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■ Glucose measurement using portable blood glucose meters in dogs and cats

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■ Η χρήση φορητής συσκευής για τη μέτρηση της συγκέντρωσης της γλυκόζης στο αίμα στο σκύλο και τη γάτα

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ABSTRACT. Portable blood glucose meters (PBGMs) are small electronic devices that measure the concentration of glucose in whole blood. Due to the technological advances, measurement of glucose concentration is carried out in a small blood volume, and it is a relatively simple, quick and inexpensive procedure. PBGMs are frequently used in companion animal medicine, especially for the diagnosis and treatment monitoring of dogs and cats with diabetes mellitus and hypoglycaemia. The main factors affecting the precision of the measurement include: a) the device (manufacturer), the consumables (reagent strips) and their storage conditions; b) environmental conditions (temperature and possibly altitude); c) blood collection technique (site of sampling, cleanliness at the site of sampling, use of anticoagulants); d) patient factors (haematocrit, blood triglycerides, creatinine, uric acid and protein concentrations, drug administration); and, e) operator errors. Due to all these factors, readings of glucose concentration by PBGMs may differ from those of chemistry analysers, and this should be taken into account when shifting from one method to the other. Furthermore, because the results depend on the PBGM, the same device should be used and all measurements should be made under the same environmental conditions and using the same blood sampling technique for serial measurements of blood glucose to be comparable. Finally, all the above mentioned limitations of glucose measurement by PBGMs should be taken into consideration and the results should be interpreted along with the clinical signs and any other laboratory findings for optimal diagnostic and therapeutic decisions.

Keywords: cat, dog, portable blood glucose meters

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ΠΕΡΙΛΗΨΗ. Οι φορητές συσκευές για τη μέτρηση της συγκέντρωσης της γλυκόζης στο αίμα (γλυκοζόμετρα) είναι ηλεκτρονικές συσκευές που προσδιορίζουν τη συγκέντρωση της γλυκόζης στο ολικό αίμα. Χάρη στην εξέλιξη της τεχνολογίας, ο προσδιορισμός αυτός πραγματοποιείται σε μικρό όγκο αίματος και αποτελεί απλή γρήγορη και χαμηλού κόστους διαδικασία. Τα γλυκοζόμετρα χρησιμοποιούνται συχνά στην ιατρική των ζώων συντροφιάς, ιδιαίτερα για τη διάγνωση και την παρακολούθηση της θεραπείας σκύλων και γατών με σακχαρώδη διαβήτη ή υπογλυκαιμία. Τα αποτελέσματα της μέτρησης της συγκέντρωσης της γλυκόζης με τα γλυκοζόμετρα επηρεάζονται από διάφορους παράγοντες, όπως: α) η ίδια η συσκευή, αφού το αποτέλεσμα ενδέχεται να διαφέρει μεταξύ γλυκοζόμετρων από διαφορετικούς κατασκευαστές, καθώς και τα αναλώσιμα (ταινίες) που τη συνοδεύουν και οι συνθήκες αποθήκευσής τους, β) οι περιβαλλοντικές συνθήκες, όπως είναι η θερμοκρασία και ενδεχομένως το υψόμετρο, γ) η τεχνική συλλογής του δείγματος (σημείο αιμοληψίας, καθαριότητα του σημείου αιμοληψίας, συλλογή του δείγματος σε φιαλίδιο με αντιπηκτικό), δ) το ίδιο το ζώο, αφού το αποτέλεσμα επηρεάζεται από τον αιματοκρίτη, τη συγκέντρωση των τριγλυκεριδίων, της κρεατινίνης, του ουρικού οξέος και των πρωτεϊνών στο αίμα και τη χορήγηση ορισμένων φαρμάκων, και ε) τα σφάλματα ανάλυσης. Λόγω της επίδρασης των παραγόντων αυτών, το αποτέλεσμα της μέτρησης ενδέχεται να διαφέρει σημαντικά όταν αυτή προσδιορίζεται με γλυκοζόμετρο σε σύγκριση με τους βιοχημικούς αναλυτές, γεγονός που πρέπει να λαμβάνεται υπόψη, ιδιαίτερα όταν δε χρησιμοποιείται η ίδια μέθοδος προσδιορισμού σε διαδοχικές μετρήσεις στο ίδιο ζώο. Επιπλέον, λόγω των διαφορών του αποτελέσματος μεταξύ συσκευών από διαφορετικούς κατασκευαστές, πρέπει να χρησιμοποιείται πάντα το ίδιο γλυκοζόμετρο και οι μετρήσεις να γίνονται κάτω από τις ίδιες περιβαλλοντικές συνθήκες και με την ίδια τεχνική αιμοληψίας προκειμένου να είναι συγκρίσιμα τα αποτελέσματα. Οι παραπάνω παράγοντες που επηρεάζουν τη συγκέντρωση της γλυκόζης στο αίμα όταν αυτή προσδιορίζεται με τη βοήθεια του γλυκοζόμετρου πρέπει να λαμβάνονται υπόψη και, όπως άλλωστε ισχύει για κάθε εργαστηριακή εξέταση, το αποτέλεσμα πρέπει να συνεκτιμάται με τα κλινικά και τα υπόλοιπα εργαστηριακά ευρήματα για τη λήψη των καλύτερων διαγνωστικών και θεραπευτικών αποφάσεων.

