



THE UNIVERSITY *of* EDINBURGH

Edinburgh Research Explorer

Precision and accuracy of a point-of-care glucometer in horses and the effects of sample type

Citation for published version:

Rendle, DI, Armstrong, S, Heller, J & Hughes, KJ 2019, 'Precision and accuracy of a point-of-care glucometer in horses and the effects of sample type', *The Veterinary Journal*, vol. 252, 105359. <https://doi.org/10.1016/j.tvjl.2019.105359>

Digital Object Identifier (DOI):

[10.1016/j.tvjl.2019.105359](https://doi.org/10.1016/j.tvjl.2019.105359)

Link:

[Link to publication record in Edinburgh Research Explorer](#)

Document Version:

Peer reviewed version

Published In:

The Veterinary Journal

General rights

Copyright for the publications made accessible via the Edinburgh Research Explorer is retained by the author(s) and / or other copyright owners and it is a condition of accessing these publications that users recognise and abide by the legal requirements associated with these rights.

Take down policy

The University of Edinburgh has made every reasonable effort to ensure that Edinburgh Research Explorer content complies with UK legislation. If you believe that the public display of this file breaches copyright please contact openaccess@ed.ac.uk providing details, and we will remove access to the work immediately and investigate your claim.



1 **Precision and accuracy of a point-of-care glucometer in horses and the effects of sample type**

2 David. I. Rendle ^{a, b}, Susan. K. Armstrong ^{a, c}, Jane. Heller ^a, Kristopher. J. Hughes ^{a, *}

3

4 *School of Animal and Veterinary Sciences, Charles Sturt University, Locked Bag 588, Boorooma Street,*
5 *Wagga Wagga, NSW 2678 Australia.*

6 ^b*Current address: Rainbow Equine Hospital, Old Malton, Malton, North Yorkshire, YO17 6SG UK.*

7 ^c*Current address: Royal (Dick) School of Veterinary Studies and Roslin Institute, University of*
8 *Edinburgh, Roslin, Midlothian, EH25 9RG UK.*

9

10 * Corresponding author. Tel.: +61 69334253.

11 *Email address: krhughes@csu.edu.au (K.J. Hughes).*

12

13

14

15

16

17

18

19

20

21

22

23

24

25

26

27

28

29

30

31

32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
61
62

Abstract

Point-of-care glucometry is used commonly in clinical and research settings; however, accuracy and precision of this method are concerns. The objectives of this study were to determine the accuracy of glucometry in adult horses and the precision of duplicate measurements. Blood samples were collected from 62 horses into one plain syringe, one ethylenediaminetetraacetic acid (EDTA) tube and three fluoride oxalate (FO) tubes. Immediately after collection, glucose concentrations in whole blood were determined, in duplicate, by glucometry from the syringe (plain whole blood [WB] group), EDTA tube (EDTA group) and one FO tube (FO group). One FO sample was used to measure plasma glucose concentration by a laboratory chemistry analyser (LAB group) ≤ 1 h after collection. The third FO tube was used to measure plasma glucose concentration by glucometry after 3 h storage (FO3hr group).

Adequate precision was present for all groups (coefficient of variation: 0.7-3.5%) except WB (5.5-9.4%). Between groups, correlations were significant ($P < 0.05$) (except WB-EDTA), varied with group comparison and tended to be lowest for comparisons involving WB. Mean bias was lowest for WB-LAB and greatest for FO-LAB and FO3hr-LAB; however, the limits of agreement were ≥ 4.65 mmol/L for WB-LAB and ≤ 2.75 mmol/L for most other comparisons. For the glucometer used, performance is influenced by sample type: WB is unsuitable, while FO or EDTA samples result in adequate precision and accuracy, provided under-estimation of glucose concentrations is accounted for by using method-specific reference ranges. Glucometer performance and optimal sample type(s) should be determined prior to use in horses.

Keywords: Horse; Glucometry; Repeatability; Agreement; Equine

63

64

65 **Introduction**

66 Regulation of blood glucose concentrations is important in critically-ill horses, as derangements
67 in glucose concentrations are associated with increased mortality (Hassel et al., 2009; Hollis et al.,
68 2007; Hollis et al., 2008a). Hyperglycaemia is a negative prognostic indicator in critically-ill human
69 patients, supporting regulation of blood glucose concentration through monitoring and intervention
70 (Bochicchio et al., 2005; Sung et al., 2005; Vogelzang et al., 2006). Point-of-care glucometry has
71 become standard in human and equine intensive care settings and testing of insulin dysregulation,
72 given the ease of measurement, small blood volumes required, and ability for immediate decision-
73 making (Hoedemaekers et al., 2008; Hollis et al., 2008b; Russell et al., 2007; Tack et al., 2012).
74 However, for optimal patient management, the precision and accuracy of individual glucometers needs
75 to be determined. Despite extensive use of glucometry in intensive care units and diabetes control in
76 human medicine, conflicting results of glucometer performance have been reported. In some studies,
77 glucometry was an accurate alternative to laboratory methods (Ray et al., 2001; Tack et al., 2012).
78 However, considerable glucometer inaccuracy was reported in other studies (Denfeld et al., 2011;
79 Hoedemaekers et al., 2008; Kanji et al., 2005), with implications for incorrect decision-making when
80 tight glycaemic control is necessary. Possible explanations for the disparity in glucometer performance
81 include haematocrit interference, sample type (whole blood/plasma, capillary/venous/arterial blood) and
82 analyser type (Gerber and Freeman, 2016; Kanji et al., 2005; Tang et al., 2000).

