Effect of pH and Storage Conditions on Measured Ionized Calcium Concentration in Horses and Donkeys

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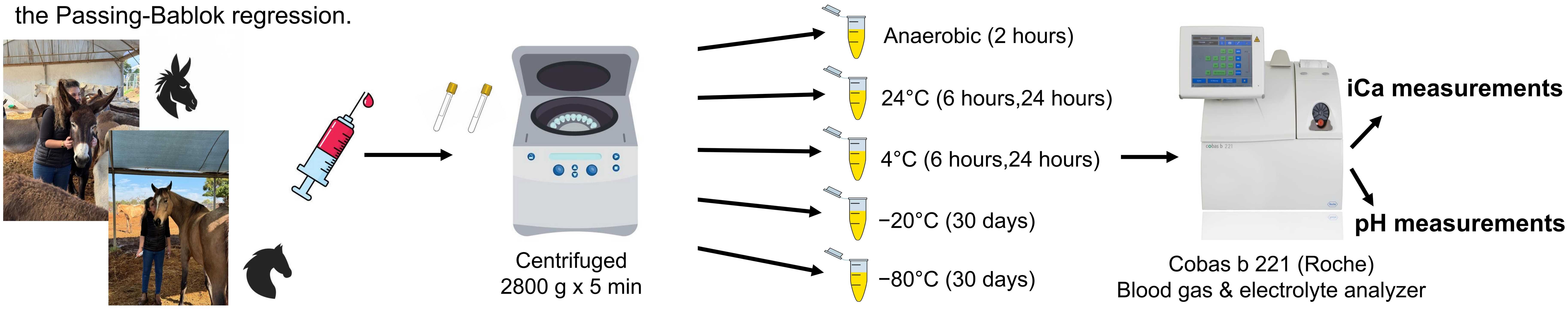
Introduction

- Calcium homeostasis is vital for many physiological functions. Free ionized calcium (iCa) is the physiologically active fraction.
- Measured iCa concentration is influenced by sample pH & calcium-protein binding properties.
- Correction formulae for sample pH & iCa concentration in human & small animal blood samples are available.
- **Aims:**
  1. **Determine effects of sample pH & storage conditions on serum iCa concentration in horses & donkeys.**
  2. **Establish sample pH-dependent correction formulae for measured iCa concentration in aerobically stored samples.**