Λέξεις ευρητηρίας: γάτα, γλυκοζόμετρο, σκύλος, φορητή συσκευή μέτρηση της γλυκόζης

INTRODUCTION

Glucose, the principal source of energy for mammalian cells, is derived from the diet, hepatic glycconeogenesis and hepatic and muscle glucogenolysis, and its concentration in blood is under the control of various hormones, including insulin, glucagon, cortisol, catecholamines and growth hormone (Feldman and Nelson, 2004; Knieriem et al., 2007). Glucose metabolism and blood concentrations are abnormal in many disease states (Table 1). For this reason, hyperglycaemia and hypoglycaemia are common laboratory findings in dogs and cats. Measurement of blood glucose concentration is essential whenever the clinical picture is compatible with hyperglycaemia or hypoglycaemia (Table 2), whenever any of the possible underlying causes is included in the differentials list (Table 1), in all critical care patients, and for general health screening purposes (Villiers and Blackwood, 2005; Wiedmeyer and DeClue, 2008; Ford and Lynch,

2013; Surman and Fleeman, 2013). Furthermore, repeated measurements of blood glucose concentration are necessary to monitor and adjust the treatment of all the diseases that cause hyperglycaemia and hypoglycaemia and they are needed on a long-term to lifelong basis in dogs and cats with diabetes mellitus and insulinoma (Feldman and Nelson, 2004; Dietiker-Moretti et al., 2011).

The methodology of glucose analysis is currently based on either chromogenic or electrochemical reactions that are mediated by an enzyme, such as glucose oxidase, glucose dehydrogenase and hexokinase (Kaneko et al., 2008; Johnson et al., 2009; Surman and Fleeman, 2013). These enzymatic methods have been applied in different equipment and devices, including in-house and reference laboratory automated chemistry analysers, point-of-care analysers, colorimetric reagent strips, portable blood glucose meters (PBMGs), and continuous glucose monitoring

Table 1. The most common causes of hyperglycaemia and hypoglycaemia in dogs and cats.

Causes of hyperglycaemia
Diabetes mellitus (non-complicated, ketoacidotic, hyperosmolar)
Acute pancreatitis
Glucagonoma
Hyperadrenocorticism
Pheochromocytoma in dogs
Acromegaly (especially in cats)
Hyperthyroidism in cats
Dioestrus in dogs
Sepsis
Neoplasia
Brain trauma
Stress (especially in cats)
Recent meal (especially if rich in monosaccharides, disaccharides or propylene glycol)
Medications: glucocorticoids, progestagens, α_2 -adrenergic agonists, thiazide diuretics, dextrose containing fluids, total parenteral nutrition
Causes of hypoglycaemia
Insulinoma
Hypoadrenocorticism
Pituitary dwarfism
Sepsis, canine babesiosis
Neoplasia (except insulinoma)
Liver diseases: glycogen storage diseases, acute liver insufficiency, portosystemic shunts, cirrhosis
Neonatal and juvenile hypoglycaemia
Hunting dog hypoglycaemia
Prolonged fasting of animals in poor body condition
Renal failure (most commonly acute renal failure)
Toxicoses: ethylene glycol, alcohol, xylitol
Medications: insulin, orally administered hypoglycaemic agents
Laboratory error (delay in serum separation), severe polycythaemia

Table 2. Clinical signs of hyperglycaemia and hypoglycaemia in dogs and cats, irrespectively of the underlying cause.