83

84 There are few published studies of the accuracy and precision of glucometers in horses
85 (Hackett and McCue, 2010; Hollis et al., 2008b; Hug et al., 2013; Russell et al., 2007) and only one
86 veterinary-specific glucometer has been validated for horses (Hackett and McCue, 2010). The
87 influences of sample type, glucose concentration and delays in analysis on results in horses are largely
88 unknown. In one equine study, glucometry with plasma had excellent agreement with a laboratory
89 method, while measurements from whole blood did not (Hollis et al., 2008b). In some human and equine
90 studies, glucometer performance was less reliable when marked hypoglycaemia or hyperglycaemia
91 was present (Hollis et al., 2008b; Kanji et al., 2005; Khan et al., 2006). Bias in glucometer results may
92 be important in critically-ill horses where performance at the extremes of blood glucose concentrations

93 may influence decision-making and accurate glycaemic control. The objectives of this study were to
94 determine (1) the accuracy of glucometry in adult horses using blood and plasma samples in
95 comparison to a laboratory standard method and (2) the precision of duplicate measurements.

96

97 **Materials and methods**

98 The study was granted approval by the Animal Care and Ethics Committee, Charles Sturt
99 University (Approval number 11/043; Approval date September 2011).

100 *Sample collection*

101 Blood samples were obtained from horses that were >1 year of age and from the research herd
102 of Charles Sturt University (CSU) or presented to the Veterinary Clinical Centre, CSU, for clinical
103 assessment.

104

105 For each horse, whole blood (WB) was collected from the jugular vein into a 3 mL syringe, one
106 blood collection tube containing ethylenediaminetetraacetic acid (EDTA) (Vacutainer, Becton
107 Dickinson) and 3 blood collection tubes containing fluoride oxalate (FO) (Vacutainer, Becton Dickinson).
108 The tubes were filled to the manufacturer's designated level. Immediately after collection, the glucose
109 concentration of WB in the syringe was determined using a human-specific POC glucometer (Accutrend
110 Plus, Roche Diagnostics), according to the manufacturer's instructions (WB group). Within 5 min of
111 blood collection, the blood glucose concentrations in the EDTA tube (EDTA group) and one of the FO
112 tubes (FO group) was determined using the glucometer. The second FO sample was used for
113 determination of plasma glucose concentration using the glucometer after storage of the sample for 3
114 h at 22 °C and separation of the plasma by centrifugation (10 min at 3000 rpm) immediately prior to
115 analysis (FO3hr group). The third FO tube was used to measure plasma glucose concentration by a
116 wet chemistry analyser using the hexokinase/glucocose-6-phosphate dehydrogenase method (Konelab
117 30i, Thermo Fisher Scientific) as the reference method, within 1 h of blood collection and after
118 centrifugation (10 min at 3000 rpm) (LAB group). For every sample, two glucose concentration
119 measurements were obtained using the same device. For the WB, EDTA, FO and FO3hr groups, the
120 same lot number for the glucometer measurement strips and a single glucometer was used throughout
121 the study. The glucose measurement strips were stored according to the manufacturer's instructions.

122

123 Using the LAB results, samples were categorised as euglycaemic (4.0-7.0 mmol/L) or hyperglycaemic
124 (>7.0 mmol/L). Raw glucose concentration data for all 62 horses are provided in Appendix A:
125 Supplementary material.

126

127 *Data analysis*

128 Within groups, precision of glucose measurement was determined by calculation of the
129 repeatability coefficient (RC: 1.96 times the SD of the differences of paired measurements) (Bland and
130 Altman, 1986). The RC is expected to contain 95% of the differences, and values of <0.8 mmol/L and
131 <2.0 mmol/L were considered acceptable for euglycaemic and hyperglycaemic samples, respectively,
132 based on criteria for glucometer performance in humans (Tonyushkina and Nichols, 2009). Within each
133 group, the coefficient of variation (CV) for repeated glucose measurements was determined: a median
134 CV of <4% was considered adequate measurement precision (Carr et al., 1995; Chen et al., 2003).
135 Blood glucose measurements within each group were compared also using Spearman's correlation as
136 data were non-normally distributed (Shapiro-Wilk test). Spearman's correlation coefficients (r_s) were
137 interpreted using the criteria of 0.90-1.00: very high, 0.70-0.89: high, 0.50-0.69: moderate, 0.30-0.49:
138 low and <0.30: little/no correlation (Domori et al., 2014).

139

140 Between groups, association of paired glucose measurements (first measurement for each
141 sample) was determined using Spearman's correlation, and agreement was determined using Lin's
142 concordance correlation (Lin, 1989) and the mean bias and 95% limits of agreement (LOA) (Bland and
143 Altman, 1986). Concordance correlation coefficients (ρ_c) were interpreted using the criteria of >0.99:
144 almost perfect, 0.95-0.99: substantial, 0.90-0.95: moderate, <0.90: poor concordance (Domori et al.,
145 2014). Based on criteria for glucometer accuracy for humans (Tonyushkina and Nichols, 2009),
146 adequate agreement was considered with LOA ≤ 1.7 mmol/L and ≤ 2.8 mmol/L for euglycaemic and
147 hyperglycaemic samples, respectively. The relative difference between paired glucose measurements
148 for each glucometer group and the LAB group was calculated (POC group-LAB/LAB x 100%) and the
149 percentage of glucometer results that were within 5%, 10%, 15% or 20% of the LAB glucose
150 concentration were calculated according to guidelines to describe glucometer performance (Krouwer
151 and Cembrowski, 2010; Tennent-Brown et al., 2011; Tonyushkina and Nichols, 2009).

