

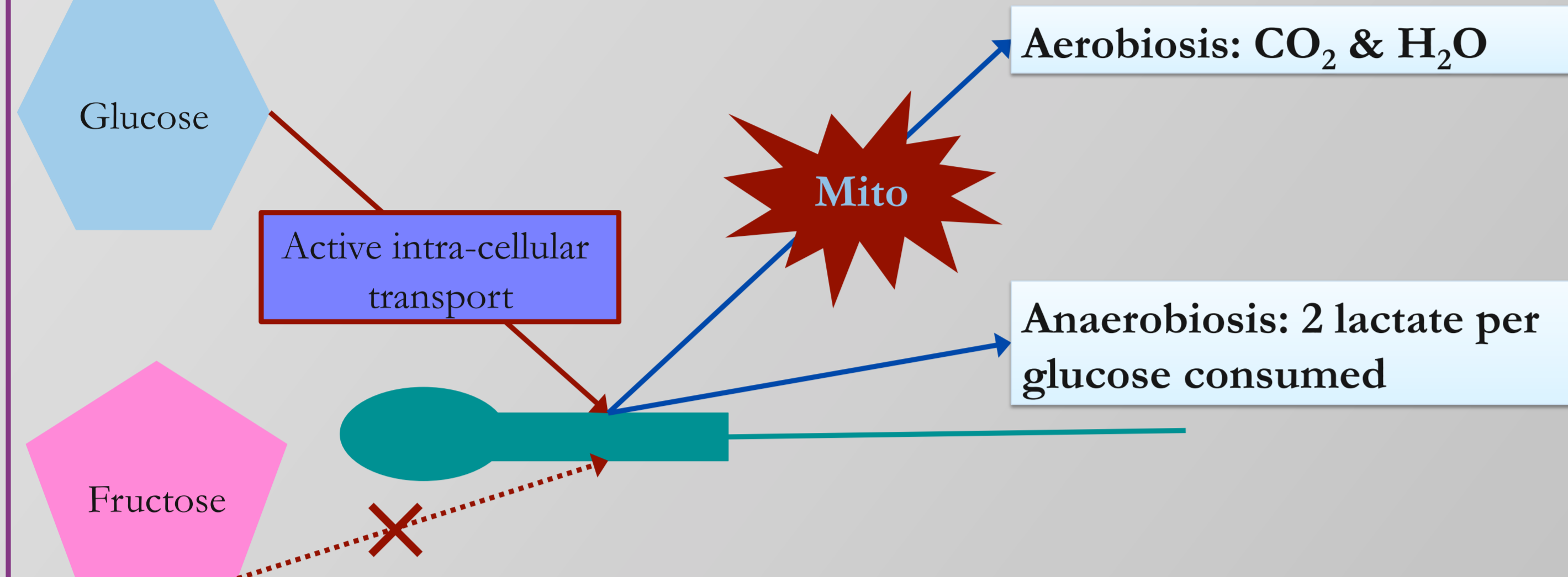
SPERM MOTILITY AND LACTATE PRODUCTION AT DIFFERENT SPERM CONCENTRATIONS

Introduction

Use of highly concentrated fresh semen:

- Deep horn insemination for low fertility mare & low quality fresh semen
- No data available about conservation
 ➔ Shipping = ?

Data about equine semen metabolism:



Material and methods

Animals: 1 Pony stallion, 4 sport stallions

Experimental design:

Semen collected 4 times

- Raw semen analysis (Concentration & motility with CASA)
- Volume containing 110, 440, 880 x 10⁶ spz sampled
- Dilution (1/4 semen, 3/4 INRA96[®])
- Cushioned centrifugation (Ioxidanol, Maxifreeze[®])
- Sperm-rich pellet re-suspended in 1ml of supernatant
- Motility analysis after 8 & 24 hours:
 - *Non Progressive Motility (NPM)*
 = Total motility – Progressive motility
 - *Preservation of Total Motility (PTM)*
 = Total Motility at 8 or 24 hours / Total Motility in raw semen
 - *Preservation of Conservation of Progressive Motility (PPM)*
 = Progressive Motility at 8 or 24 hours / Progressive Motility in raw semen

Lactate assay:

- After centrifugation (t0) and after 24h (t24)
- ¹H Nuclear Magnetic Resonance integrating the methyl signal of lactate

Statistical methods:

