EFFECT OF MORINGA OLIFERA LEAVES AND SEEDS SUPPLEMENTATION ON SEMEN CHARACTERISTICS, TESTES AND EPIDIDYMIS BIOMETRY AND SOME BLOOD PARAMETERS IN BARKI RAMS.

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ABSTRACT: This study aimed to investigate the effect of MOL or MSC supplementation on some blood constituents, semen characteristics, testis and epididymis measurements in Barki rams. Fifteen Barki rams aged 10-12 months divided into three similar groups (5 each). Rams in G1 were fed the basal diet supplemented by MOL (5% of CFM), rams in G2 fed the basal diet supplemented by MSC (10% of CFM) while rams in G3 (control) were fed the basal diet without supplements. Blood samples were collected and erythrocytes, Hb, PLT, PCV, MCV, MCH, MCHC, ALT, AST, glucose, uric acid, creatinine and testosterone were determined. Semen was collected and semen physical characteristics as well as of the epididymis and testes biometric were evaluated. Results indicated that MOL and MSC supplementation to ram rations significantly affected glucose, creatinine, urea, ALT, AST and testosterone levels, and insignificantly on erythrocyte, Hb, PLT, PCV, MCV, MCH, and MCHC.

In this regard, MOL supplementation to ram rations increased MCV, PLT and decreased RBCs, Hg, MCH, MCHC, and PCV; while, MSC supplementation increased RBCs, Hg, MCHC, PLT, and urea. The addition of MOL or MSC increased ALT, AST, and creatinine, and decreased testosterone and glucose levels.

Both supplementation (MOL or MSC) in Barki rams' feed improved most characteristics of semen such as volume, motility, vitality and normality, and reduced mortality and abnormalities and there is no significant effect on testes measurements. The highest testicular weight and width were observed in the MOL group. The testicular size was relatively higher for rams fed a moringa-free diet compared to those supplemented with MOL or MSC. It could be concluded that supplementation of MOL and MSC rations into Barki rams' rations improved most of the semen characteristics (volume, motility, vitality and normality) and reduced mortality and malformations. However, it negatively affects kidney function (increased urea and creatinine) and liver enzyme activities (ALT & AST).

Key words: Barki rams, semen characteristics, Moringa leaves (MOL), Moringa seeds (MSC).

INTRODUCTION

Ram's reproductive ability has a significant impact on herd reproductive performance and profitability (Shi et al., 2010). Nutrition is one of the various factors affecting male fertility, semen quality and the ability of sperm to fertilize (Canyurt & Akhan 2008; Lahnsteiner 2009; Martin et al., 2010). In this regard, Wafa et al., (2017) explained that nutrition regulates sperm production, gonadotropin secretion, and male sexual development.

Moringa oleifera (MO) is one of thirteen known species belonging to the family Moringaceae that grows in many countries (Olson, 2002, Cuellar-Nuñez et al., 2018), and is a good source of high-quality feed for farm animals such as cattle, buffalo, rabbits, goats and sheep (Fahey, 2005; Nayel, 2021 and Syarifuddin et al., 2022). In addition, its various parts have beneficial properties and are recognized as nutritional and medicinal value (Abdel-Rahman et al., 2018; Dhakad et al., 2019 and Liang et al., 2019). Several chemical compounds have been isolated from Moringa leaves such as flavonoids, phenolic acid, glucosinolates, and isothiocyanates, as well as
possessing many biological activities, including hypocholesterolemic, antidiabetic, (Anwar et al., 2007, Bichi, 2013, Baldisserotto et al., 2018 Dhakad et al., 2019 and Nayel, 2021). At the same time, Jaiswal et al., (2009) and Mahfuz & Piao., (2019) reported that the extract of M. oleifera supplementation showed a great benefits on blood metabolites and a strong immune modulating effect, can modulate B-cell activation and stimulate immunoglobulins production (Ojeka et al. 2016 and Nayel, 2021). It is indicated that Moringa oleifera affects male reproductive capacity, as it contains several important factors for spermatogenesis such as the minerals zinc and selenium (Cheah and Yang, 2011), the amino acid arginine (Numan et al., 2014) as well as vitamins A, C, E (Agarwal, 2014). In addition, it includes the putrescine precursor and spermidine which are important for sperm motility (LeFèvre et al., 2011), along with the long-chain polyunsaturated fatty acid (PUFA/PUFA) α-linolenic (Moyo et al., 2011) that maintain sperm cell survival, maturation and fertility (Conquer et al., 2000). It could be suggested that Moringa Oleifera leaves enhances the male reproductive potentials through lowering the free radical concentration in sperm cytoplasm (D’cruz and Mathur 2005), in this regard, Farouk et al., (2007) and Pakade et al., (2013) presented that Moringa contains more than 40 natural antioxidant compounds that have the ability to scavenge free radicals.