152

153 All statistical analyses were performed using SPSS Statistics version 20 (IBM). Significance was set at
154 $P < 0.05$.

155

156 **Results**

157 Blood samples were obtained from 62 horses (30 Thoroughbreds, 19 Standardbreds, 5 Quarter
158 horses, 4 ponies, 3 Arabians, 1 Warmblood and 1 Clydesdale) with a median age of 9.5 years (range
159 1-23). The study population consisted of 33 geldings, 6 entire male horses and 23 mares. Forty-two
160 animals were research horses, of which 10 underwent combined glucose and insulin testing performed
161 for the investigation of insulin dysregulation and had hyperglycaemia. Twenty horses were client-owned
162 animals that had blood collected for clinical assessment. Forty-nine and 13 horses were euglycaemic
163 and hyperglycaemic, respectively. No horses were hypoglycaemic.

164

165 For euglycaemic and hyperglycaemic samples, RC and CV values indicated adequate
166 repeatability for all groups with the exception of WB (Table 1). For all glucose concentration categories,
167 the RC and CV values were smallest and largest for the LAB and WB groups, respectively. For all
168 glucose measurements, r_s was significant and the strength of correlation was moderate to very high
169 (Table 1).

170

171 Between groups, r_s was significant for all comparisons, with the exception of WB-EDTA for
172 euglycaemic samples (Table 2). Correlation and concordance of glucose measurements varied
173 substantially with group comparison and glucose concentration (Table 2). For all categories, r_s and
174 concordance tended to be lowest for comparisons between WB and other groups. The bias and 95%
175 LOA for agreements between groups are provided in Table 3. Bland-Altman plots are provided in
176 Appendix A: Supplementary material.

177

178 No group had 95% of the glucose measurements within 5-20% of the LAB results, with the
179 exception of EDTA for hyperglycaemic samples (Table 4).

180

181 **Discussion**

182 The results of this study demonstrate that glucometer precision and accuracy can be influenced
183 by blood sample type and storage. Poor glucometer performance occurred with WB without
184 anticoagulant, while anticoagulated blood (EDTA, FO) and plasma (FO3hr) resulted in improved
185 glucometer repeatability and agreement with the laboratory standard, albeit with underestimation of
186 glucose concentration. In addition, precision and accuracy varied across the glycaemic range,
187 supporting the recommendation that glucometer performance should be assessed in clinically-relevant
188 glucose ranges (hypo-, eu- and hyperglycaemia) (Gerber and Freeman, 2016). Determination of
189 glucometer performance is important for patient care, particularly in intensive care settings. In both
190 humans and horses, glycaemic abnormalities are associated with a worse prognosis for survival
191 (Bochicchio et al., 2005; Hassel et al., 2009; Hollis et al., 2008a; Sung et al., 2005) and monitoring and
192 regulation of blood glucose concentrations is associated with improved outcomes in critically-ill humans
193 (Lewis et al., 2004; Sung et al., 2005). While glucometry provides rapid results for immediate patient-
194 side decision making, poor accuracy and precision may lead to erroneous clinical assessment and
195 management (Hoedemaekers et al., 2008; Kanji et al., 2005; Russell et al., 2007).

196

197 For all categories, WB had poor precision (CV: 5.5-9.4%, RC: 2.14-2.94 mmol/L). For
198 euglycaemic animals, a difference of over 2.14 mmol/L may alter clinical interpretations, suggesting that
199 WB is inappropriate for this glucometer. The reason for this poor precision is unknown; however, it may
200 involve the interaction of horse erythrocytes with the test strip. The strips filter erythrocytes while plasma
201 diffuses to the reagents and sensor for glucose measurement and differences in the sizes of human
202 and horse erythrocytes and rouleaux formation may be sources of pre-analytic error when WB was
203 used. Further, glucometers designed for humans assume a stable relationship of glucose
204 concentrations between erythrocytes and plasma, and differences in glucose distribution between
205 humans and horses are likely (Coldman and Good, 1967; Hackett and McCue, 2010). In one study
206 (Hackett and McCue, 2010), a veterinary glucometer coded for equine use (and likely species-specific
207 assumption of glucose distribution) accurately measured glucose concentrations in WB, while in other
208 studies, plasma provided superior agreement with a laboratory method than WB when human-specific
209 glucometers were used in horses (Hollis et al., 2008b) and alpacas (Tennent-Brown et al., 2011). These
210 findings indicate that the performance of individual glucometers, including influence of sample type,

211 should be assessed prior to use in horses, as discrepancies in accuracy and precision may influence
212 decision-making.

213

214 For both euglycaemic and hyperglycaemic samples, the RC and CV values for the EDTA, FO
215 and FO3hr groups were consistent with acceptable precision. Glucometer precision in equine practice
216 has been reported once previously, where the CV of a veterinary glucometer that uses WB was 1.3%
217 (Hackett and McCue, 2010). Using described values (<3.5-4%) for glucometry in human medicine (Carr
218 et al., 1995; Chen et al., 2003), the CVs for EDTA, FO and FO3hr in our study indicate that the precision
219 of the glucometer with these sample types is adequate for horses, while WB resulted in poor precision.

220

221 When all samples were considered, all glucometer groups had moderate or high correlation
222 with LAB. However, r_s increases with a greater range of measured values (Bland and Altman, 1986)
223 and the strengths of association may have been artificially improved. Importantly, only low to moderate
224 correlations between groups and LAB were present for euglycaemic samples, emphasising the
225 importance of determining glucometer performance within clinically-relevant glucose concentration
226 ranges. Similarly, ρ_c values were influenced by glucose concentration category, and demonstrated poor
227 agreement of the glucometer with LAB for euglycaemic samples and often poor agreement between
228 glucometer groups for euglycaemic and hyperglycaemic samples, suggesting that results should not be
229 used interchangeably with reference laboratory methods or when different sample types are used.
230 However, the magnitude of the lack of agreement cannot be determined using this method.