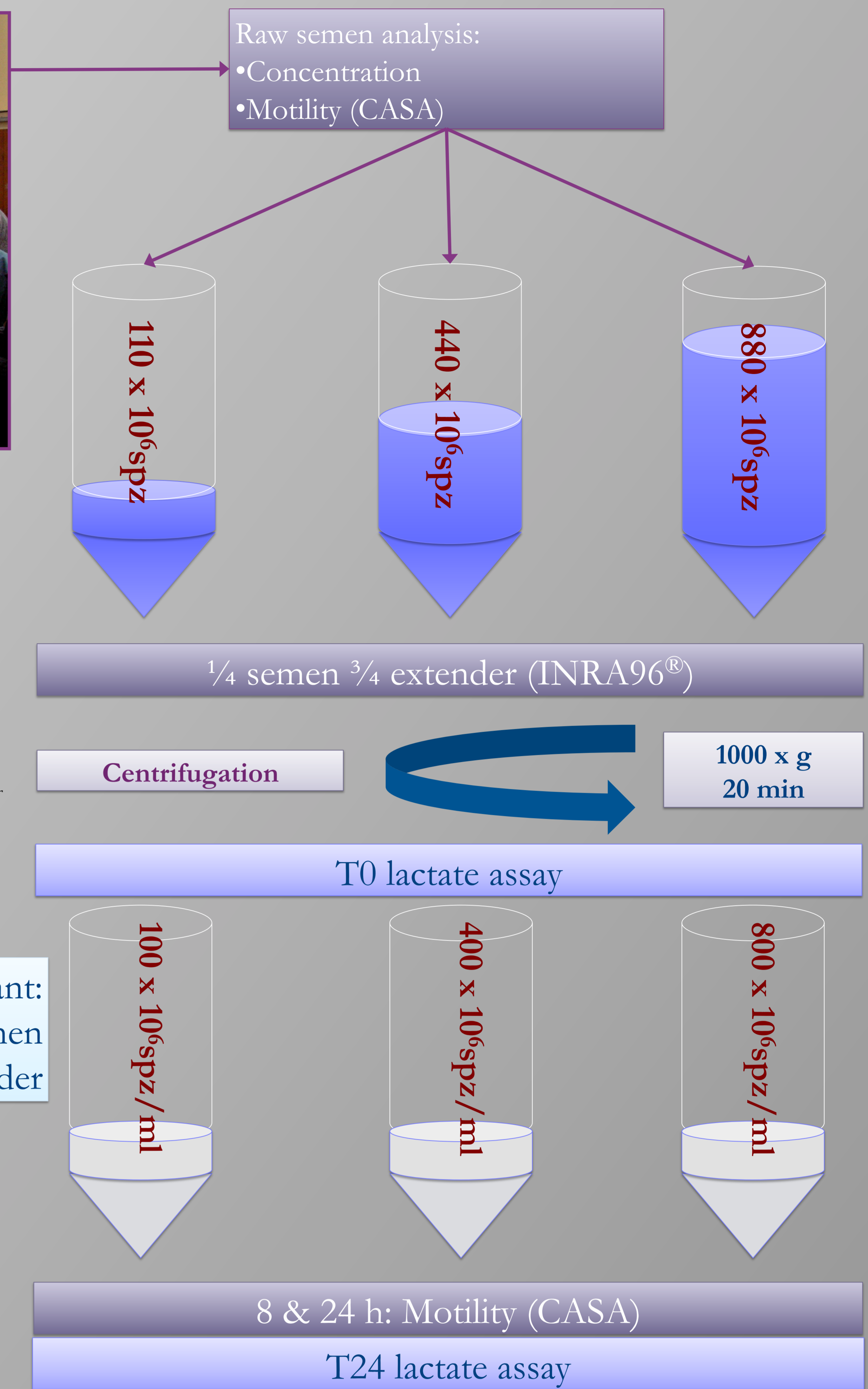
Friedmann test and Dunn's post-test for median differences – Spearman test for correlations

Results

- Spermatozoa recovery rate lower in low concentration samples (p<0.001)
- Final concentrations in groups: 70.45±30.59, 434.82±120.02 and 879.97±241.15x10⁶ spz/ml
- Lactate concentration at t0:
 - No differences between groups
 - Directly correlated to raw semen volume (r=0.5032; p=0.0103)
- PTM decreases after 8 hours of conservation in 800x10⁶ spz/ml samples (p<0.001)
- PPM decreases after 24 hours of conservation in 800x10⁶ spz/ml samples (p<0.001)
- PTM & PPM after 24h negatively correlated to lactate at t0 (r=-0.4568; p=0.0217 and r=-0.4684; p=0.0182)
- Concentration of TM and PM after 8 & 24h negatively correlated to lactate concentration at t0 and t24
- Concentration of NPM (non-progressive motile spermatozoa) after 24h is directly correlated to lactate concentration after 24h (r=0.6767; p<0.0001)

Conclusions

- Lactate concentration at T0 is correlated to volume of semen showing an influence of semen collection conditions
 ➔ Effect of lactate concentration at T0 on motility after 24h
- Conservation with high concentration is rapidly (8 hours) deleterious for Total motility whereas Progressive motility is only decreased after 24 hours
 ➔ **HIGH CONCENTRATION FRESH SEMEN DOES SUITABLE FOR USE WITHIN 8 HOURS**
- High and strong association between (NPM) Non-Progressively Motile spermatozoa and lactate concentration after 24 hours
 ➔ **LACTATE SEEMS TO BE MAINLY PRODUCED BY NON-PROGRESSIVELY MOTILE SPERMATOZOA DURING STORAGE**
- **NON PROGRESSIVELY MOTILE SPERMATOZOA MAINLY USE ANAEROBIC GLYCOLYSIS**



Lactate concentration (mmol/L) according to spermatozoa concentration in samples and storage

Different letters in superscript indicate statistically different values.