Methods

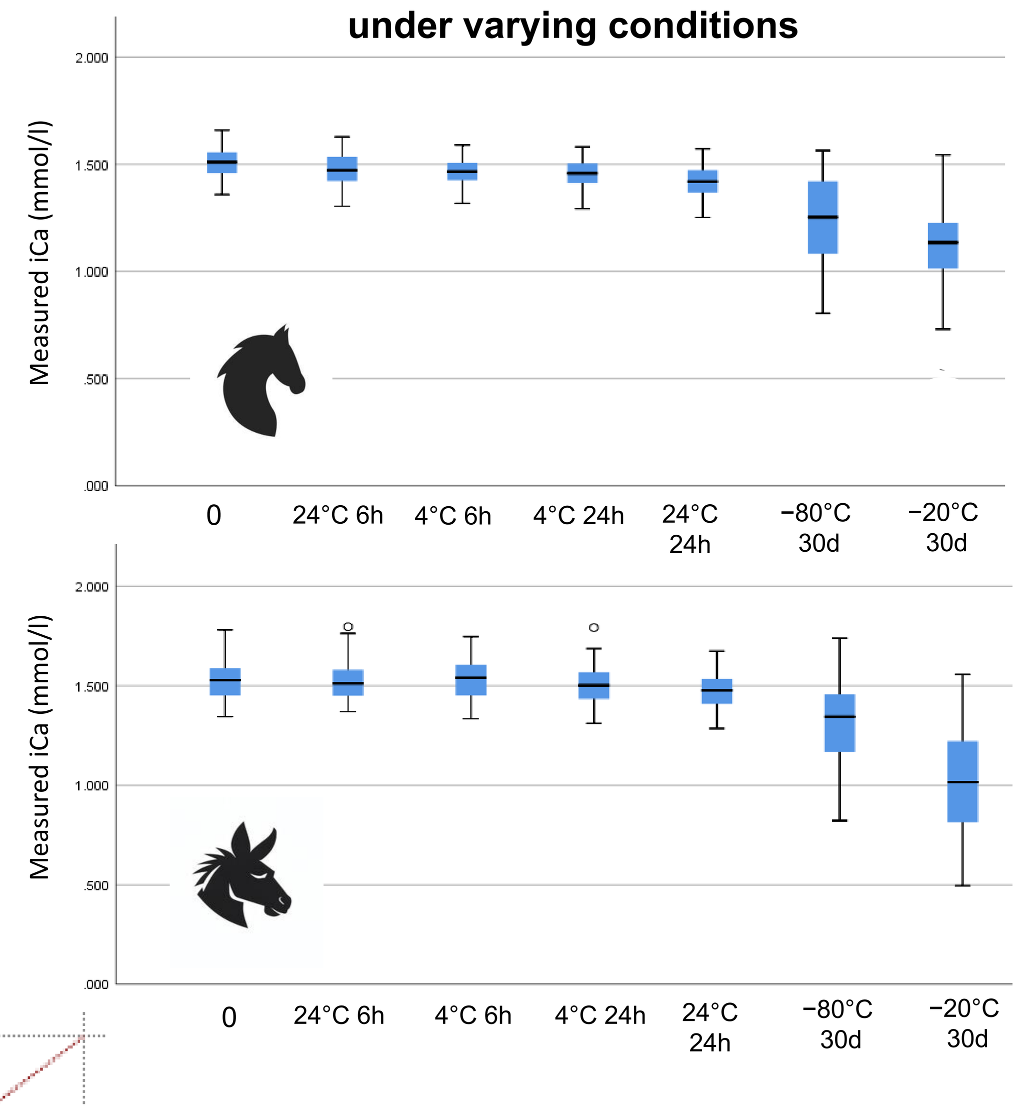
- Serum iCa concentration and sample pH of **40 horses & 53 donkeys** kept anaerobically under cooling conditions were measured within 2 hours from sampling and later, samples were stored under several aerobic conditions.
- Generalized Estimating Equations were used to compare iCa concentrations or pH among the different storage conditions.
- The agreement between the serum calculated 'corrected iCa' and the 'true iCa', measured under anaerobic conditions was determined using the Passing-Bablok regression.



Results

- In horses, iCa concentrations were significantly (P <0.05) lower in all aerobically stored samples compared to anaerobic samples (measured 2 hours from collection).
- In donkeys, iCa concentrations were significantly (P <0.05) lower in frozen stored samples.
- The largest significant differences were noted in samples stored frozen at −20°C & −80°C for 30 days.

