



Effect of Adding Quercetin to Tris Extender on Some Semen Biochemical Characteristics of Awassi Rams Following Different Cooling Periods

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Abstract

The study was planned to evaluate the effect of adding Quercetin in some characteristics of the sperm of the ram. This study was conducted in the animal field of the Animal Production Department / Faculty of Agricultural Engineering Sciences / University of Baghdad, for the period 2021/12/5 to 2022/2/20. In this experiment, 3 rams were used at the age of 2-2.5 years and weighed 50-55 Kg. The semen was collected early in the morning and once a week and the semen was pooled to remove the individual differences. The treatments were divided: qur- free control group, treatment T1 (3mm/ ml qur), T2 treatment (6 mm/ ml qur), T3 treatment (9mm/ml qur). the result of the study showed significant decreased in the concentration of AST in treatment T3 at time (2 and 72 hours) compared with the other treatments. While there were significant decreased in the concentration of ALT in the treatment T1 and T3 at time (2 and 72 hours) respectively. The T3 treatment was showed significant decreased in the concentration of Malondialdehyde at time (0 and 72 hours) compared the other treatments. The study showed no significant effect of Qur on total antioxidant between the treatments.

Key Words: Quercetin, Tris Extender, Semen Biochemical Characteristics, of Awassi Rams.

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DOI Number: 10.14704/nq.2022.20.6.NQ22264

NeuroQuantology 2022; 20(6):2717-2719

Introduction

Quercetin is part of the flavonoids and is commonly found in foods such as fruits and vegetables, has many biological activities including being an antioxidant (Ben Abdallah et al, 2011, Gibb et al, 2013). Ram sperm contain a high percentage of unsaturated fatty acid in the plasma membrane, so they are more sensitive to oxidative stress and free radical formation during cooling periods (Diaz et al, 2011). Studies have indicated that quercetin has positive effects on fresh sperm and after thawing in different types of agricultural animals (Gibb et al, 2013, Seifi-jamadi et al, 2016) the quercetin was prevent lipid peroxidation by inhibiting the production of free radicals with Alpha- tocopherol to delay the oxidation and stimulating gene expression of enzymes such as glutathione s-transferase and glucuronosyl transferase (Wang et al, 2016). Therefore, the research aims to explain

the effect of adding quercetin to the Tris in some semen characteristics of the ram.

Materials and Methods

This study was conducted in the animal field of the Animal Production Department / collage of Agricultural Engineering Sciences / University of Baghdad for the period from 5/12/2021 to 20/2/2022. The process of collecting sperm from 3 rams by artificial vagina and once a week. The samples were folded and diluted 1:10. The strain dilator was prepared by Salamon and Maxwell (2000) and three concentrations of Quercetin, the first treatment (3 µl / ml), the second (6 µl / ml) and the third (9 µl / ml) for diluted semen plus control group.

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Attributes studied the researchers calculated the concentration according to the method of researchers Guidet and Shah, (1989). Effectiveness of ALT and AST: Effectiveness was estimated by Retman and Frankel (1957) based on the kit prepared by French biomerieux. The Statistical Analysis System (SAS) (2012) was used for data analysis, and the differences between the averages were compared with the Duncan test (1955).

Results

The results of the study showed a significant decrease (p<0.01) in treatment T3 in AST concentration at 2 and 72 hours in seminal plasma of Awassi rams it was (47.79±4.30 IU/L) compared with the T2 and T1 (97.04±11.23, 84.52± 10.63 IU/L) respectively at 2 hours of cooling period. After 72 hours, the concentration of AST enzyme was significantly decreased (p<0.01) in seminal plasma (48.36 ± 1.07 IU/L) compared with the control treatment (84.25± 8.80 IU/L) (table 1).

Table 1. Effect of adding different concentration of quercetin to Tris extender in AST enzyme level (IU/L) at (2 and 72 hrs.) of cooling period (5°C) for Awassi rams semen (mean ± standard error)

| Treatment | Time | |
|-----------|-----------------|-------------------|
| | 0 hour | 72 hour |
| Control | 84.25 ± 8.80 a | 57.05 ± 2.93 bc |
| T1 | 84.63 ± 10.63 a | 108.14 ± 15.55 ab |
| T2 | 97.04 ± 11.23 a | 144.68 ± 32.69 a |
| T3 | 47.79 ± 4.30 b | 48.36 ± 7.07 c |
| P VALUE | ** | ** |

Different superscripts within column are significantly different (P<0.01) **

The results of the study showed a significant decreased (p<0.01) in the concentration of ALT enzyme in the T1 treatment at 2 hrs. of cooling periods it was (356.17 ± 22.08 IU/L) compared with T2 and T3 treatment (311.27 ± 13.58, 251.80 ± 9.98 IU/L) Respectively (table 2).

