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EFFECT OF POST FOOT AND MOUTH DISEASE INFECTION ON SOME SEMEN ATTRIBUTES AND BLOOD BIOCHEMICAL PARAMETERS OF HOLSTEIN BULLS

[1] Sajeda Mahdi Eidan, [2]Faris Feisal Ibrahim, [1]Talal Anwer Abdulkareem & [2]Kamil Fayadh Hurish
[1]Department of Animal Production, College of Agriculture, University of Baghdad, Iraq.
[2]Artificial Insemination Department, Ministry of Agriculture, Iraq.

ABSTRACT
This study was undertaken to investigate the influence of 14 months- post foot and mouth disease (FMD) infection on some semen characteristics (mass activity, individual motility, post-thawed individual motility) and blood biochemical parameters (plasma glucose, cholesterol, AST, ALP, total protein, albumin and globulins quantities, albumin and globulins percentages, albumin to globulins ratio, Na and Mg) of Holstein bulls. This study was carried out at the Artificial Insemination Department, Ministry of Agriculture; Iraq using ten intact and 21 post FMD infected bulls. Four weekly ejaculates and blood samples were collected from each healthy and post FMD infected bull for this purpose. Sperm mass activity, individual motility and post-thawed individual motility were declined ($P \leq 0.01$) in post FMD infected bulls as compared with healthy bulls. Plasma glucose, albumin percentage, albumin to globulin ratio and Mg concentrations were lowered ($P \leq 0.01$) and plasma AST, ALP, total protein, globulins quantity and percentage and Na concentrations were higher ($P \leq 0.01$) in post FMD infected bulls as compared with healthy one. In conclusion, 14 months post FMD infection had residual negative effects on semen quality and blood biochemical parameters of Holstein bulls. These have a negative impact on post-freezing semen quality and cow's fertility rates consequently.

KEYWORDS: FMD, Semen, Blood, Holstein bulls.

INTRODUCTION

Footand mouth disease (FMD) is a highly contagious viral vesicular disease of cloven-hoofed animals, characterized by fever, depression, anorexia and excessive stingy or foamy salivation with vesicles or blisters appearing on the tongue (Kitching, 2002, Barkakati *et al.*, 2015). Mortality loss, salvage sale loss, and milk loss accounted for the greatest percentage of the total economic cost due to FMD in small, medium, and large herds, respectively (Jamal and Belsham, 2013, Baluka, 2016). Bhakat *et al.* (2008) reported that application of FMD vaccine has significant ($P \leq 0.05$) adverse effect on most seminal attributes during post vaccination period in Sahiwal bulls. In Iraq, Al-Badry *et al.* (2012) demonstrated that FMD had negatively affected semen quality in Holstein bulls. This prompted us to explore the effect of 14 months post FMD infection on some semen characteristics and blood biochemical parameters of Holstein bulls.

MATERIALS & METHODS

Experimental animals and design

This study was carried out at the Artificial Insemination Department pertaining to the Directorate of Animal Resource / Ministry of Agriculture, Abu-Ghraib, 25Km west of Baghdad, Iraq during the period from October 2014 to February 2016 using ten intact and 21 fourteen months-post FMD infected bulls of 2.5-3 years old and 500-750kg live body weight. Four weekly ejaculates and blood samples were collected from each intact and post FMD infected bull for this purpose. Serum was harvested from blood following centrifugation of the sample (1409g for 15 min.) and was stored at -20˚C until assay. Semen was collected from each bull separately. Mass activity was determined according to Rollinson *et al.* (1970) and Salisbury *et al.* (1978) by placing a drop of semen on a warmed microscope slide with cover slip and examining the edge of the drop (x 100 power). Ejaculate mass activity was estimated based on speed of wave motion. Fresh semen and post-thawing Individual motility was determined for fresh and frozen semen according to Walton (1933) procedure. One drop of semen was mixed with one drop of sodium citrate (2.9%) solution, put on warmed slide with a cover slip and an examined under light microscope (×400). Glucose (Cooper, 1973) and cholesterol (Allain *et al.*, 1974) concentrations were quantitatively determined. Total protein was assessed using Biuret's method (Green *et al.*, 1982). Bush (1998) was used for quantitative determination of albumin concentration. A Globulins concentration and albumin to globulins ratio were assessed according to Otto *et al.* (2000). Aspartate aminotransferase (AST) activity was assessed using Reitman and Frankel (1957) method. Kind and King (1954) was used to estimate alkaline phosphatase activity (ALP). Serum sodium and magnesium were measured using a Perkin-Elmer Model 5100 PC atomic absorption spectrophotometer (Shelton, CT).