Clinical signs of hyperglycaemia	
Polyuria-polydipsia	
Clinical signs of hypoglycaemia	
Due to neuroglycosopenia	Due to sympathetic stimulation
Reduced level of consciousness, disorientation	Nervousness, restlessness
Seizures	Mydriasis
Blindness	Muscle tremors
Ataxia, paresis	Polyphagia, increased body weight
Muscle weakness	

systems (CGMSs) (Wiedmeyer and DeClue, 2008; Surman and Fleeman, 2013).

Automated chemistry analysers are used by most commercial diagnostic laboratories as in-house equipment and they determine blood glucose concentrations in plasma or serum samples employing colorimetric endpoint or kinetic assays. The former are inaccurate with lipemic samples, since turbidity decreases light transmittance, which is particularly important for dogs and cats with diseases that simultaneously affect glucose and lipid metabolism, like diabetes mellitus. The latter detect changes in the optical density of the specimen over time. Other widely used methods for determination of blood glucose concentrations involve enzyme-catalysed reactions coupled to a colour detection system, oxidation-reduction methods, and methods that result in the generation of an electrical current (Hagvik, 2007; Johnson et al., 2009). Although automated chemistry analysers are usually very accurate, their major disadvantages when used for the determination of blood glucose concentrations are the relatively large volume of blood and the long time to obtain the results. For these reasons, they are usually preferred when additional biochemical parameters, besides glucose, should be measured on a non-emergency setting (Surman and Fleeman, 2013).

Point-of-care analysers usually operate with dry chemistry slides or strips, require only small amounts of whole blood, serum or plasma, and provide rapid results. Besides glucose, they can also measure, using different slides or strips, other analytes that are helpful for the evaluation of the acid-base and the electrolyte status of the patient. For these reasons they are especially valuable in emergency cases. However, point-of-care analysers are relatively expensive and can only be used by trained personnel (Cohn et al., 2000).

Colorimetric reagent strips, which are read visually or with a reflectance meter, are available for estimating glucose concentration in fresh whole blood (Sherwood et al., 1983). Currently, they are rarely, if ever, used because they are the least precise of all the available methods.

Portable blood glucose meters were originally developed to permit human diabetic patients to self-monitor blood glucose concentration using a drop of capillary blood (Ginsberg, 2009a). Following an enzymatic reaction (glucose oxidase, glucose dehydrogenase, glucose oxidase-peroxidase) that is coupled to a chromogen or to an electron-donating mediator system, blood glucose concentration is measured via reflectance photometry (photometric methods) or via an electrode (electrochemical methods), respectively (Cohn et al., 2000; Johnson et al., 2009; Surman and Fleeman, 2013; Ford and Lynch, 2013). PBGMs are commonly used to measure blood glucose concentrations in dogs and cats. They require a small volume of venous or capillary blood, they have a relatively wide working range (from 10-20 to 500-600 mg dL⁻¹, depending on the device), they provide immediate results, they are of low cost and they are easy to handle, which permits their use even by animal owners after a short period of training (Cohen et al., 2009; Johnson et al., 2009; Dobromylskij and Sparkes, 2010). These attributes make PBGMs the ideal way to measure blood glucose concentrations in emergency patients with hyperglycaemia or hypoglycaemia, in all critical care patients and for the long-term monitoring of diabetic dogs and cats in the hospital or at home (Alt et al., 2007; Rios and Ward, 2008; Cohen et al., 2009). The accuracy of PBGMs is highly important because the glucose readings that will be obtained will dictate the course of clinical action (Cohn et al., 2000).

Continuous glucose monitoring systems are also electronic devices that measure glucose concentrations in the interstitial fluid with the aid of a small flexible sensor that is inserted into the subcutaneous tissue. The measurement is based on the glucose oxidase-mediated reaction between glucose and oxygen (Rios and Ward, 2008; Wiedmeyer and DeClue, 2008; Moretti et al., 2010; Dietiker-Moretti et al., 2011; Fleeman, 2011; Surman and Fleeman, 2013). Glucose concentrations in the blood and the interstitial fluid are usually highly correlated, and the calibration of the CGMS using peripheral blood samples with known glucose concentrations per-