231

232 The mean bias and 95% LOA results revealed variable agreement between glucometer groups
233 and LAB. Whole blood had the poorest agreement (LOA ≥ 4.7 mmol/L), representing differences with
234 risk of clinical misinterpretation. Similarly, when used in human-specific glucometers, WB was
235 associated with poor agreement with a laboratory method in foals (Russell et al., 2007), adult horses
236 (Hollis et al., 2008b), alpacas (Tennent-Brown et al., 2011) and dogs (Cohen et al., 2009). In contrast,
237 good accuracy of veterinary-specific glucometers with WB has been demonstrated for horses (Hackett
238 and McCue, 2010) and alpacas (Tennent-Brown et al., 2011), which may reflect more accurate
239 programmed assumptions of glucose distribution between plasma and erythrocytes. The selection of
240 acceptable LOA is a clinical, rather than a statistical decision, and for the current study, <1.7 mmol/L

241 (euglycaemic samples) and <2.8 mmol/L (hyperglycaemic samples) were chosen, based on glucometer
242 performance criteria for humans (Tonyushkina and Nichols, 2009). Using these LOA values, acceptable
243 agreement was present for LAB-EDTA, LAB-FO, LAB-FO3hr and FO-FO3hr (euglycaemic samples)
244 and LAB-FO, LAB-FO3hr and FO-FO3hr (hyperglycaemic samples). The FO and FO3hr groups had
245 the smallest LOA ranges, which are unlikely to alter clinical interpretation, irrespective of glucose
246 concentration. While acceptable agreement between the EDTA and LAB groups was present for
247 euglycaemic samples, proportional bias with increasing glucose concentration was present, suggesting
248 that EDTA is not suitable for interchangeable use with the laboratory method when hyperglycaemia is
249 present. Proportional bias has occurred in previous studies of glucometer performance in veterinary
250 species (Cohen et al., 2009; Tennent-Brown et al., 2011), indicating that assessment of glucometer
251 accuracy over a wide glucose concentration range is necessary. In the current study, there was constant
252 bias with underestimation of glucose concentrations by the glucometer for the EDTA, FO and FO3hr
253 groups. For FO and EDTA, this may, in part, reflect higher glucose concentrations in plasma, which are
254 approximately 10-15% higher than those in blood (Gerber and Freeman, 2016), emphasising that
255 results are often not interchangeable. The mean bias for FO-LAB (-1.17 mmol/L) is similar to an
256 estimated difference of 10-15% between plasma (LAB) and blood (FO). Constant bias with glucometer
257 use in horses and other species has been demonstrated previously (Beemer et al., 2013; Cohen et al.,
258 2009; Russell et al., 2007; Tennent-Brown et al., 2011), although the direction of the bias varied
259 between studies. Given the small size of the hyperglycaemic group in the current study, further
260 investigation of the effects of hyperglycaemia on glucometer performance using larger numbers of
261 hyperglycaemic and systemically-ill horses is warranted.

262

263 With the exception of FO-FO3hr, there was, at best, only moderate agreement between
264 glucometer groups, demonstrating that different sample types should not be used for monitoring of
265 glucose concentrations in an individual animal. The results of the current and previous studies (Hollis
266 et al., 2008b; Russell et al., 2007) suggest that human-specific glucometers may be inaccurate when
267 used in horses (due to systematic bias), and careful consideration of appropriate sample type is
268 necessary for specific glucometers. The poor agreement between WB and other groups, can be
269 anticipated from the large RC for WB, as repeatability limits the amount of achievable agreement (Bland
270 and Altman, 1986).

271

272 No glucometer group had 95% of the measurements within 5-20% of the LAB results, with the
273 exception of the EDTA group for hyperglycaemic samples. In human medicine, recommended
274 glucometer performance criteria include 95-100% of measurements within 5-20% of the laboratory
275 method for acceptable accuracy (Tonyushkina and Nichols, 2009). While similar standards for
276 glucometer accuracy in horses have not been established, our results, overall, reflected glucometer
277 inaccuracy, irrespective of sample type. Similarly, glucometer performance often failed to achieve
278 adequate accuracy in alpacas (Beemer et al., 2013) and dogs (Cohn et al., 2000). Glucometer
279 inaccuracy may result in erroneous clinical decisions if glucose reference ranges for a reference
280 laboratory method are used. However, systematic bias may not preclude individual glucometer use if a
281 conversion factor is applied. Alternatively, for sample types with acceptable repeatability (e.g. EDTA,
282 FO, FO3hr), establishing glucometer-specific reference ranges for each sample type would maximise
283 the application of the glucometer and avoid errors in clinical-decision making.

284

285 The accuracy of glucometry using FO3hr was lower than for the FO group. In blood, glucose
286 continues to be utilised by erythrocytes at a rate of approximately 5-7% per h (Chan et al., 1989), and
287 while FO prevents glycolysis through inhibition of enolase, enzymes 'upstream' of enolase are
288 unaffected and continue to metabolise glucose-6-phosphate (Shi et al., 2009). Consequently, glucose
289 concentrations in FO blood samples decrease over the first h after collection and immediate separation
290 of serum or plasma is recommended to preserve glucose concentrations (Chan et al., 1989). In humans,
291 glucose concentrations in FO blood decreased, on average, by 0.39 mmol/L during 4 h transit to the
292 laboratory (Shi et al., 2009). Glucose utilisation after blood collection may have, in part, reduced the
293 repeatability and agreement for the FO3hr group in comparison to the FO group.