Testicular size is a good indicator of present and future sperm production (Osinowo et al., 1981; Gage & Freckleton 2003), and there is often a positive correlation between sperm production and testicular volume within animal species (Morris et al. 1999). Moringa leaves have been used as animal feed to improve the measurement of testicular and sperm fertility in rabbits (Abu and Ahemen., 2013; Khalifa et al., 2016), enhancing libido and sperm quality in Bali cattle (Syarifuddin et al., 2017). As well as improving the sperm characteristics of buffaloes (Wafa et al., 2017). Concerning to M. oleifera seeds, Liyasu et al., (2021) reported the efficacy of M. oleifera seed extract in increasing fertility hormones (LH, FSH, serum testosterone) and improving semen properties in rats. On the other hand, M.O leaves supplementation increased the values of testicular size and weight in rabbits (Ojo & Abdurahman., 2017) as well as in mice (Devil et al., 2019). The aim of this study was to investigate the effect of Moringa oleifera supplementation (leaves or seeds) on semen characteristics, biometric measurement of the testis and epididymis and some hematological variables in Barki rams.

**MATERIALS AND METHODS**

**Animals and management:**

This study was conducted using fifteen adult barki rams (aged 8-12 months, weighing 33.23±3.09 kg live body weigh) belonging to the herd of Animal Production Department, Faculty of Agriculture, Minoufia University, Shebin El-Kom, Egypt.

These rams were randomly housed in three separate groups of closed, well-ventilated and lighted pens (5 rams each). The animals were healthy, free of external and internal parasites; the experiment was conducted from January to May 2020 Rams in the three trial groups were fed to meet their requirements of DM according to (NRC, 1985). Roughage: concentrate ratio were 40: 60%, the composition of the basic concentrate feed mixture (CFM) was yellow corn, 50%; soybean meal, 8%; cotton seed meal, 17%; wheat bran, 21%; Table salt, 1%; mineral mix, 0.3%; vitamin mix 0.1%; sodium bicarbonate, 0.5%; sodium bicarbonate, 0.5% and calcium carbonate, 1.6%.

Rams in group 1 (G1) were fed the basal diet supplemented by *moringa olifera* leaves powder (MOL) at a concentration of 5% of CFM, whereas rams in groups 2 (G2) fed the basal diet supplemented by moringa seed cake (MSC) at a concentration of 10% of CFM whereas group 3 (control) were fed the basal diet without supplements. The experimental rations were offered to all animals twice a day at 8 a.m. and 2 p.m. in equal portions. Daily feed allowances were changed quantitatively according to the change in body weight. Water was offered freely all the day.
Blood samples:

Blood samples were collected from the jugular vein of each ram at morning before feeding or drinking in two different vials, the first vial was heparinized with K3-EDTA (1 mg/ml) as an anticoagulant.

The whole blood was shortly analyzed after collection using a hematology analyzer (Sysmex KX-21N Auto Hematology Analyzer, KOBE, JAPAN) for: total count of Erythrocytes (RBCs, x106 / mm3), Hemoglobin concentration (Hb, gm/dl) platelet count (PLT), hematocrit (Packed cell volume, PCV%), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC).

The second vial was free of anticoagulant, it was left until clotting, thereafter, serum was separated carefully after centrifugation at 3000 r.p.m for 20 minutes and stored at -20°C until further analysis:

Total protein and protein fraction:

Concentration of total protein in blood was determined according to the colometric method of Henry, (1964) and the serum albumin according to Rodkey, (1965) and Webster, (1974). The globulin concentration was calculated as the difference between total serum protein and serum albumin. Also the A/G ratio was calculated.

Transaminases:

Activities of Alanin transaminase (ALT) and aspartate aminotransferase (AST) were measured coloimetrically according to Reitman and Frankel (1957)

In addition, uric acid according to Caraway, (1963), creatinine according to Murray, 1984 and Young, 1995) and glucose according to (Trinder, 1969) were determined in blood serum.

Concentration of testosterone in blood serum was estimated by radioimmunoassay according to the procedure described by Ekins (1984).

Semen collection and evaluation:

Semen was collected twice weekly by electro-ejaculation method (Blackshow, 1954) using a Lane electroejaculator with a ram rectal probe. Immediately after semen collection, the semen samples were individually evaluated for the following physical characteristics: Ejaculate volume to the nearest 0.1 ml, ejaculate color and viscosity, pH, sperm wave motility score was estimated according to Amman and Hammerstedt (1980) and sperm abnormality (Blom, 1983). Also, the response of spermatozoa to HOS-test was assessed in term of percentage of curled spermatozoa at 50 mOsm/l for 30 min according to El-Sherbieny (2004).