mits the device to accurately estimate blood glucose (Rios and Ward, 2008; Wiedmeyer and DeClue, 2008; Moretti et al., 2010; Dietiker-Moretti et al., 2011; Hoenig et al., 2012; Surman and Fleeman, 2013). CGMSs typically record glucose concentrations every 3-5 minutes for up to 72 hours and are mainly used to regulate insulin treatment of stable diabetic dogs and cats (Wiedmeyer and DeClue, 2008; Moretti et al., 2010; Dietiker-Moretti et al., 2011; Fleeman, 2011; Surman and Fleeman, 2013). Additional indications may include monitoring of dogs and cats with diabetic ketoacidosis, of critical care patients, of dogs with juvenile hypoglycaemia as well as the intra- and post-operative monitoring of surgical patients at risk for hypoglycaemia or hyperglycaemia (Wiedmeyer and DeClue, 2008); however, the accuracy of CGMS for these purposes has not been fully evaluated and/or it is debatable (Surman and Fleeman, 2013).

INDICATIONS FOR USE OF PORTABLE BLOOD GLUCOSE METERS IN DOGS AND CATS

Hyperglycaemia or hypoglycaemia in dogs and cats (Table 1) may be an emergency, as in the case of ketoacidotic or hyperosmolar diabetes mellitus or hypoglycaemia-induced seizures (Dobromylskyj and Sparkes, 2010; Koenig, 2013). In such cases, the rapid diagnosis of abnormal blood glucose concentration is essential for the initial therapeutic decisions. In all dogs and cats with clinical signs compatible with a diabetic emergency or with hypoglycaemia (Table 2) measurement of blood glucose concentration with a PBGM is strongly advised as one of the initial diagnostic steps. If the blood glucose readings of the PBGM are above or below the reference range, the result can be confirmed using an automated chemistry analyser, since a complete serum biochemical profile will be anyway necessary to identify the full range of biochemical abnormalities that accompany diabetic emergencies or to investigate the cause of hypoglycaemia (Koenig, 2013).

In addition, PBGMs are indispensable for monitoring of the emergency treatment of hyperglycaemia (e.g. regular insulin administration) or hypoglycaemia (e.g. dextrose infusion) which is based on serial measurements of blood glucose concentrations (Feldman and Nelson, 2004; Koenig, 2013).

Non-diabetic critical care dogs and cats, with or without one of the known causes of hyperglycaemia (Table 1), may present disturbances of glucose metabolism leading to increased blood glucose concentrations (Knieriem et al., 2007). Possible underlying mechanisms include increased adrenal secretion of glucocorticoids and catecholamines and insulin resistance, in addition to the possible hyperglycaemic effects of various therapeutic interventions (Knieriem et al., 2007). Increased blood glucose concentrations have been correlated with the severity of head trauma in dogs and cats (Syring et al., 2001) and with a poor outcome in dogs with congestive heart failure (Brady et al., 2004) and perhaps in dogs with sepsis (Hardie et al., 1985). Intensive insulin therapy has been shown to reduce the incidence of complications and to improve prognosis in human critical care patients with hyperglycaemia (Van den Berghe et al., 2003; Krinsley, 2004). Although there is a paucity of similar studies in companion animals, the use of regular insulin has been suggested in hyperglycaemic dogs and cats with head trauma or sepsis in an effort to strictly control blood glucose concentration in a range between 85 and 130 mg dL⁻¹ (Knieriem et al., 2007). Obviously, if this therapeutic intervention is adopted, repeated measurements of blood glucose concentrations with PBGMs is the only practical way to monitor treatment. At least in theory, a possible alternative could be the use of a CGMS with real-time display of interstitial fluid glucose concentration (Wiedmeyer and DeClue, 2008).

Blood glucose curves (BGC) are essential for monitoring and adjusting insulin treatment of dogs and cats with diabetes mellitus (Alt et al., 2007; Martin and Rand, 2007; Rios and Ward, 2008; Affenzeller et al., 2011; Dietiker-Moretti et al., 2011; Smith et al., 2012; Roomp and Rand, 2013). They can be generated in either hospitalized patients or