294

295 A limitation of this study was the lack of hypoglycaemic samples. A hypoglycaemic group would
296 have allowed determination of glucometer performance across a larger glycaemic range. However, the
297 tendency of the glucometer to under-estimate glucose concentrations is less likely to result in erroneous
298 treatment decisions in hypoglycaemic horses. Further, the effect of haematocrit on glucometer accuracy
299 was not assessed. The haematocrit can influence glucometer accuracy in humans, alpacas and dogs,
300 with decreased and increased haematocrit levels resulting in a positive and negative bias, respectively

301 (Paul et al., 2011; Tang et al., 2000; Tennent-Brown et al., 2011). In a previous equine study (Hollis et
302 al., 2008b), there was no influence of haematocrit on glucometer accuracy. While haematocrit was not
303 assessed in our study, an influence of erythrocytes on glucometer performance due to interactions with
304 the test strip filters cannot be discounted. In addition, other sources of analytic error from interference
305 (e.g. lipaemia, bilirubin, pH, drugs) (Gerber and Freeman, 2016) were not recorded. To minimise
306 methodological influences on glucometer performance (Beemer et al., 2013; Hollis et al., 2008b),
307 measurements were made by a single operator using one glucometer, all test strips had the same lot
308 number and were used within the expiry date and the glucometer and test strips were used according
309 to the manufacturer's instructions, including calibration procedure, storage and operation temperature
310 and humidity.

311

312 **Conclusion**

313 The precision and accuracy of the glucometer was influenced by sample type. Whole blood is
314 unsuitable for glucometry using this equipment as inaccuracy has potential for erroneous clinical
315 decisions. Glucometry using blood collected into FO or EDTA tubes results in precision and accuracy
316 sufficient for clinical use, provided trends in under-estimation of blood glucose concentration are
317 accounted for by determination of method-specific reference ranges. Given the lack of readily available
318 validated equine-specific glucometers, these findings emphasise the importance of assessing individual
319 glucometer performance and optimal sample type prior to use in horses.

320

321 **Conflict of interest statement**

322 None of the authors have financial or personal relationships that could inappropriately influence
323 or bias the content of this paper.

324

325 **Acknowledgements**

326 The authors are grateful for the assistance provided for this study by the staff and students at
327 the Veterinary Clinical Centre and Veterinary Diagnostic Laboratory, Charles Sturt University. The
328 glucometer test strips were provided by Roche Diagnostics. This research did not receive any specific
329 grant from funding agencies in the public, commercial, or not-for-profit sectors.

330

331 **Appendix A: Supplementary material**

332 Supplementary data associated with this article can be found, in the online version, at
333 <https://doi.org/...>

334

335

336

337

338

339

340

341

342

343

344

345

346

347

348

349

350

351

352

353

354

355

356

357

358

359

360

361 **References**

- 362 Beemer, O., Byers, S., Bohn, A., 2013. Evaluation of four point-of-care glucose meters in alpacas.
363 *Journal of Veterinary Internal Medicine* 27, 990-995.
- 364 Bland, J.M., Altman, D.G., 1986. Statistical methods for assessing agreement between two methods of
365 clinical measurement. *Lancet* 1, 307-310.
- 366 Bochicchio, G.V., Sung, J., Joshi, M., Bochicchio, K., Johnson, S.B., Meyer, W., Scalea, T.M., 2005.
367 Persistent hyperglycemia is predictive of outcome in critically ill trauma patients. *Journal of*
368 *Trauma-Injury Infection and Critical Care* 58, 921-924.
- 369 Carr, S.R., Slocum, J., Tefft, L., Haydon, B., Carpenter, M., 1995. Precision of office-based blood
370 glucose meters in screening for gestational diabetes. *American Journal of Obstetrics and*
371 *Gynecology* 173, 1267-1272.
- 372 Chan, A.Y., Swaminathan, R., Cockram, C.S., 1989. Effectiveness of sodium fluoride as a preservative
373 of glucose in blood. *Clinical Chemistry* 35, 315-317.
- 374 Chen, E.T., Nichols, J.H., Duh, S.H., Hortin, G., 2003. Performance evaluation of blood glucose
375 monitoring devices. *Diabetes Technology and Therapeutics* 5, 749-768.
- 376 Cohen, T.A., Nelson, R.W., Kass, P.H., Christopher, M.M., Feldman, E.C., 2009. Evaluation of six
377 portable blood glucose meters for measuring blood glucose concentration in dogs. *Journal of*
378 *the American Veterinary Medical Association* 235, 276-280.
- 379 Cohn, L.A., McCaw, D.L., Tate, D.J., Johnson, J.C., 2000. Assessment of five portable blood glucose
380 meters, a point-of-care analyzer, and color test strips for measuring blood glucose
381 concentration in dogs. *Journal of the American Veterinary Medical Association* 216, 198-202.
- 382 Coldman, M.F., Good, W., 1967. Distribution of Sodium Potassium and Glucose in Blood of Some
383 Mammals. *Comparative Biochemistry and Physiology* 21, 201-206.
- 384 Denfeld, Q.E., Goodell, T.T., Stafford, K.N., Kazmierczak, S., 2011. Precision and Accuracy
385 Comparison of Point-of-Care and Laboratory Glucose Concentrations in Cardiothoracic
386 Surgery Patients. *Journal of Cardiovascular Nursing* 26, 512-518.
- 387 Domori, A., Sunahara, A., Tateno, M., Shimokawa Miyama, T., Setoguchi, A., Endo, Y., 2014. The
388 clinical utility of two human portable blood glucose meters in canine and feline practice.
389 *Veterinary Clinical Pathology* 43, 55-62.
- 390 Gerber, K.L., Freeman, K.P., 2016. ASVCP guidelines: quality assurance for portable blood glucose
391 meter (glucometer) use in veterinary medicine. *Veterinary Clinical Pathology* 45, 10-27.
- 392 Hackett, E.S., McCue, P.M., 2010. Evaluation of a veterinary glucometer for use in horses. *Journal of*
393 *Veterinary Internal Medicine* 24, 617-621.
- 394 Hassel, D.M., Hill, A.E., Rorabeck, R.A., 2009. Association between Hyperglycemia and Survival in 228
395 Horses with Acute Gastrointestinal Disease. *Journal of Veterinary Internal Medicine* 23, 1261-
396 1265.
- 397 Hoedemaekers, C.W.E., Gunnewiek, J.M.T.K., Prinsen, M.A., Willems, J.L., Hoeven, J.G.D., 2008.
398 Accuracy of bedside glucose measurement from three glucometers in critically ill patients.
399 *Critical Care Medicine* 36, 3062-3066.
- 400 Hollis, A.R., Boston, R.C., Corley, K.T.T., 2007. Blood glucose in horses with acute abdominal disease.
401 *Journal of Veterinary Internal Medicine* 21, 1099-1103.