iCa measurements samples stored under varying conditions

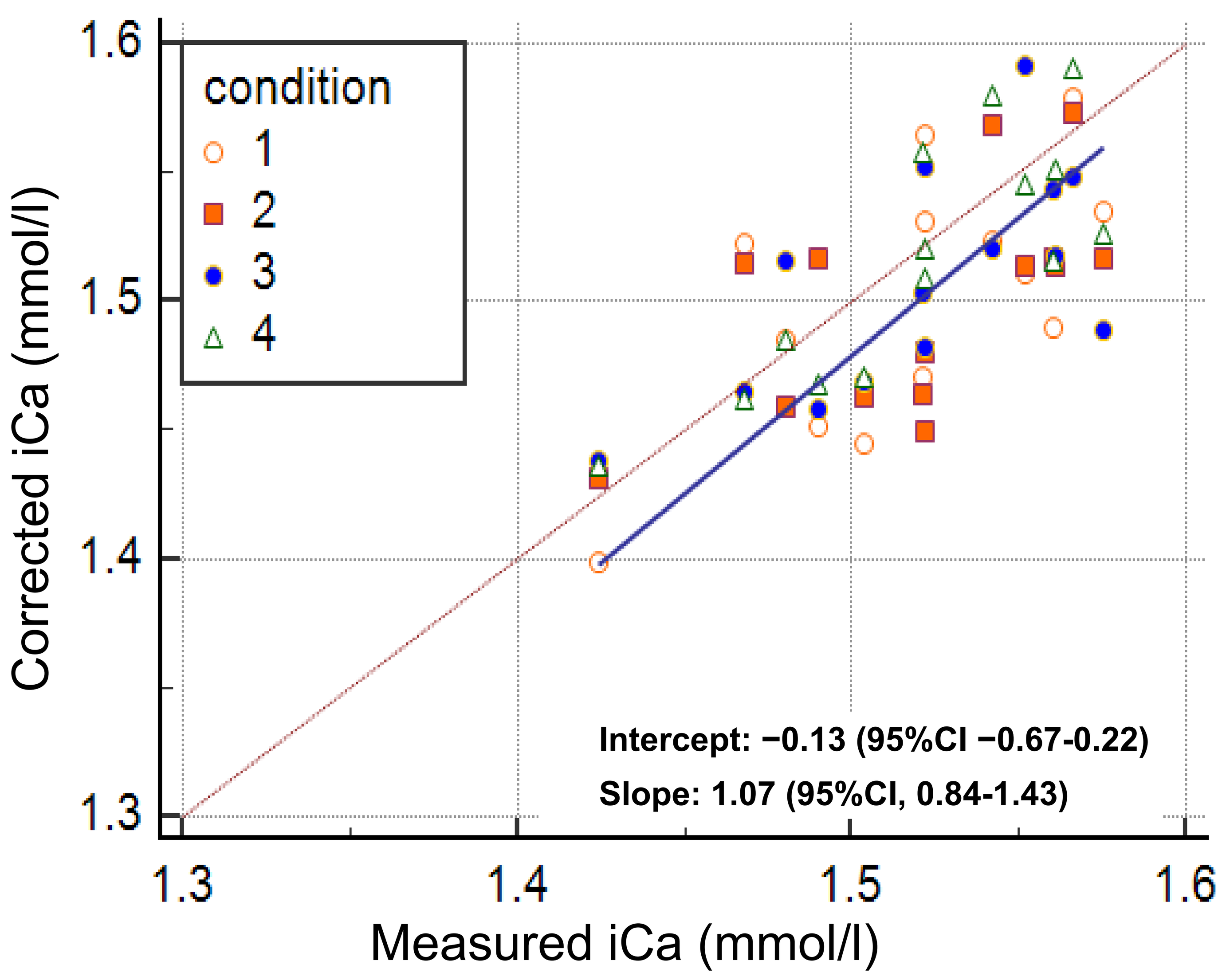


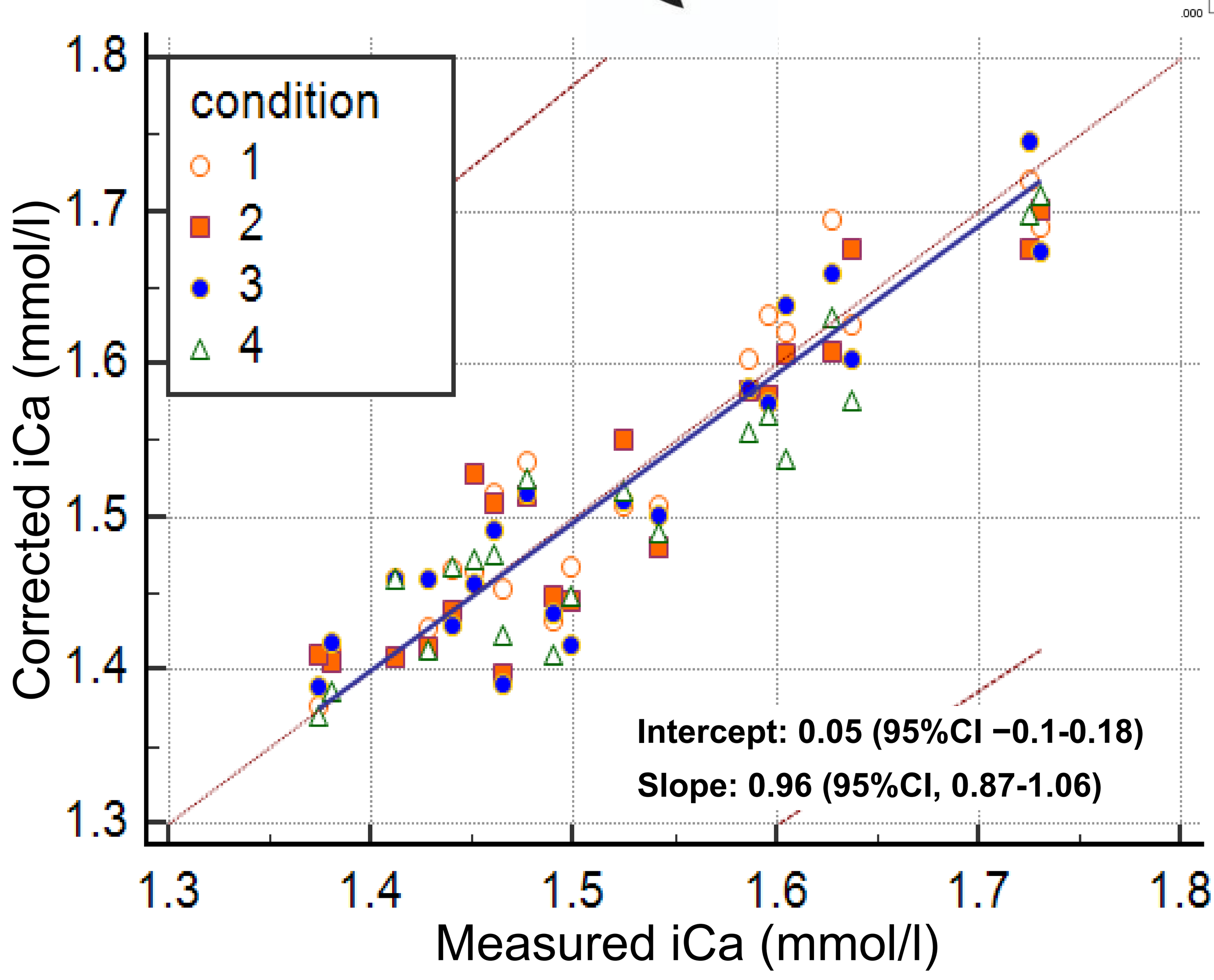
Formula for correcting [iCa] for sample pH:

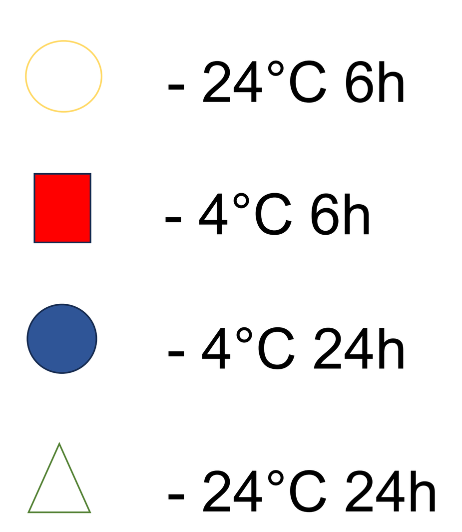
Corrected iCa = measured iCa + β coefficient × (measured pH–mean pH)

Species	β coefficient	Mean pH
Horses	0.295	7.45
Donkeys	0.262	7.42

Passing-Bablok regression







Conclusions

- Horse & donkey sera for iCa measurement can be stored at 4°C or at 24°C for 24 hours.
- Storage at −20°C or −80°C for longer periods leads to statistically & clinically significant lower results, and hence, is not recommended.
- **Correcting measured equine & asinine serum iCa concentration for sample pH can be made using the above-mentioned generated formulae**.
- Future studies: Assess measured & corrected equine sera iCa in hypocalcemic & hypercalcemic animals.

References will be available upon request

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