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LIST OF ABBREVIATION

AI	Artificial insemination
EY	Egg yolk
SBL	Soybean lecithin
BHT	Butylated hydroxytoluene
Tris	hydroxymethylamino methane
PUSFA	Polyunsaturated fatty acids
ROS	Reactive oxygen species
Se-NPs	Selenium-nanoparticles
LPO	lipid peroxidation
LDL	Low density lipoprotein
HDL	High density lipoprotein
TAC	Total antioxidant capacity
MDA	Malondialdehyde
LDH	Lactate dehydrogenase
HOS-t	hypo-osmotic swelling test
LM	Light microscopy
EM	Electron microscopy
TEM	Transmission electron microscopy
ODF	Outer dislocated dense fibers
PS	Phosphatidylserine
CIDR	Control Internal drug release
FC	Flow Cytometric
A	Annexin-V
PI	Propidium iodide
DNA	Deoxy ribonucleic acid
PBS	Phosphate buffer solution
FITC	fluorescein isothiocyanate

5. SUMMARY AND CONCLUSION

The experimental work was carried out at physiology and biotechnology lab, in Animal Production Department, Faculty of Agriculture, Mansoura University, in cooperation with Animal production Research Institute, Agricultural Research Center, Egypt, during the period from September 2016 till August, 2017.

This study aimed to evaluate the effect of supplementation of selenium Nano-particles or *A. harveyi* leaves extract with different concentrations to bull semen extender on sperm characteristics, chromatin damage, ultrastructure changes and apoptosis of spermatozoa, total antioxidant and lipid peroxidation in seminal plasma as well as fertility rate of cryopreserved bull semen.

Five healthy, fertile Friesian bulls were used, and the ejaculates were obtained using an artificial vagina. Semen of all bulls were pooled and diluted in a tris-yolk fructose (TYF) extender supplemented with Se-NPs (1^{st} experiment) or methanol extract of *A. harveyi* leaves (2^{nd} experiment) at concentrations of 0 (control), 0.5, 1.0 and 1.5 µg/ml for a final sperm concentration of 80×10^6 sperm cells/ml diluted semen.

Diluted semen was packed in straws (0.25 ml) and stored in liquid nitrogen (−196 ◦C) for one month. After thawing, semen of each treatment was evaluated for sperm quality parameters, including sperm progressive motility, livability, morphological abnormalities, plasma membrane integrity and chromatin integrity. Apoptosis and sperm ultrastructure were also examined.

Total antioxidant capacity and lipid peroxidation markers were determined in seminal plasma of semen in each treatment. Finally, the effect of Se-NPs on fertilization capacity was checked *in vivo* using n=81cows.

The obtained results can be summarized as follows:

First Experiment: Selenium Nano-particles:-

- The supplementation of Se-NPs at levels of 0.5 and 1.0 µg/ml to semen extender had a positive effect on sperm progressive motility, livability and membrane integrity as compared to the control.
- Adding Se-NP at a level of 1.0 µg/ml improved percentage of viable sperm, while decreased percentages of early apoptotic, apoptotic and necrotic sperm cells as compared to the control.
- Total antioxidants capacity (TAC) in seminal plasma increased and malondialdhyde (MDA) concentration decreased in semen supplemented with Se-NPs up to 1.0 µg/ml.
- Addition of Se-NPs with concentration of 1.5µg/ml had negative effect on all parameters studied.
- Treated bull semen extender with Se-NPs at a level of 1 µg/ml improved *in vivo* fertility rate (90%) compared with control (59%).

Second Experimental: *A. harveyi* Leaf extract:-

- The leaf extract ameliorated the damaging effects of the frozen-thawing process in cryopreserved bull semen. In a dose dependent pattern, sperm motility, viability, and membrane integrity were improved compared to the untreated control.
- Furthermore, the extract increased the percentage of viable sperm and decreased the percentages of early apoptotic and apoptotic sperm cells as well as the damage in sperm ultra-structure.
- These activities are in agreement with the robust antioxidant properties *in vitro* and in the seminal fluid as observed in the total antioxidant capacity and the lipid peroxidation parameter malondialdehyde.

Conclusion

In conclusion, an overall improvement in progressive motility, livability, chromatin damage, sperm ultrastructure and *in vivo* fertility rate was observed when bull semen was extended with TEY supplemented with 1 µg/ml of Se-NPs in during cryopreservation which appears to have positive impact on antioxidant activity. These encouraging results suggest that Se-NPs could be easily included in the freezing protocols to improve overall quality of spermatozoa to enhance in vivo fertility rates in Friesian bull.

Also, the *A. harveyi* leaf extract is rich in flavonoids namely myricetin, quercetin and kaempferol glycosides and polyphenol's. The extract exhibited strong antioxidant activities in DPPH and FRAPS assays, as well as, in cryopreserved bull semen against the deleterious effects of freezing - thawing process when semen was supplemented with 1.5 µg/ml of Albizia harveyi leaf extract in the tris egg yolk fructose extender. Albizia harveyi is a promising plant with potential improve in cryopreservation with oxidative stress during freezing semen.