- 402 Hollis, A.R., Furr, M.O., Magdesian, K.G., Axon, J.E., Ludlow, V., Boston, R.C., Corley, K.T.T., 2008a.
403 Blood glucose concentrations in critically ill neonatal foals. *Journal of Veterinary Internal*
404 *Medicine* 22, 1223-1227.
- 405 Hollis, A.R., Schaer, B.L.D., Boston, R.C., Wilkins, P.A., 2008b. Comparison of the accu-chek aviva
406 point-of-care glucometer with blood gas and laboratory methods of analysis of glucose
407 measurement in equine emergency patients. *Journal of Veterinary Internal Medicine* 22, 1189-
408 1195.
- 409 Hug, S.A., Riond, B., Schwarzwald, C.C., 2013. Evaluation of a continuous glucose monitoring system
410 compared with an in-house standard laboratory assay and a handheld point-of-care glucometer
411 in critically ill neonatal foals. *Journal of Veterinary Emergency and Critical Care* 23, 408-415.
- 412 Kanji, S., Buffie, J., Hutton, B., Bunting, P.S., Singh, A., McDonald, K., Fergusson, D., McIntyre, L.A.,
413 Hebert, P.C., 2005. Reliability of point-of-care testing for glucose measurement in critically ill
414 adults. *Critical Care Medicine* 33, 2778-2785.
- 415 Khan, A.I., Vasquez, Y., Gray, J., Wians, F.H., Kroll, M.H., 2006. The variability of results between point-
416 of-care testing glucose meters and the central laboratory analyzer. *Archives of Pathology and*
417 *Laboratory Medicine* 130, 1527-1532.
- 418 Krouwer, J.S., Cembrowski, G.S., 2010. A review of standards and statistics used to describe blood
419 glucose monitor performance. *Journal of Diabetes Science and Technology* 4, 75-83.
- 420 Lewis, K.S., Kane-Gill, S.L., Bobek, M.B., Dasta, J.F., 2004. Intensive insulin therapy for critically ill
421 patients. *Annals of Pharmacotherapy* 38, 1243-1251.
- 422 Lin, L.I., 1989. A Concordance Correlation-Coefficient to Evaluate Reproducibility. *Biometrics* 45, 255-
423 268.
- 424 Paul, A.E.H., Shiel, R.E., Juvet, F., Mooney, C.T., Mansfield, C.S., 2011. Effect of hematocrit on
425 accuracy of two point-of-care glucometers for use in dogs. *American Journal of Veterinary*
426 *Research* 72, 1204-1208.
- 427 Ray, J.G., Hamielec, C., Mastracci, T., 2001. Pilot study of the accuracy of bedside glucometry in the
428 intensive care unit. *Critical Care Medicine* 29, 2205-2207.
- 429 Russell, C., Palmer, J.E., Boston, R.C., Wilkins, P.A., 2007. Agreement between point-of-care
430 glucometry, blood gas and laboratory-based measurement of glucose in an equine neonatal
431 intensive care unit. *Journal of Veterinary Emergency and Critical Care* 17, 236-242.
- 432 Shi, R.Z., Seeley, E.S., Bowen, R., Faix, J.D., 2009. Rapid blood separation is superior to fluoride for
433 preventing in vitro reductions in measured blood glucose concentration. *Journal of Clinical*
434 *Pathology* 62, 752-753.
- 435 Sung, J., Bochicchio, G.V., Joshi, M., Bochicchio, K., Tracy, K., Scalea, T.M., 2005. Admission
436 hyperglycemia is predictive of outcome in critically ill trauma patients. *Journal of Trauma-Injury*
437 *Infection and Critical Care* 59, 80-83.
- 438 Tack, C., Pohlmeier, H., Behnke, T., Schmid, V., Grenningloh, M., Forst, T., Pflutzner, A., 2012.
439 Accuracy Evaluation of Five Blood Glucose Monitoring Systems Obtained from the Pharmacy:
440 A European Multicenter Study with 453 Subjects. *Diabetes Technology and Therapeutics* 14,
441 330-337.
- 442 Tang, Z.P., Lee, J.H., Louie, R.F., Kost, G.J., 2000. Effects of different hematocrit levels on glucose
443 measurements with handheld meters for point-of-care testing. *Archives of Pathology and*
444 *Laboratory Medicine* 124, 1135-1140.