Biometric evaluation of epididymis and testicles:

At the end of the feeding period, two rams from each group were slaughtered, and then the genital tracts were removed, freed from extraneous tissues, put into labeled plastic bags and transported in an ice pack to the laboratory where the epididymides were separated from testes. The measurements of Paired testes (left and right) of each animal, namely, testicular length and width (cm) as well as testicular weight (gm) were evaluated and recorded. The biometry of testes and epididymis were evaluated for every ram according to the method of Thakur and Dixit (2006) & Amini and Kamkar, (2005) as follow: Testicular width was measured around the widest point at an area that is equidistant to the testicular poles as well as, testicular length (cm) was also measured along the longitudinal axis of the testis beginning from one pole of the testis to the other pole.

Testicle size: Volume of each testis was estimated by water displacement to the nearest cubic centimeter for each ram. The right and left epididymis of each ram were divided into head, body and caudal parts. The weight (g) as well as the length and width (cm) of each piece were determined and recorded.

Statistical Analysis:

Percentages of semen characteristics such as motility, death, abnormalities.... etc. were
recorded as percentages and subjected to arcsin transform for analysis of variance. Statistical differences between the means of various groups were evaluated by one-way analysis of variance (ANOVA) followed by Duncan's test using SPSS program.

RESULTS AND DISCUSSION

I- The effect of moringa olifera leaves and moringa olifera cake seeds supplementation on the Hematological parameters:

Results in Table (1) showed no significant changes as responded to the MOL and MSC supplementation to the Barki rams rations on all investigated Haematological parameters. Leaves powder Supplementation in MOL slightly increased the mean corpuscular volume (MCV) (103.74±4.81 fl) and platelets (PLT) (258.75±42.81 10³/m), While it slightly decreased red blood cells (RBCs) count 10⁶/m, hemoglobin (HGB) g/dl, the mean corpuscular hemoglobin (MCH) pg, the mean corpuscular hemoglobin concentration (MCHC) g/dl and packed cell volume (PCV.) (3.30%±0.23, 9.94±0.53, 29.87±0.41, 29.45±1.06 and 33.77±0.67, respectively) compared to the control group (3.47±0.217, 10.35±0.50, 29.90±0.39 and 34.95±0.63, respectively). Supplementing ration with MSC insignificantly increased RBC (3.87±0.27), HGB (10.97±0.610), MCHC (31.30±1.22 g/dl), PLT (269.00±4 9.4310³/m), though it decreased MCV (95.60±5.56 fl) and MCH (28.55±0.47 pg) compared to the control group. Results of hematological parameters were within normal ranges of animal response to external environment (Isikwenu et al., 2012).

The non-significant changes observed among the different groups in our hematological studies are in agreement with Luqman et al. (2012) and Omran (2008); however, on contrary with the results of Babeker and Abdalbagi (2015) on Sudan Nubian goats fed MOL and Wafa et al. (2017) who found significant (P<0.05) increase in PCV, hemoglobin Hb and RBCs as affected by MOL supplementation to buffalo bulls diets and Akinyemi et al. (2010) also found that include MOL in the diet improved all hematological parameters in West African Dwarf rams. Furthermore, Fayomi et al. (2014) obtained similar results in yearling rams. The increase in RBC concentration could be due to the antioxidant properties of the extract as reported by Revin et al. (2019).

Table (1): The effect of moringa olivera leaves and moringa cake seeds on the hematological parameters.

<table>
<thead>
<tr>
<th>Groups Parameters</th>
<th>MOL (5 % leave powder)</th>
<th>MSC (10% cake seeds)</th>
<th>Control Without suppl.</th>
<th>Probability</th>
</tr>
</thead>
<tbody>
<tr>
<td>RBC (10⁶/m)</td>
<td>3.30±0.23</td>
<td>3.87±0.27</td>
<td>3.47±0.217</td>
<td>N.S.</td>
</tr>
<tr>
<td>(Hb, gm/dl)</td>
<td>9.94±0.53</td>
<td>10.97±0.610</td>
<td>10.35±0.50</td>
<td>N.S.</td>
</tr>
<tr>
<td>MCV fl</td>
<td>103.74±4.81</td>
<td>95.60±5.56</td>
<td>102.07±4.57</td>
<td>N.S.</td>
</tr>
<tr>
<td>MCH pg</td>
<td>29.87±0.41</td>
<td>28.55±0.47</td>
<td>29.90±0.39</td>
<td>N.S.</td>
</tr>
<tr>
<td>MCHC. g/dl</td>
<td>29.45±1.06</td>
<td>31.30±1.22</td>
<td>29.84±1.00</td>
<td>N.S.</td>
</tr>
<tr>
<td>PLT (10³/m)</td>
<td>258.75±42.81</td>
<td>269.00±49.43</td>
<td>256.65±40.61</td>
<td>N.S.</td>
</tr>
<tr>
<td>PCV (%)</td>
<td>33.77±0.67</td>
<td>35.13±0.77</td>
<td>34.95±0.63</td>
<td>N.S.</td>
</tr>
</tbody>
</table>