In cooling preservation after 72 hrs., T3 treatment showed significantly decreased (p<0.01) in the enzyme level (97.62 ± 25.80 IU/L) compared with control treatment (235.00 ± 18.74 IU/L) (table 2).

Table 1. Effect of adding different concentration of quercetin to Tris extender in ALT enzyme level (IU/L) at (2 and 72 hrs.) of cooling period (5°C) for Awassi rams semen (mean ± standard error)

| Treatment | Time | |
|-----------|------------------|-------------------|
| | 0 hour | 72 hour |
| Control | 120.65 ± 17.97 c | 216.65 ± 19.65 ab |
| T1 | 85.36 ± 9.35 c | 166.66 ± 20.73 bc |
| T2 | 174.40 ± 20.23 b | 246.16 ± 6.12 a |
| T3 | 243.34 ± 20.75 a | 124.81 ± 26.72 c |
| P VALUE | ** | ** |

Different superscripts within column are significantly different (P<0.01) **

The results of the current study showed significantly decreased (p<0.01) at time (2 and 72 hrs.) Of cooling preservation of T3 treatment (2.79 ± 0.25, 2.48 ± 0.26 μmol / 10⁹ sperm) respectively compared with the other treatments (table 3).

Table 3. Effect of adding different concentration of quercetin to Tris extender in Malodialdehyde (μmol/10⁹ sperm) at (2 and 72 hrs.) of cooling period (5°C) for Awassi rams semen (mean ± standard error)

| Treatment | Time | |
|-----------|-------------------|------------------|
| | 0 hour | 72 hour |
| Control | 271.57 ± 12.96 bc | 235.00 ± 18.74 b |
| T1 | 356.17 ± 22.08 a | 206.70 ± 18.31 b |
| T2 | 311.27 ± 13.58 b | 500.45 ± 25.67 a |
| T3 | 251.80 ± 9.98 c | 97.62 ± 25.80 c |
| P VALUE | ** | ** |

Different superscripts within column are significantly different (P<0.01) **

The results of the current study showed no significant effect of adding quercetin to tris extender of awassi ram semen in total antioxidants level at time (2 and 72 hrs.) after cooling preservation between all the treatments (table 4).

Table 4. Effect of adding different concentration of quercetin to Tris extender in total antioxidants level (μmol) at (2 and 72 hrs.) of cooling period (5°C) for Awassi rams semen (mean ± standard error)

| Treatment | Time | |
|-----------|---------------|---------------|
| | 0 hour | 72 hour |
| Control | 2.42 ± 0.16 a | 2.47 ± 0.44 a |
| T1 | 2.59 ± 0.23 a | 2.56 ± 0.17 a |
| T2 | 2.48 ± 0.37 a | 2.47 ± 0.22 a |
| T3 | 2.79 ± 0.25 a | 2.48 ± 0.26 a |
| P VALUE | N.S | N.S |

Different superscripts within column are significantly different (P<0.01) **



Discussion

The addition of quercetin had a significant effect in decreasing the level of AST and ALT activity in seminal plasma of awassi rams at cooling preservation. The level of enzymes outside the cell is a good indicator of semen quality because it is a measure of the stability of the sperm membrane. This is due to the importance of quercetin in maintaining the integrity of the sperm membrane and reducing the levels of abnormalities in the membranes, thus preventing the leakage of enzymes into the seminal plasma (Gunogan, 2006). The adding of quercetin to tris extender of semen improved acrosome integrity and reduced abnormality of sperm that prevents the AST AND ALT enzymes from leaking out to seminal plasma. This is due to quercetin having antioxidant activity and preventing oxidation of polyunsaturated fatty acid in sperm membrane (Gunogan, 2006, Fayyad and Mahmood, 2019).

The results of this study showed significantly decreased MDA levels after adding quercetin to tris extender of awassi rams semen. The occurrence of oxidation in the lipids present in the membranes of the sperm is an important measure of high concentrations of MDA. Therefore, quercetin reduces the damage to the sperm membranes and thus preserves the fertility of the semen (Moretti et al 2012, Seifi-jamadi et al 2017, Saieed et al, 2019). The current results showed no significant effect of adding quercetin to tris extender of awassi rams semen in total antioxidant activity at cooling preservation between all treatments.

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