at home (Alt et al., 2007; Rios and Ward, 2008; Dietiker-Moretti et al., 2011; Smith et al., 2012; Roomp and Rand, 2013; Ford and Lynch, 2013). Most diabetic pets are administered insulin twice daily; thus the BGC typically lasts for 10-12 hours and blood sampling for measurement of glucose concentration is typically performed every 1-2 hours during this period, although sampling every 3-4 hours may be carried out in cats at home (Moretti et al., 2010; Affenzeller et al., 2011; Dietiker-Moretti et al., 2011; Roomp and Rand, 2013). Since an important objective of the BGC is to determine the minimum glucose concentration or nadir (Martin and Rand, 2007; Dietiker-Moretti et al., 2011), more frequent sampling (usually every 30 min) is practiced by the authors during the period of maximum insulin activity (between 2 and 6 hours after insulin administration) and/or when a trend of progressively declining blood glucose concentration is noticed. Therefore, at least 7 and up to 20 or even more blood glucose measurements may be performed during a BGC. Obviously, PBGMs and not automated chemistry analysers are used to measure blood glucose concentrations when BGC is conducted at home by the owner (Alt et al., 2007; Dobromylskyj and Sparkes, 2010; Ford and Lynch, 2013), and PBGMs are clearly preferred in the hospital environment due to the small blood volume that is required (avoidance of blood loss anaemia) and the option to use capillary instead of venous blood (less stressful sampling). On the other hand, current data suggest that CGMSs may be preferable over PBGMs for BGCs, mainly because of their superiority in terms of recording glucose nadir, zenith and daily fluctuations, of the option for continued monitoring of glucose concentrations for up to 72 hours, and of the avoidance of repeated patient restraint for blood sampling (Rios and Ward, 2008; Wiedmeyer and DeClue, 2008; Moretti et al., 2010; Affenzeller et al., 2011; Dietiker-Moretti et al., 2011;). However, the cost to purchase the equipment may not be always affordable, the accuracy of the measurements may be lower compared to PBGMs, especially when blood glucose concentrations are at the hypoglycaemic

range, thus necessitating periodic use of a PBGM and therapeutic decision-making may not significantly differ depending on the use of CGMS or a PBGM (Moretti et al., 2010; Dietiker-Moretti et al., 2011; Fleeman, 2011).

Home monitoring of blood glucose concentration of diabetic dogs and cats decreases the medical cost (although it does not decrease the frequency of re-examinations), is feasible for most owners and it allows their active participation in the medical care of their pet (Roomp and Rand, 2009; Smith et al., 2012). Besides the traditional BGC that can be performed at home, current research suggests that especially in diabetic cats it is possible and safe to implement intense control of hyperglycaemia with a combination of long-acting insulin administration and dietary modifications, provided that the owners are willing and able to measure blood glucose concentration with a PBGM at least 3 times per day and optimally 5 times per day, on an everyday basis for a few weeks up to 4 months (Roomp and Rand, 2009; Roomp and Rand, 2013; Ford and Lynch, 2013). The intense protocol has the advantage that in more than 80% of newly diagnosed cats diabetic remission can be achieved and insulin administration can be discontinued, on a temporary or even permanent basis (Roomp and Rand, 2009; Roomp and Rand, 2013).

Elective (e.g., neutering of female dogs) and non-elective surgery of diabetic dogs and cats poses a challenge in terms of controlling blood glucose concentration to avoid severe hyperglycaemia and hypoglycaemia. Lack of food intake, influence of the anaesthetic medication and the surgical stress on glucose metabolism, and inability to monitor for clinical signs suggestive of abnormal blood glucose concentrations are the main considerations on the day of surgery and until the diabetic animal is awake and resumes eating. The typical recommendation during this period is to monitor blood glucose concentration every 30-60 min and to establish tight control ($60\text{--}250\text{ mg dL}^{-1}$) with the administration of regular insulin and the appropriate fluid (normal saline 0.9%, normal saline 0.45% and dextrose 2.5%, dextrose 5%) (Feldman and Nelson, 2004). Obviously,

PBGMs are indispensable for the measurement of blood glucose concentrations and guiding treatment during this period.