Statistical analyses

Statistical analysis was performed using general linear model (GLM) procedure in the SAS program (SAS, 2012) to investigate the effect of status (Intact and post-FMD infection) on some fresh and post-thawed semen

characteristics as well as blood biochemical parameters. The statistical model for analysis of variance was:

$Y_{ij} = \mu + S_i + e_{ij}$

Where:

Y_{ij} = dependent variable (Mass activity, individual motility, glucose, cholesterol, total protein, albumin, globulins, albumin: globulins ratio, AST, ALP, Na, Mg).

μ = Overall mean.

S_i = effect of status (S= Intact and post-FMD infection).

e_{ij} = Error term.

Differences among means were computed using the Duncan multiple range test (Duncan, 1955).

RESULTS & DISCUSSION

Semen characteristics

Higher (P≤0.01) fresh semen mass activity was noticed in intact bulls as compared with post FMD-infected ones (Table 1). Concomitantly, fresh sperm individual motility took similar trend, being higher (P≤0.01) in intact than post FMD-infected bulls (Table 1). On the other hand, intact bulls exhibited an obvious (P≤0.01) post-thawed sperm individual motility as compared with post-FMD infected bulls (Table 1). These results were agreed with those observed by Al-Badry *et al.* (2012) who found a deteriorated semen properties two and four months post FMD infection forhighly and medium efficient-Holstein bulls respectively in comparison with pre-FMD infection. These results were also in line with Bhakat *et al.* (2008) who noticed decreased in mass activity percentage following FMD vaccination in Sahiwal bulls. FMD infection caused rise in body temperature, leading to the deleterious influence on both the process of spermatogenesis (Anderson, 2001).

TABLE 1: Sperm mass activity, individual motility and post-thawing motility of healthy and post food and mouth disease-infected bulls (Mean ± SE).

Parameters / Group	Mass activity (%)	Individual motility (%)	Post-thawing individual motility (%)
Healthy (n=10)	66 ± 1.8a	69.5±2.2a	56.5 ±1.8a
Post FMD infection (n=21)	45.7 ± 4.8b	54.5 ± 4.0b	32.4 ± 5.7b
Level of significance	P≤0.01	P≤0.01	P≤0.01

a, b, Mean±SE with different superscripts in the column differ significantly (P ≤ 0.01).