RBC: red blood cells, Hb: Hemoglobin concentration (gm/dl), MCV: the mean corpuscular volume, MCH: The mean corpuscular hemoglobin, MCHC: The mean corpuscular hemoglobin concentration, PLT: platelets, PCV: Packed cell volume.
II-The effect of moringa olifera leaves and moringa olifera seeds cake supplementation on the some blood metabolites:

The results in Table (2) show that adding MOL or MSC to Barki ram diets had a significant impact on glucose, creatinine, urea, ALT, AST, and testosterone levels. In this regard, MOL supplementation significantly increased ALT, AST and testosterone (57.64±5.53, 87.91±6.15, 5.27±0.27, respectively) and insignificantly for creatinine (1.34±0.04) compared to those rams fed a ration free of Moringa. While rams fed MSC in diet showed significant increase in creatinine, urea, ALT and AST levels (1.39±0.03, 4.71±0.11, 57.19±5.19 and 84.88±5.77, respectively, Table, 2). Conversely, the addition of MOL or MSC resulted in a significant decrease of glucose conc. (71.27±2.01 and 87.98±1.89) in the treated groups, respectively compared to those of rams fed control ration (99.66±2.39), moreover, leaves in group one insignificantly underestimated the rams, plasma urea conc. (3.87±0.12). The blood testosterone concentration in rams given MSC was considerably lower (2.96 ±0.25) than in rams in the control group (4.49±0.31).

Generally, the quantity and quality of feed, as well as the level of anti-nutritional substances or factors present in the feed, impact both the biochemical and hematological blood components (Akinmutimi, 2004). It is also a crucial indicator of physiological and pathological condition in both animals and humans (Harper, 1973).

According to Olophobo et al. (1992) a normal blood glucose level shows that animals are not living at the expense of bodily tissues. Glucose conc. had been decreased significantly as affected by the experimental additives in this study and the same results obtained by Luqman et al., 2012 and Jaiswal et al., (2009). This might indicate that Moringa has an insulin-like impact on peripheral tissues, either by boosting glucose uptake and metabolism or by inhibiting gluconeogenesis. It is likely that the aqueous extract of the moringa leaves has some effect of increasing the tissue utilization of glucose (Gray et al., 2000, Oyedepo et al., 2013) by inhibiting hepatic gluconeogenesis or absorption of glucose into the muscles and adipose tissues (Pradana, et al., 2021). However, Ahemen et al. (2013) reported insignificant influence of diet containing MOL on glucose in rabbit serum. The increase in ALT, AST values as affected by MOL and MSC in our study is in line with the obtained results by Ghebreselassie et al., (2011) who treated mice with doses of 600, 750 and 900mg/kg as compared to the control group although it was not statistically significant. this could be returned to the presence of glucosinolate in Moringa leaf extract (Mekonnen et al., 2003, Bennet et al., 2003), as its described that rats fed diets containing individual glucosinolates produce small increase in the weights of liver and kidneys.

Table (2): Effect of Moringa leaves and Moringa seeds supplementation in Barki rams diets on some blood metabolites.

<table>
<thead>
<tr>
<th>Groups</th>
<th>Glucose (mg/dl)</th>
<th>Creatinin (mg/dl)</th>
<th>Urea (mg/dl)</th>
<th>ALT (u/l)</th>
<th>AST (u/l)</th>
<th>Testosterone (ng/dl)</th>
</tr>
</thead>
<tbody>
<tr>
<td>MOL (G1)</td>
<td>71.27±2.01&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.34±0.04&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.87±0.12&lt;sup&gt;b&lt;/sup&gt;</td>
<td>57.64±5.53&lt;sup&gt;a&lt;/sup&gt;</td>
<td>87.91±6.15&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.27±0.27&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>MSC (G2)</td>
<td>87.98±1.89b</td>
<td>1.39±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.71±0.11&lt;sup&gt;a&lt;/sup&gt;</td>
<td>57.19±5.19&lt;sup&gt;a&lt;/sup&gt;</td>
<td>84.88±5.77&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.96±0.25&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Control (G3)</td>
<td>99.66±2.39&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.33±0.04&lt;sup&gt;b&lt;/sup&gt;</td>
<td>4.10±0.14&lt;sup&gt;b&lt;/sup&gt;</td>
<td>46.22±6.56&lt;sup&gt;b&lt;/sup&gt;</td>
<td>82.04±7.30&lt;sup&gt;c&lt;/sup&gt;</td>
<td>4.49±0.31&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

| probability | ** | *** | ** | * | * | ** |

<sup>a, b, c</sup>: Means in the same column bearing different superscripts are significantly different.* significant at P ≤ 0.05, ** significant at P ≤ 0.01, ALT: Alanine transaminase, AST: Aspartate transaminase.
Our trend in ALT and AST changes in the rams of the experimental groups were different from data of Khalel et al. (2014) who reported that feeding lactating cows on MOL up to 40% of the whole daily ration did not badly affect liver or kidney functions; Luqman et al., (2012) who recorded non-significant changes in GPT, GOT, as affected by different moringa byproducts extracts, the extracts were well tolerated till dose of 100 mg/kg b/w, with no toxicity. Eshak and Osman (2013) observed that MOL aqueous extract had a therapeutic action through enhancing of liver enzyme activities (AST and ALT) in irradiated rats by gamma irradiation.

