

# Accepted Manuscript

Does MnTBAP ameliorate DNA fragmentation and in vivo fertility of frozen-thawed Arabian stallion sperm?

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PII: S0093-691X(17)30561-7

DOI: [10.1016/j.theriogenology.2017.11.019](https://doi.org/10.1016/j.theriogenology.2017.11.019)

Reference: THE 14351

To appear in: *Theriogenology*

Received Date: 15 August 2017

Revised Date: 15 November 2017

Accepted Date: 20 November 2017

Please cite this article as: Shojaeian K, Nouri H, Kohram H, Does MnTBAP ameliorate DNA fragmentation and in vivo fertility of frozen-thawed Arabian stallion sperm?, *Theriogenology* (2017), doi: 10.1016/j.theriogenology.2017.11.019.

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1 **Does MnTBAP ameliorate DNA fragmentation and in vivo fertility of frozen-thawed**  
2 **Arabian stallion sperm?**

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10 **Abstract**

11 Overproduction of reactive oxygen species during sperm freeze-thawing process leads to  
12 membrane lipid peroxidation, DNA damage, motility loss, and subsequent death. This oxidative  
13 stress can be alleviated by the addition of some antioxidants to semen extenders prior to freezing.  
14 This study was performed to evaluate the in vitro effectiveness of MnTBAP (a cell permeable  
15 antioxidant) on stallion sperm freezability and in vivo fertility rate. Twenty-one ejaculates were,  
16 collected with missouri model artificial vagina (n = 3 stallions, seven ejaculate each), and diluted  
17 (1:2 v/v) with phosphocaseinate base INRA extender, containing 0 (control), 100, 200 and  
18 300 $\mu$ M of MnTBAP and frozen using a controlled-rate freezing system. The following  
19 parameters were determined: sperm motility, viability, membrane integrity, acrosome  
20 abnormalities, lipid peroxidation and DNA fragmentation. MnTBAP improved horse semen  
21 quality parameters in a dose-dependent manner. The 100 $\mu$ M concentration of MnTBAP did not  
22 show a significant difference in semen parameters compare with control group ( $p > 0.05$ ).  
23 Accordingly, the extender supplemented with 200 $\mu$ M resulted in higher sperm total and  
24 progressive motility ( $55.3 \pm 4.28\%$  and  $33.2 \pm 2.90\%$ ), viability ( $43.9 \pm 2.14\%$ ), and membrane  
25 integrity ( $50.8 \pm 2.14\%$ ), provided a greater protective effect in the percentage of total  
26 abnormalities compare to other groups ( $p < 0.05$ ), and showed lower sperm with damaged DNA  
27 with lower MDA levels ( $p < 0.001$ ). Higher concentrations (300 $\mu$ M) not only did not improve the  
28 results but inversely affected sperm parameters. Twelve mares were used for fertility trial in the  
29 cross over study of 60 deep horn inseminations performed using control (9/30 pregnancy/mare)  
30 and 200 $\mu$ M - MnTBAP (14/30 pregnancy /mare) groups frozen semen. The Average pregnancy

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