The diagnosis of insulinoma is based on the normal or increased serum insulin concentration in the face of hypoglycaemia ($<50\text{-}60\text{ mg dL}^{-1}$) (Koenig, 2013). In some dogs and cats with this tumour, blood glucose concentration fluctuates between low-normal and subnormal levels thus necessitating supervised fasting and repeated measurements in order to identify a hypoglycaemic serum sample where insulin concentration can be measured (Ford and Lynch, 2013; Koenig, 2013;). PBGMs are advised to monitor blood glucose concentration during the fasting period, although confirmation of hypoglycaemia in an automated chemistry analyser is needed before the serum sample is submitted to the laboratory for measurement of insulin concentration (Cohen et al., 2009)

FACTORS AFFECTING GLUCOSE MEASUREMENT BY PORTABLE BLOOD GLUCOSE METERS

Different devices, employing different analytical methodologies, are characterized, not only by variable method-specific interferences (e.g. the effect of blood oxygen tension on glucose oxidase method), but also by important differences in their inherent random (imprecision) and systematic (bias) error (Jensen and Kjølgaard-Hansen, 2006; Ginsberg, 2009a; Johnson et al., 2009). In human medicine, the need for an internationally accepted reference method, like isotope dilution gas chromatography-mass spectrometry (Hagvik, 2007), to evaluate the accuracy and to calibrate blood glucose measuring devices, including PBGMs, has long been recognized. Until such a goal is achieved and is applicable for dogs and cats, it is recommended to consistently use the same device for the same patient, especially if monitoring of the trends in blood glucose concentration over time is clinically important (Johnson et al., 2009; Paul et al., 2011).

In general terms, the accuracy of blood glucose concentrations that are measured by PBGMs

differ depending on whether the equipment has been designed for human or for veterinary use, and for the former it also depends on whether they have been calibrated to measure whole blood or plasma-equivalent glucose concentrations (Roomp and Rand, 2013). More specifically, veterinary PBGMs, like AlphaTrak (Abbot Animal Health) and GlucoPet (Animal Diabetes) that have been specifically designed for dogs and cats (Cohen et al., 2009; Johnson et al., 2009; Moretti et al., 2010; Dietiker-Moretti et al., 2011; Ford and Lynch, 2013), give plasma-equivalent glucose concentrations that can be lower or higher but are strongly correlated with those expected with an automated chemistry analyser and the differences are not large enough to negatively affect clinical decisions or patient outcome (Cohen et al., 2009; Johnson et al., 2009; Roomp and Rand, 2009; Ford and Lynch, 2013; Roomp and Rand, 2013; Surman and Fleeman, 2013;). Other PBGMs for veterinary use include the g-Pet (Woodley Equipment) and the i-Pet (UltiCare); their accuracy seems to be adequate based on manufacturers' results but, at the time of writing, there are no independent studies published in the peer-reviewed literature to support this (Surman and Fleeman, 2013). On the contrary, most human whole-blood PBGMs, which are the most widely available devices, systemically underestimate glucose concentrations in dogs and cats (by as much as 16-40% or by approximately 18-36 mg dL^{-1} at normal and low glucose levels and at a greater extend for hyperglycaemic samples) and human plasma-equivalent PBGMs give intermediate values (Cohen et al., 2009; Roomp and Rand, 2009; Ford and Lynch, 2013; Roomp and Rand, 2013; Surman and Fleeman, 2013). The reason for this is suspected to be the different distribution of glucose between plasma (approximately 50% in humans, 87.5% in dogs and 93% in cats) and red blood cells (approximately 50% in humans, 12.5% in dogs and 7% in cats) along with the fact that the porous membrane of PBGMs test strips separates red blood cells from plasma and the analysis is performed only in the latter (Johnson et al., 2009; Roomp and Rand, 2009;

Surman and Fleeman, 2013). To further complicate this issue, the human PBGM Wavesense Presto (AgaMatrix) has been shown to give overall higher glucose concentration values, to overestimate low and to underestimate high glucose concentrations in cats (Hoenig et al., 2012) and in another study of six different human PBGMs in cats, both higher and lower glucose values than those obtained by a chemistry analyser were obtained (Dobromylskyj and Sparkes, 2010). For all these reasons, it is recommended to use a veterinary PBGM, and preferably one of those that have been evaluated in independent studies, to measure blood glucose concentration in dogs and cats. If this is not feasible due to lack of commercial availability, a human device (preferably a plasma-equivalent PBGM) that has been shown to be characterized by a systemic bias when used in dogs and cats should be selected and the results should be interpreted keeping in mind the expected error of the measurements (Table 3). For example, in diabetic cats treated with a long-acting insulin and a very-low carbohydrate diet with the aim of achieving diabetic remission, the lower acceptable blood glucose concentration would be 50 mg dL⁻¹ or 80 mg dL⁻¹ depending on whether a whole-blood human or a veterinary PBGM is used for home glucose monitoring, respectively (Roomp and Rand, 2009). Finally, any spurious results from



Figure 1. After venous blood sampling, the needle has been removed from the syringe, and the reagent strip of the portable blood glucose meter is loaded using the drop of blood that has been created on the adaptor of the syringe.

the PBGM should be confirmed by a chemistry analyser (Dobromylskyj and Sparkes, 2010).