Increased serum urea concentration may suggest an increase in activities of urea enzymes ornithine, carbonyl transferase and orginase, which may also indicate kidney damage (Ajagbonna et al., 1999). Also, Hoffmann et al. (2003) found high utilization of MO nitrogen making them available in the small intestine in an intact form led to lower blood urea level. On the other hand, Ahemen et al. (2013) reported insignificant effect of MOL meal diet on concentration of creatinine and urea as well as AST and ALT activity in blood serum of rabbit, while total blood protein and creatinine contents have been shown to depend on the quantity and quality of dietary protein (Eggum, 1970; Iyayi & Tewe, 1998; Awosanya et al., 1999; Esonu et al., 2001). Source of excess creatinine in the blood of animals is normal due to creatinine phosphate catabolism (Bell et al., 1992).

Ojo & Abdurahman., 2017 reported that the testosterone concentration insignificantly increased in bucks treated by MOL extract as compared to control group, This could be due to the saponins content in Moringa leaves which can boost the level of testosterone in the body (Tesfay et al., 2011) and to the significant increase in LH hormone concentration which could lead to increased testosterone secretion from leydig cells (Thakur and Dixit., 2006). The decrease of testosterone concentration in MSC was in contrary with what found by Devi et al. (2019) who confirmed that Moringa seeds can increase male fertility hormones particularly testosterone, follicle-stimulating hormone (FSH) and luteinizing hormone (LH) and sperm analyses in adult male rats.

### III- The effect of moringa olifera leaves and moringa olifera seeds cake supplementation on total protein and protein fraction in blood of Barki rams:

Results in Table (3) showed decrease in all blood proteins parameters in the experimental groups (MOL and MSC) compared to the control one; some of the decreases in proteins parameters were significant i.e., total protein (T.P) (6.53±0.13 and 7.89±0.13), Globulin (G) (3.40±0.07 and 4.13±0.06) and A /G ratio (0.98±0.02 and 0.98±0.02) in MOL and MSC, respectively compared to those in the control group (9.59±0.16, 4.76±0.08 and 1.02±0.03, respectively), on the other hand there were insignificant decrease in Albumin con. in the treated groups vs. control one.

Table (3): The effect of moringa olifera leaves and moringa olifera cake seeds supplementation on total protein and protein fraction in blood serum of Barki rams:

<table>
<thead>
<tr>
<th>Groups</th>
<th>Total protein (g/dl)</th>
<th>Albumin (A) (g/dl)</th>
<th>Globulin (G) (g/dl)</th>
<th>A /G</th>
</tr>
</thead>
<tbody>
<tr>
<td>MOL</td>
<td>6.53±0.13&lt;sup&gt;c&lt;/sup&gt;</td>
<td>4.38±0.736&lt;sup&gt;a&lt;/sup&gt;</td>
<td>3.40±0.07&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.98±0.02&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>MSC</td>
<td>7.89±0.13&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.76±0.690&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.13±0.06&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.98±0.02&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Control</td>
<td>9.59±0.16&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.84±0.873&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.76±0.08&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.02±0.03&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>probability</td>
<td>**</td>
<td>N.S.</td>
<td>**</td>
<td>**</td>
</tr>
</tbody>
</table>

<sup>a, b, c</sup>: Means in the same column bearing different superscripts are significantly different.

* significant at $P \leq 0.05$, ** significant at $P \leq 0.01$, N.S.: not significant, A /G: Albumin/ Globulin ratio.
Effect of moringa olifera leaves and seeds supplementation on semen characteristics, testes …

According to Eggum (1970), Iyayi & Tewe (1998); Awosanya et al. (1999); Esonu et al. (2001) blood total proteins depend on the quantity and quality of dietary protein. The depletion of total proteins in MOL and MSC comparing to control may be refer to decrease of the protein retained in the animal body as reported by (Esonu et al., 2001) also Wafa et al. (2017) indicated that MOL treatment had impact on decreasing protein metabolites and higher protein utilization in bulls fed MOL than in control. These results are opposite to those of Garg et al. (1992) who explained that MOL chemical composition may increase rumen undegradable protein utilization and improve synthesis of microbial protein in the rumen (Soliva et al., 2005). Also, feeding ruminants on MOL can help in carbohydrates absorption and increasing metabolizable energy (Khalel et al., 2014).

