were reported by previous studies [17,18]. Burns lead to an increase in edema formation that in turn leads to cell damage, with the release of hepatic enzymes [19]. The increase in the liver enzymes activities was due to the systemic inflammatory syndrome induced by thermal injury. Importantly, total protein and albumin decreased in burn animals. This study was supported by Demling [20] who recorded that marked decreased in plasma proteins occurs early post burn.

Other study also pointed out that burns injury results in dramatic changes in plasma proteins in which the concentration of protein was significantly lower in serum of burn animals than the control [21]. These results occur because local inflammatory cytokines enter the circulation and result in systemic inflammatory response. As burns approach 25% of TBSA, this will lead the microvascular escape to become generalized and permit the loss of fluid and protein from the intravascular to the extravascular spaces and finally they are lost through the wound [3].

The serum sodium of burnt animals was significantly decreased in comparison to healthy controls. This results agreed with Huang et al. [22] who states that during the first 3 days after burn, serum sodium levels were moderately elevated, as well as, these results were supported by Ge et al. [23] who explained that serum Na⁺ decreased post-burn and increased after resuscitation. The full explanations for these results are in the major burns, the intravascular volume is lost in burned and unburned tissues. Losing of the intravascular volume is due to increase the vascular permeability as well as the interstitial osmotic pressure in burn tissue and cellular edema developed.

The concentration of serum potassium is significantly increased in burnt animals. Our data are supported by other study, which states that in major burns, the initial resuscitation period (between 0 and 36 h) characterized by hyperkalemia because of the massive tissue destruction and necrosis [24]. As well as, Demling et al. [20] stated that potassium ions will increase if severe hemolysis has occurred or renal impairment is present. Our results reveled also increase in the level of urea and creatinine, the markers of kidney failure this come in agreement with Anandani [18]. Multiple conditions contribute to renal failure in the burn animals such as hypovolemia, cardiac dysfunction, release of inflammatory mediators and denatured proteins (from extensive tissue destruction), and nephrotoxic drugs [25,26]. Finally, the increase or decrease in the hematological and biochemical parameters may be due to the hypermetabolic state of the body which resulted from the increase in the adrenaline release, loss of fluid and electrolytes, hemolysis and sepsis.

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