

# Comparison of biochemical parameters of Muscovy drake semen diluted and stored at 4 °C in three buffers

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## Abstract

A comparison study of biochemical parameters of semen from Muscovy drakes diluted and stored at 4 °C in three buffers—IMV-buffer (France), HIA-1 and AU (Bulgaria) was carried out. The ejaculates were collected twice a week from ten 1-year-old Muscovy drakes using laying Muscovy females as teaser. Semen was diluted immediately, respectively, with IMV-buffer, HIA-1 and AU, and cold-stored (4 °C) for 1, 3 and 6 h. The intensities of oxygen uptake at the third hour in semen diluted, respectively, with IMV-buffer ( $200 \pm 1.6$  nAO/10<sup>9</sup> sperm cells min), with HIA ( $224 \pm 44$  nAO/10<sup>9</sup> sperm cells min) and with AU ( $238 \pm 48$  nAO/10<sup>9</sup> sperm cells min) were highly significant in comparison with neat semen ( $75 \pm 0.7$  nAO/10<sup>9</sup> sperm cells min).

The observed intensity of fructolysis was highest when using AU, followed by HIA-1 and IMV-buffer. During the first hour of storage the level of pyruvic acid was significantly lower in semen diluted with Bulgarian extenders, and this stability for AU referred to the entire period. For lactic acid, the differences were not statistical significant. Our investigations do not show significant differences concerning the dynamics of inorganic phosphate and total lipids after dilution with all tested extenders. On the contrary, high increase of cholesterol efflux from spermatozoa to seminal plasma—diluent were obtained after 6 h of storage.

All extenders, IMV-buffer (France), HIA-1 and AU (Bulgaria) for diluting and short time storage of semen from Muscovy drakes at 4 °C maintain the necessary comfort of energy metabolism of the spermatozoa.

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## 1. Introduction

The neat drake semen, several hours after collection, is inadequate to allow for artificial insemination (Choi and Song, 1998; Kasai et al., 2001). The use of semen extenders with suitable ingredients and physical–chemical parameters optimizes energy metabolism and preserves the fertility of the male gametes. It is a guarantee for obtaining high reproductive results by artificial insemination of waterfowls (Bottwalla and Miles, 1992).

In this aspect the biochemical assessment of fresh and stored drake semen quality is an additional method which can be used. The investigations of Sexton (1984) and Atanasov et al. (1994, 1998, 1999) established intensive oxygen consumption of rooster and tom spermatozoa. Glycolysis and the tricarboxylic acid cycle (TCA cycle) appear to be the main source of adenosine triphosphate (ATP) for spermatozoa. But as for the drake semen there is no data about sperm glycolysis, respiration and oxidative phosphorylation. Surai and Wishart (1996) suggested that in drake and gander spermatozoa, mainly anaerobic metabolic processes dominated and their markers are higher levels of lactate dehydrogenase (LDH) isozymes 4 and 5, than 1 and 2. In this aspect the investigation of fructose, pyruvate, lactate, inorganic phosphate and oxygen consumption is compulsory and will describe the intensity of Embden–Meyerhof pathway and oxidative phosphorylation.

The technology of artificial insemination for producing *Mule ducks* includes some processes—obtaining ejaculates, quality estimation, dilution, and semen application carried out mainly in aerobic conditions that influence the fertility of gametes (Gvoryahu et al., 1984).

According to Penfold et al. (2001) cold storage of drake semen provides an effective means of short-term storage with no loss of fertility.

The purpose of the study was to establish the influence of the IMV-buffer (France) and two newly created by Gerzilov (2002) semen extenders HIA-1 and AU on some biochemical parameters in short-time cool-stored (4 °C) Muscovy semen.

## 2. Materials and methods

Semen was collected from ten 1-year-old Muscovy drakes (White variety) twice a week using laying Muscovy females as teaser (Tan, 1980; Gerzilov, 2000). During the reproductive period (April–July) the males were kept in individual cages with size 0.6/0.8/0.6 m. Sperm concentration, sperm motility and live/dead spermatozoa were determined for each ejaculate by standard methods (Bakst and Cecil, 1997). Only good quality ejaculates (color, pearly white; purity, free of any contamination with cloacal products; volume, above 0.3 ml; sperm motility, above 70%, sperm concentration, above  $1 \times 10^9$  sperm cells/ml) have been used for the study.

The pooled semen was divided into four equal parts. One part of the semen was separated as undiluted, and the others were diluted in a ratio 1:1 (v/v) with the IMV-buffer (patented by IMV-Technologies, France), and the newly created extenders HIA-1 and AU (Gerzilov, 2002):

- The HIA-1 extender consists of 0.25 g D-glucose, 0.25 g D-fructose, 0.07 g saccharose, 0.50 g sodium citrate, 9.00 g sodium chloride, and 100 ml double distilled water. The osmolarity was 290 mOsmol/kg and pH 7.00.
- The AU extender consists of 0.40 g D-glucose, 0.80 g D-fructose, 0.80 g saccharose, 0.90 g sodium citrate, 0.84 g sodium glutamate, 0.40 ml glycerol, 0.04 g ethylene diamine tetra acetic acid disodium salt dihydrate, and 100 ml double distilled water. The osmolarity was 320 mOsmol/kg and pH 7.00.

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