IV- The effect of moringa olifera leaves and moringa olifera seeds cake supplementation on semen characteristics:

Table (4) shows the effect of MOL and MSC supplementations on the physical semen properties of Barki rams. Both supplements to the experimental ration had positive effects on some important physical semen characteristics examined in this study as volume, percentages of sperm motility, livability, dead, abnormality, normality and curled sperms found in hypo osmotic compared to the rams fed un-supplemented rations.

Our results proved that MOL insignificantly increased, Color, viscosity, pH, (3.39±0.28, 2.91±0.22, 6.95±0.05, respectively) in the same time it significantly increased the motility, livability and normality% (0.72±0.16, 93.90±0.47, 95.32±0.39, respectively) compared to the control group; Whereas supplementing Barki rams with MSC significantly increased volume (1.10±0.19 ), pH (7.02±0.06 ), normality% (94.18±0.39) and color (3.36±0.32), and insignificantly increased viscosity (3.34±0.25), motility (73.58±3.53), livability (92.98±0.47) compared to the same semen parameters recorded to the control group. Conversely, rams fed MOL and MSC containing diets had a lower percentage of abnormal, dead, and curled tail sperms than rams fed the control. In this regard, some of the experimental semen characteristics were insignificantly higher in MSC than in MOL such as volume, pH and motility and other were insignificantly higher in MOL than in MSC i.e., color, viscosity, livability, except abnormality was significantly higher in MOL than in MSC.

Table (4): The effect of moringa olifera leaves and moringa olifera seeds cake supplementation on semen characteristics.

<table>
<thead>
<tr>
<th>Traits</th>
<th>MOL</th>
<th>MSC</th>
<th>Control</th>
<th>probability</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volume (ml)</td>
<td>0.72±0.16&lt;sup&gt;a&lt;/sup&gt;b</td>
<td>1.10±0.19&lt;sup&gt;a&lt;/sup&gt;b</td>
<td>0.44±0.19&lt;sup&gt;b&lt;/sup&gt;</td>
<td>*</td>
</tr>
<tr>
<td>Color (1-5)</td>
<td>3.39±0.28</td>
<td>3.36±0.32</td>
<td>3.15±0.33</td>
<td>N.S.</td>
</tr>
<tr>
<td>Visco. (1-5)</td>
<td>2.91±0.22</td>
<td>3.34±0.25</td>
<td>2.89±0.25</td>
<td>N.S.</td>
</tr>
<tr>
<td>pH</td>
<td>6.95±0.05&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>7.02±0.06&lt;sup&gt;a&lt;/sup&gt;</td>
<td>6.82±0.06&lt;sup&gt;b&lt;/sup&gt;</td>
<td>*</td>
</tr>
<tr>
<td>HOST (%)</td>
<td>18.38±2.04</td>
<td>19.88±2.44</td>
<td>22.79±2.37</td>
<td>N.S.</td>
</tr>
<tr>
<td>Motility (%)</td>
<td>72.43±3.53</td>
<td>73.58±3.53</td>
<td>64.53±3.53</td>
<td>N.S.</td>
</tr>
<tr>
<td>Dead sperm (%)</td>
<td>6.10±0.47&lt;sup&gt;b&lt;/sup&gt;</td>
<td>7.02±0.47&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>7.87±0.47&lt;sup&gt;a&lt;/sup&gt;</td>
<td>*</td>
</tr>
<tr>
<td>Live sperm (%)</td>
<td>93.90±0.47&lt;sup&gt;a&lt;/sup&gt;</td>
<td>92.98±0.47&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>92.43±0.47&lt;sup&gt;b&lt;/sup&gt;</td>
<td>*</td>
</tr>
<tr>
<td>Abnormal sperm (%)</td>
<td>4.68±0.39&lt;sup&gt;c&lt;/sup&gt;</td>
<td>5.82±0.39&lt;sup&gt;ab&lt;/sup&gt;</td>
<td>8.28±0.39&lt;sup&gt;a&lt;/sup&gt;</td>
<td>**</td>
</tr>
<tr>
<td>Normal sperm (%)</td>
<td>95.32±0.39&lt;sup&gt;a&lt;/sup&gt;</td>
<td>94.18±0.39&lt;sup&gt;b&lt;/sup&gt;</td>
<td>91.73±0.39&lt;sup&gt;c&lt;/sup&gt;</td>
<td>**</td>
</tr>
</tbody>
</table>

<sup>a, b, c</sup>: Means in the same column bearing different superscripts are significantly different.