Blood biochemical parameters

The concentration of different serum biochemical constituents (mean ±SE values) are presented in Table 2. Serum glucose concentrations were lower (P ≤ 0.01) in post FMD infected bulls than those of intact bulls. This result was disagreed with those of Barkakati *et al*. (2015) who noticed declining in serum glucose of FMD-infected cows as compared with recovered and apparently healthy once. Decreased glucose levels could be due to the hypocalcaemia which stimulate the secretion of insulin from the pancreas during and post FMD infection (Kaneko *et al*., 1997). The average values of serum cholesterol in post FMD-infected bulls were similar between the two groups. It is known that conditions such as pancreatic dysfunction that decrease cholesterol synthesis will also decrease insulin synthesis, resulting in hypocalcaemia, hypoproteinaemia and hyperglycemia (Kaneko *et al.*, 1997). The serum AST activity was significantly (P ≤ 0.01) lower in intact than in the post FMD-infected bulls. The post FMD-infected bulls exhibited higher (P ≤ 0.01) ALP activity than intact bulls. Elevations in ALP are usually secondary to tissue damage. Concentration of total protein is significantly (P ≤ 0.01) increased in post FMD-infected bulls as compared with intact bulls. The decreasing in the total serum protein concentration in the intact bulls might be due to the anorexia and may result from insufficient intake of protein. Roussel *et al.* (1997) stated that decreasing of total protein
concentration is associated with hepatic and renal damage, starvation; enteropathies resulting in protein loss. This result was disagreed with those of obtained by Barkakati *et al.* (2015) who observed decreased in total protein concentration in FMD- affected animals. Similarly, serum globulins concentrations and percentage were increased (P ≤ 0.01) in post FMD-infected bulls as compared with intact bulls. Higher globulins concentrations in FMD affected animals may return to the liver and kidney damage caused by FMD infection (Roussel *et al.*, 1997). This protein is important in preventing and fighting infection. Some gamma globulins bind to foreign substances (such as virus or bacteria) and destroying them by the immune system. Serum albumin concentrations did not changed between post FMD infected and intact bulls, however, albumin percentage was higher (P≤ 0.01) in intact bulls in comparison with FMD infected one. This may reflected the liver (cirrhosis) and kidney damage of FMD affected bulls. Albumin is the most abundant protein produced in the liver, constituting about half of the blood serum protein, responsible for hormones and fatty acids transport. Accordingly, the albumin: globulin ratio is greater (P ≤ 0.01) in intact as compared with FMD-infected bulls. Post FMD infected bulls exhibited higher (P ≤ 0.01) Na concentrations than intact bulls. In contrast, Mg was lower (P≤ 0.01) in post FMD infected bulls than intact bulls. Increasing serum Na concentrations (hypernatremia) and decreasing of serum Mg (hypomagnesaemia) in FMD-affected bulls resulted from dehydration as a result of kidney disorder (Kaneko, 1997). The biochemical alteration indicates the development of pancreatic dysfunction in post FMD affected bulls.

TABLE 2: Some blood biochemical parameters of healthy and post Food and mouth disease-infected bulls (Mean ± SE).

Parameters \ Group	Healthy (n = 10)	Post FMD Infection (n= 21)	Level of significance
Glucose (mg/dl)	70.1 ±1.8[a]	42.9± 2.9[b]	P≤0.01
Cholesterol (mg/dl)	96.4 ± 6.5[a]	88.5 ± 5.2[a]	NS
AST (U/ L)	68 ± 2.3[b]	106.1 ±8.2[a]	P≤0.01
ALP (U/ L)	66 ± 7.9[b]	136.7 ± 6.6[a]	P≤0.01
Total protein (g/ L)	69.4 ± 1.4[b]	91.9 ±1.3[a]	P≤0.01
Albumin (g/ L)	32.2 ±0.3	31.1 ± 0.4	NS
Globulins (g/ L)	37.2 ±1.3[b]	60.7 ± 1[a]	P≤0.01
Albumin (%)	46.5 ±0.9[a]	33.9 ± 0.4[b]	P≤0.01
Globulin (%)	52.5 ±0.9[b]	66.4 ± 0.4[a]	P≤0.01
Albumin:globulin ratio	0.87 ± 0.03[a]	0.51 ± 0.0[b]	P≤0.01
Na (mmol/l)	130.4 ± 0.4[b]	140.2 ± 0.6[a]	P≤0.01
Mg (mg/dl)	3.8 ± 0.1[a]	2.5 ± 0.13[b]	P≤0.01

NS = Non-significant

a, b, Mean±SE with different superscripts in the column differ significantly (P ≤ 0.05)

CONCLUSION

14 months post FMD infection had residual negative effects on semen quality and blood biochemical parameters of Holstein bulls. These have a negative impact on post-freezing semen quality and cow's fertility rates consequently.

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