445 Tennent-Brown, B.S., Koenig, A., Williamson, L.H., Boston, R.C., 2011. Comparison of three point-of-
446 care blood glucose meters for use in adult and juvenile alpacas. *Journal of the American*
447 *Veterinary Medical Association* 239, 380-386.

448 Tonyushkina, K., Nichols, J.H., 2009. Glucose meters: a review of technical challenges to obtaining
449 accurate results. *Journal of Diabetes Science and Technology* 3, 971-980.

450 Vogelzang, M., Nijboer, J.M., van der Horst, I.C., Zijlstra, F., ten Duis, H.J., Nijsten, M.W., 2006.
451 Hyperglycemia has a stronger relation with outcome in trauma patients than in other critically ill
452 patients. *Journal of Trauma-Injury Infection and Critical Care* 60, 873-879.

453

454

455

456

457

458

459

460

461

462

463

464

465

466

467

468

469

470

471

472

473

474

475

476 **Table 1:** Repeatability coefficient (RC), median [interquartile range] coefficient of variation (CV) and
 477 Spearman's correlation coefficient (r_s) for paired measurements of glucose concentration for each group
 478 and for each category of glucose concentration. (EDTA = ethylenediaminetetraacetic acid, FO = fluoride
 479 oxalate, FO3hr = fluoride oxalate and 3 h of storage, LAB = fluoride oxalate and wet chemistry analyser
 480 at laboratory, WB = whole blood).

481

482

| Glucose concentration category | Group | RC (mmol/L) | CV (%) | Spearman's correlation | |
|--------------------------------|-------|-------------|----------------|------------------------|--------|
| | | | | r_s | P |
| All samples (n=62) | WB | 2.36 | 8.1 [3.3-14.6] | 0.79 | <0.001 |
| | EDTA | 0.86 | 2.9 [1.5-5.3] | 0.85 | <0.001 |
| | FO | 0.61 | 2.2 [1.6-4.1] | 0.92 | <0.001 |
| | FO3hr | 0.82 | 3.5 [1.9-6.3] | 0.92 | <0.001 |
| | LAB | 0.34 | 1.4 [1.1-2.5] | 0.95 | <0.001 |
| Euglycaemic samples (n=49) | WB | 2.14 | 9.4 [3.6-16.5] | 0.60 | <0.001 |
| | EDTA | 0.74 | 3.1 [1.6-5.9] | 0.71 | <0.001 |
| | FO | 0.50 | 3.3 [1.7-4.1] | 0.85 | <0.001 |
| | FO3hr | 0.78 | 3.5 [1.9-7.3] | 0.62 | <0.001 |
| | LAB | 0.33 | 1.5 [1.3-2.8] | 0.90 | <0.001 |
| Hyperglycaemic samples (n=13) | WB | 2.94 | 5.5 [2.5-9.0] | 0.83 | <0.001 |
| | EDTA | 1.24 | 1.9 [0.9-2.5] | 0.93 | <0.001 |
| | FO | 0.93 | 2.0 [1.0-3.0] | 0.98 | <0.001 |
| | FO3hr | 0.94 | 2.1 [1.3-3.9] | 0.99 | <0.001 |
| | LAB | 0.39 | 0.7 [0.4-1.3] | 1.00 | <0.001 |

483

484

485

486 **Table 2:** Lin's concordance (ρ) and Spearman's correlation coefficient (r_s) analysis of paired glucose
 487 measurements between groups for each glucose concentration category. (EDTA =
 488 ethylenediaminetetraacetic acid, FO = fluoride oxalate, FO3hr = fluoride oxalate and 3 h of storage,
 489 LAB = fluoride oxalate and wet chemistry analyser at laboratory, WB = whole blood).

| Group comparison | Glucose concentration category | | | | | | | | | | | |
|---------------------|--------------------------------|-----------|-------|--------|-------------------------------|-----------|-------|--------|----------------------------------|-----------|-------|--------|
| | All samples (n=62) | | | | Euglycaemic samples (n=49) | | | | Hyperglycaemic samples (n=13) | | | |
| | ρ | 95% CI | r_s | P | ρ | 95% CI | r_s | P | ρ | 95% CI | r_s | P |
| WB-LAB | 0.94 | 0.90-0.96 | 0.68 | <0.001 | 0.31 | 0.13-0.46 | 0.37 | 0.01 | 0.89 | 0.68-0.97 | 0.87 | <0.001 |
| WB-EDTA | 0.93 | 0.89-0.96 | 0.60 | <0.001 | 0.18 | 0.03-0.33 | 0.20 | 0.17 | 0.94 | 0.83-0.98 | 0.94 | <0.001 |
| WB-FO | 0.86 | 0.79-0.91 | 0.66 | <0.001 | 0.11 | 0.01-0.22 | 0.33 | 0.01 | 0.77 | 0.48-0.91 | 0.81 | <0.001 |
| WB-FO3hr | 0.88 | 0.81-0.92 | 0.68 | <0.001 | 0.19 | 0.07-0.31 | 0.37 | 0.00 | 0.76 | 0.45-0.91 | 0.81 | <0.001 |
| EDTA-LAB | 0.98 | 0.96-0.99 | 0.83 | <0.001 | 0.49 | 0.33-0.63 | 0.66 | <0.001 | 0.93 | 0.81-0.98 | 0.93 | <0.001 |
| EDTA-FO | 0.94 | 0.92-0.96 | 0.79 | <0.001 | 0.38 | 0.22-0.52 | 0.57 | <0.001 | 0.79 | 0.55-0.91 | 0.91 | <0.001 |
| EDTA-FO3hr | 0.95 | 0.93-0.97 | 0.73 | <0.001 | 0.48 | 0.29-0.64 | 0.45 | 0.00 | 0.79 | 0.54-0.91 | 0.81 | <0.001 |
| FO-LAB | 0.93 | 0.90-0.95 | 0.84 | <0.001 | 0.23 | 0.14-0.32 | 0.67 | <0.001 | 0.84 | 0.67-0.93 | 0.91 | <0.001 |
| FO-FO3hr | 0.99 | 0.98-0.99 | 0.85 | <0.001 | 0.66 | 0.48-0.78 | 0.69 | <0.001 | 0.98 | 0.95-0.99 | 0.98 | <0.001 |
| FO3hr-LAB | 0.94 | 0.92-0.96 | 0.76 | <0.001 | 0.28 | 0.16-0.39 | 0.51 | <0.001 | 0.86 | 0.69-0.94 | 0.91 | <0.001 |