* Significant at P ≤ 0.05, ** significant at P ≤ 0.01, N.S: not significant, HOST: hypo osmotic swelling test.
Our results proved that supplementing rams ration with MOL and MSC can enhance semen quality; this may be due to the fact that MOL are valuable in minerals such as Fe, vitamins A, B complex, C, K and E which are of importance in enhancing semen quality and reaction time (Odeyinka et al., 2008; Faye et al., 2011; Dougnon et al., 2012) as well as the seeds (Iliyasu et al., 2021 and Devi et al., 2019), also Shokry et al. (2020) refer that to their ability to improve the characters of the fresh and cryopreserved semen of Barki rams via improving seminal plasma antioxidant defense mechanism. It was stated that the sperms motility is the most important parameters about fertility (Castellini & Lattaioli, 1999 and Brun et al., 2002), and a good indicator of viability (Mohan et al., 2021).

In the present study the sperm motility was increased insignificantly by the dietary treatment with MOL and MSC; this was in agreement with Abu and Ahemen, (2013) who has observed that MOL meal did not significantly increased sperm active motility and George et al. (2017) and Shokry et al. (2020) who observed significant improvement in sperm motility which was noticed in rabbit fed 30g of Moringa oleifera. Also Khalifa et al. (2016) and Oyeyemi et al. (2008) found that MOL significantly increased percentage of semen motility. Oyeyemi and Okediran (2007) also declared that quality nutrition with high percentage of protein will improve motility and concentration of spermatozoa and Moringa leaves is known to have high crude protein content. Also Toleng and Yusuf (2021) explained that the antioxidant compounds from the Moringa seed extract help in maintaining the sperms to move progressively.

Semen viscosity is the liquid flow of semen after ejaculation. Herbert (1992) also reported that the appearance of semen is a part of important characteristic of quality; high semen viscosity causes low sperm mobility, if the sperm does not liquefy or remains very thick, fertilization maybe difficult due to the sperm not being able to reach an egg. Good quality semen should have a uniformly milky appearance which gives the indication of high sperm concentration (George et al., 2017). Our results shows that appearance of the Barki rams semen and viscosity was insignificantly increased by the treatments, as semen usually exhibit milky and homogeneous stickiness with medium viscosity. These results are similar to the results of Mohan et al. (2021). The percentage of normal sperm cells in our study showed significant increased pattern affected by inclusions of MOL and MSC, our findings were supported by those of Ojo & Abdurahman (2017) and Mohan et al. (2021).

The percentage live sperm cells are those present for use during fertilization (Ajala et al., 2001). The percentage of live sperm cells in this study were significantly influenced by dietary supplement of MOL and MSC whereas Mohan et al. (2021) and Shokry et al. (2020) found no significant increase by dietary supplement of MOL. Also Mohan et al. (2021) illustrated that there is no influence of treatment recorded in semen appearance and semen viscosity while Shokry et al. (2020) reported that MOL extract increased Barki rams semen volume.

V- Effect of adding Moringa oleifera leaves and Moringa oleifera seed cake on the biometrics of testis and epididymis.

The results in Table (5) showed that MOL and MSC supplementation in the feed of Barki rams had no significant effect on the measurements of the right and left testicles (weight, width and length), while the differences in testicle size of the left testis between the experimental groups were significant. The highest recorded values of the testis weight and width were observed in MOL group. However, the testicular size was relatively higher for the rams fed moringa-free diet compared to those supplemented with MOL or MOS.

Results in Table (6) show that adding MOL to Barki ram diets insignificantly increase the measurements of three epididymal segments (caput, body, caudal) of the left or right epididymis as compared to those recorded in the control group rams, except for the length of caudal segment of the left testicle. On the other
hand, the results of the current study (Table, 6) also showed that MSC supplementation to the diet of rams led to a decrease in the measurements of different epididymal parts except for body weight in the right or left epididymis (4.10±0.81, 3.95±0.61 and 10.50±0.87, respectively) which were slightly higher than that recorded in the control group (4.10±0.23, 3.25±0.83 and 10.50±0.29, respectively).

Table (5): The effect of moringa olifera leaves and moringa olifera seeds cake supplementation on testicular measurements and testicle size (ml) in Barki rams.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Testicular measurements</th>
<th>Testicle size (ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Weight (gm)</td>
<td>Width (cm)</td>
</tr>
<tr>
<td>Right testis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MOL</td>
<td>118.00±1.73</td>
<td>8.50±0.29</td>
</tr>
<tr>
<td>MSC</td>
<td>95.50±15.88</td>
<td>7.50±0.87</td>
</tr>
<tr>
<td>Control</td>
<td>109.50±14.15</td>
<td>7.75±0.14</td>
</tr>
<tr>
<td>Left testis</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MOL</td>
<td>115.00±1.73</td>
<td>8.50±0.29</td>
</tr>
<tr>
<td>MSC</td>
<td>95.50±16.45</td>
<td>8.00±0.58</td>
</tr>
<tr>
<td>Control</td>
<td>112.50±14.15</td>
<td>7.25±0.43</td>
</tr>
</tbody>
</table>

a, b, c: Means in the same column bearing different superscripts are significantly different.