Another important attribute of the device is the blood volume that is needed to obtain a reading, especially when lancing is employed to obtain a drop of capillary blood (Ford and Lynch, 2013; Roomp and Rand, 2013). Veterinary PBGMs, like AlphaTrak, that require small blood volume (e.g. 0.3 µL) are preferable to those requiring larger volume (e.g. 0.6-4 µL for most human PBGMs or 1.5-1.6 µL for the g-Pet and the i-Pet meters), because the chances to get an accurate reading are increased, especially if the device is used by the owners (Cohen et al., 2009; Ford and Lynch, 2013; Roomp and Rand, 2013; Surman and Fleeman, 2013).

Some human PBGMs can also measure blood b-hydroxybutyrate concentrations using different strips than those used to measure blood glucose concentrations (Roomp and Rand, 2013). In diabetic animals that develop ketosis blood b-hydroxybutyrate increases earlier than urine acetoacetic acid (which is predominantly measured with urine dipsticks) and for this reason such devices have the potential for earlier diagnosis of this complication of diabetes mellitus (Roomp and Rand, 2013).

Different reagent strips covered with a specific enzyme are used by all PBGMs. There is a strip-to-strip variation, which may lead to some inaccuracy in blood glucose readings. Furthermore, slight variation in the size of the reaction wells of the strips as well as in their enzyme coverage (the amount of enzyme used and its dispersion on the well) may also influence the results (Ginsberg, 2009a). To account for the variation among different lots of strips, most PBGMs are calibrated for each new lot using a control solution, a lot-specific calibration strip or a lot-specific code (Cohen et al., 2009; Johnson et al., 2009).

The stability of the strips is guaranteed and the corresponding expiry date provided by the manufacturer is valid when they are stored in their tightly sealed package under the recommended storage conditions. A shorter life span may result from high temperature, high humidity or a strip package that

Table 3. Selected commercially available portable blood glucose meters (PBGMs) and their accuracy for measurement of blood glucose concentration in dogs and cats

PBGm	Designed for ^a	Tested in ^a	Blood glucose concentration ^b			Source
			Low	Normal	High	
Accu-Chek	H	D	U	U	U* ¹	(1)
Accu-Chek Active	H	D	U	U	U	(2)
Accu-Chek Active	H	C	NC	NC	NC* ¹	(8)
Accu-Chek Compact	H	C	NC	NC	NC* ¹	(8)
Accu-Chek Easy	H	D	U	U	U	(3)
Accu-Chek Simplicity	H	D	NC	U	U	(6)
Accu-Chek Simplicity	H	C	U	U	U* ¹	(4)
Ascensia Breeze	H	C	NC	NC	NC* ¹	(8)
AlphaTrak	D/C	C	U	U	O	(5)
AlphaTrak	D/C	D	O	O	O* ¹	(1)
Ascensia Elite	H	C	U	U	U	(5)
Contour	H	D	U	U	U* ¹	(1)
Elite	H	D	U	U	U* ¹	(1)
ExacTech RSG	H	D	U	U	U	(3)
FreeStyle	H	C	NC	NC	U* ¹	(8)
Glucometer DEX	H	D	NC	U	NC	(6)
Glucometer DEX	H	C	U	U	NC* ¹	(4)
Glucometer Elite	H	D	U	U	U	(6)
Glucometer Elite	H	D	U	U	U	(3)
Glucometer Elite	H	C	U	U	U* ¹	(4)
Glucometer Elite 2000	H	D/C	U	U	U* ¹	(7)
Glucometer Encore	H	D	U	U	U	(3)
Glucometer Glucofilm	H	D	U	U	U* ¹	(3)
GlucoPet	D	D	O	O	O	(2)
Glucotrend	H	D/C	U	U	U* ¹	(7)
OneTouch	H	D	U	U	U* ¹	(1)
OneTouch Ultra	H	C	NC	NC	NC* ¹	(8)
Precision	H	D	U	U	U* ¹	(1)
Precision QID	H	D	NC	U	U	(6)
Precision QID	H	C	NC	NC	NC* ¹	(4)
Supreme Plus	H	C	NC	NC	U* ¹	(8)
SureStep	H	D	NC	U	U	(6)
SureStep	H	C	U	U	U* ¹	(4)

a: C: cats; D: dogs; H: humans

b: NC: non-consistent overestimation and/or underestimation of blood glucose concentration; O: overestimation of blood glucose concentration;