490

491

492

493 **Table 3:** Summary of the Bland Altman analyses of agreement for glucose measurement between
 494 groups for each category of glucose concentration. (EDTA = ethylenediaminetetraacetic acid, FO =
 495 fluoride oxalate, FO3hr = fluoride oxalate and 3 h of storage, LAB = fluoride oxalate and wet chemistry
 496 analyser at laboratory, WB = whole blood).

| Group comparison | Glucose concentration category | | | | | | | | |
|------------------|--------------------------------|------------------|-------|----------------------------|------------------|-------|-------------------------------|------------------|-------|
| | All samples (n=62) | | | Euglycaemic samples (n=49) | | | Hyperglycaemic samples (n=13) | | |
| | Mean bias | 95% LOA (mmol/L) | | Mean bias | 95% LOA (mmol/L) | | Mean bias | 95% LOA (mmol/L) | |
| | (mmol/L) | | | (mmol/L) | | | (mmol/L) | | |
| WB-LAB | 0.07 | 2.44 | -2.31 | 0.11 | 2.44 | -2.21 | -0.12 | 2.53 | -2.76 |
| WB-EDTA | 0.44 | 2.91 | -2.04 | 0.62 | 3.08 | -1.84 | -0.26 | 1.83 | -2.35 |
| WB-FO | 1.24 | 3.79 | -1.30 | 1.18 | 3.67 | -1.31 | 1.47 | 4.27 | -1.33 |
| WB-FO3hr | 1.04 | 3.56 | -1.48 | 0.98 | 3.33 | -1.37 | 1.26 | 4.40 | -1.88 |
| EDTA-LAB | -0.37 | 0.94 | -1.68 | -0.51 | 0.25 | -1.26 | 0.15 | 2.38 | -2.09 |
| EDTA-FO | 0.80 | 2.42 | -0.81 | 0.56 | 1.40 | -0.29 | 1.73 | 4.16 | -0.69 |
| EDTA-FO3hr | 0.60 | 2.32 | -1.11 | 0.36 | 1.24 | -0.52 | 1.52 | 4.25 | -1.20 |
| FO-LAB | -1.17 | -0.27 | -2.07 | -1.07 | -0.36 | -1.77 | -1.58 | -0.47 | -2.70 |
| FO-FO3hr | -0.20 | 0.57 | -0.97 | -0.20 | 0.54 | -0.93 | -0.21 | 0.70 | -1.12 |
| FO3hr-LAB | -0.97 | 0.05 | -2.00 | -0.87 | -0.06 | -1.67 | -1.38 | 0.00 | -2.75 |

497

498

499

500

501

502

503 **Table 4:** Percentage of glucose measurements determined for each blood glucose category and each
504 glucometer group that were within 5, 10, 15 or 20% of the glucose measurement determined by the
505 laboratory reference method. (EDTA = ethylenediaminetetraacetic acid, FO = fluoride oxalate, FO3hr =
506 fluoride oxalate and 3 h of storage, LAB = fluoride oxalate and wet chemistry analyser at laboratory,
507 WB = whole blood).
508
509

| Group | Category | Within 5% | Within 10% | Within 15% | Within 20% |
|--------------|-------------------------------|------------------|-------------------|-------------------|-------------------|
| WB | All samples (n=62) | 24.2 | 41.9 | 56.5 | 67.7 |
| | Euglycaemic samples (n=49) | 20.4 | 36.7 | 51.0 | 61.2 |
| | Hyperglycaemic samples (n=13) | 38.5 | 61.5 | 76.9 | 92.3 |
| EDTA | All samples (n=62) | 22.6 | 56.7 | 83.3 | 93.3 |
| | Euglycaemic samples (n=49) | 14.3 | 51.0 | 81.6 | 91.8 |
| | Hyperglycaemic samples (n=13) | 53.8 | 76.9 | 84.6 | 100 |
| FO | All samples (n=62) | 1.6 | 8.3 | 26.7 | 56.7 |
| | Euglycaemic samples (n=49) | 2.0 | 4.1 | 14.3 | 44.9 |
| | Hyperglycaemic samples (n=13) | 0.0 | 23.1 | 69.2 | 92.3 |
| FO3hr | All samples (n=62) | 8.1 | 23.3 | 40.0 | 70.0 |
| | Euglycaemic samples (n=49) | 6.1 | 16.3 | 32.7 | 65.3 |
| | Hyperglycaemic samples (n=13) | 15.4 | 53.8 | 69.2 | 84.6 |

510
511