Table (6): Effect of Moringa Oleifera Seed Cake and Moringa Oleifera Leaf Supplementation on Epididymal Biometrics in Barki rams.

<table>
<thead>
<tr>
<th>Measurements</th>
<th>Treatments</th>
</tr>
</thead>
<tbody>
<tr>
<td>R Caput</td>
<td>MOL MSC Control</td>
</tr>
<tr>
<td>Weight, g</td>
<td>9.25±0.14</td>
</tr>
<tr>
<td>Length, cm</td>
<td>6.00±0.00a</td>
</tr>
<tr>
<td>R Body</td>
<td>MOL MSC Control</td>
</tr>
<tr>
<td>Weight, g</td>
<td>4.25±1.01</td>
</tr>
<tr>
<td>Length, cm</td>
<td>11.50±0.58</td>
</tr>
<tr>
<td>R caudal</td>
<td>MOL MSC Control</td>
</tr>
<tr>
<td>Weight, g</td>
<td>10.50±0.29</td>
</tr>
<tr>
<td>Length, cm</td>
<td>4.25±0.14a</td>
</tr>
<tr>
<td>L Caput</td>
<td>MOL MSC Control</td>
</tr>
<tr>
<td>Weight, g</td>
<td>9.50±0.29</td>
</tr>
<tr>
<td>Length, cm</td>
<td>6.00±0.00a</td>
</tr>
<tr>
<td>L Body</td>
<td>MOL MSC Control</td>
</tr>
<tr>
<td>Weight, g</td>
<td>4.00±0.58</td>
</tr>
<tr>
<td>Length, cm</td>
<td>11.50±0.87</td>
</tr>
<tr>
<td>L caudal</td>
<td>MOL MSC Control</td>
</tr>
<tr>
<td>Weight, g</td>
<td>10.00±0.58</td>
</tr>
<tr>
<td>Length, cm</td>
<td>4.00±0.29a</td>
</tr>
</tbody>
</table>

a, b, c: Means in the same row bearing different superscripts are significantly different (p<0.05).
This could be explained by the higher level of testosterone recorded in the blood of rams fed MSC (5.27 ± 0.27 ng/dL) compared to the level recorded in the group of rams supplemented with MOL (2.96 ± 0.25 ng/dL) or those of rams of the control group (4.49 ± 0.31 ng/dL) as shown in Table (2). Watcho et al., (2017) reported that the testes, epididymis, and other reproductive organs are structurally and physiologically dependent on testosterone and other androgens. Since testosterone enhances the development and secretory activity of reproductive organs (Gornal et al., 1949). These observations are in agreement with Ojo & Abdurahman et al., (2017) who reported that rabbits took MOL in diets had the highest recorded values of the paired testis weight, volume, width and testis length compared to the control group, and Devi et al. (2019) found that rats treated with Moringa oleifera leaves showed significant (P≤0.01) dose-dependent increase in body weight and sexual organs. Devi et al. (2019) found that rats treated with Moringa oleifera leaves showed significant (P≤0.01) dose-dependent increase in body weight and sexual organs.

**Conclusion:**

It could be concluded that supplementation of MOL and MSC rations into Barki rams’ rations improved most of the semen characteristics (volume, motility, vitality and normality) and reduced mortality and malformations. However, it negatively affects kidney function (increased urea and creatinine) and liver enzyme activities (ALT & AST). This means that there may have been a change in the histological structure of the liver and kidneys, so it is suggested that in future studies of additives or supplements, histological studies should be done on the liver, kidneys, spleen, etc., in addition to blood analysis.

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تأثير إضافة أوراق وبذور المورينجا على خصائص السائل المنوي ومقاييس الخصية والبربخ وبعض قياسات الدم في كبش البرقي.

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الملخص العربي

هدفت هذه الدراسة إلى دراسة تأثير إضافة مكملات مسحوق أوراق المورينجا أوليفيرا (MOL) أو تقل بذور المورينجا (MSC) على علاق الكبش البرقي على بعض مكونات الدم وخصائص السائل المنوي وقياسات الخصية والبربخ في كبش البرقي. تم استخدام خمسة عشر كبدًا برضفي تراوح أعمارهم بين 12-21 شهرًا تم تقسيمهم إلى ثلاث مجموعات مماثلة (5 كبدًا/كل مجموعة). تم تغذية الكباه في المجموعة الأولى (G1) بالمكمل MOL بكمية موزعة (5% من خليط الطبق المركزي)، بينما تغذية الكباه في المجموعة الثانية (G2) بكمية موزعة (5% من خليط الطبق المركزي) بالنمط الغذائي الأساسي دون مكملات. ثم جمع عينات الدم وتكييف الكباه، أستجرت المنظمة الدولية للمختبرات للتدقيق في الكبد والبربخ، AST, ALT, MCH, MCV, PCV, PLT, Hb, MCHC، AST، AST، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، 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ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، ALT، 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