



# trial for cryopreservation of rumen protozoa in sheep

H.M.Abdelaziz, H.Y.Hassan, Y.M.Abd-El-Raof, Hala A.A.Abou-Zeina and S.A.Galbt

## Abstract:

Rumen protozoa play an important role in the digestion of cellulose, protein and regulation of rumen pH as well as elimination of pathogen from GIT. Because of their central biological importance, this work aimed to cryopreserve rumen protozoa to establish a rumen protozoal bank followed by using this cryopreserved rumen protozoa in refaunation of experimentally-defaunated sheep. Therefore two experiments were conducted. In the first experiment, the rumen fluids were collected from slaughterhouse and from live sheep by stomach tube and were filtered and mixed with either one of three cryoprotectants: glycerol, ethylene glycol and Dimethyl sulfoxide (DMSO) with different concentration (4, 5, and 5%). The method used was the slow two-step freezing method in which straws containing the mixture of rumen protozoa and cryoprotectant were placed in a cooling water bath at 5°C for 30 minute and then hold in nitrogen vapor for 45 minute (holding time) then directly immersed in liquid nitrogen. Viability was checked monthly till 6 month post-freezing. Results showed that the viability was maximal when using the DMSO (5%) as a cryoprotectant. Consequently, the second experiment was conducted in which 9 female sheep were allocated into 3 equal groups. The first group served as control. The second group was defaunated by single oil drench (cooking oil at 5ml / kg BW). The third group was defaunated by oil drench followed by refaunation by intraruminal inoculation of the content of one straw that contains cryopreserved rumen protozoa with DMSO 5% as a cryoprotectant. Result showed that refaunation successfully regained the total protozoal count to near control value, significantly increased the ammonia nitrogen concentration, body weight gain and the rumen pH after being reduced by defaunation. Therefore, it was concluded that successful cryopreservation of rumen protozoa in sheep can be attained by slow two-step freezing method and that cryopreserved protozoal bank could be successfully used for refaunation of sheep that have been defaunated due to acidosis. Consequently, this cryopreserved protozoal bank can be employed in farms to improve digestibility, increase body weight and treat rumen acidosis.

## Keywords:

cryopreservation - rumen - protozoa

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