U: underestimation of blood glucose concentration

*1: bias increased at higher blood glucose concentrations

Sources: (1) Cohen et al., 2009; (2) Johnson et al., 2009; (3) Cohn et al., 2000; (4) Wess and Reusch, 2000a; (5) Zini et al., 2009 (6) Wess and Reusch, 2000b (7) Wess and Reusch, 2000c (8) Dobromylskij and Sparkes, 2010



Figure 2. Capillary blood sampling from ear pinna of a dog for measurement of blood glucose concentration with a portable blood glucose meter: a) the margin of the pinna has been shaved; b) the puncture site is warmed with a damp cloth; c) a gauge is held under the puncture site; d) the lancing device is opposed to the puncture site; e) a drop of capillary blood appears immediately after lancing; f) the capillary blood is used to load the reagent strip, and g) within a few seconds glucose reading appears on the screen of the portable blood glucose meter

has been left open. In such cases, at least for humans, the divergence of the readings is unpredictable and underestimation or overestimation of blood glucose concentration may equally occur (Bamberg et al., 2005). Therefore, strips should be stored properly and the package with the unused strips should be tightly closed immediately after the removal of a strip. Also, use of expired strips is strongly discouraged; some PBGMs automatically detect the expiration date and they alert the operator.

The accuracy of blood glucose measurements can be influenced by environmental factors, such as altitude and temperature. In addition to data provided by the manufacturers, the influence of these factors on the human applications of PBGMs has been assessed under extreme conditions (Gautier et al., 1996). Glucose oxidase, but not glucose dehydrogenase, biosensor strips are sensitive to blood oxygen concentration. A decrease in blood oxygen concentration that may occur at high altitude, can lead to overestimation of the glucose value and vice versa. For the same reason and because of the higher oxygen concentration in capillary compared to venous blood, glucose oxidase strips may be more accurate when used with the former samples which have been used for their calibration. Furthermore, the accuracy of PBGMs using such strips may be compromised in patients receiving supplemental oxygen (Cohen et al., 2009; Ginsberg, 2009b).

The influence of the temperature on glucose measurements is unpredictable. For this reason, temperature sensors have been incorporated into modern PBGMs to warn operator against extreme temperatures (Fazel et al., 1996; Oberg and Ostenson, 2005).

Blood glucose concentration measurement by PBGMs can be performed with venous or with capillary blood samples and the latter can be obtained from different sites (ear pinnae or carpal/metacarpal pads). When capillary blood sampling is selected, the reagent strip of the PBGM is directly touched to the drop of blood on the animal's skin, whereas when venous blood is used a drop of blood from the syringe can be placed on the wrapper of the strip (Cohen et al., 2009) or a drop can be created on the adaptor of the syringe after removal of needle (Fig. 1). Different blood samples may affect readings due to differences in glucose and oxygen concentration, local blood circulation and possible contamination by natural or chemical substances. Apart from the higher oxygen tension, capillary blood also has higher glucose concentrations compared to venous blood, due to tissue utilization of glucose and this difference is higher postprandially than during fasting (Johnson et al., 2009). However, capillary and venous blood glucose concentrations seem to have a good correlation in companion animals (Alt et al., 2007; Cohen et al., 2009).

The most frequently used site for capillary blood sampling in dogs and cats is the lateral margin of the



Figure 3. Capillary blood sampling from the carpal pad of a dog for measurement of blood glucose concentration with a portable blood glucose meter: a) the sampling site is pinched with two fingers; b) a drop of capillary blood appears immediately after lancing.

ear pinna (Ford and Lynch 2013) or the inner surface of the pinna (Rios and Ward, 2008). Blood collection may be facilitated by shaving (lateral margin technique), warming the puncture site for 30-60 sec with a damp cloth, applying a thin layer of petroleum jelly, holding a cotton ball or a gauze under the pinna and pricking close to but not onto a visible vein (Ford and Lynch, 2013) (Fig. 2). Recently, the carpal (pisiform or wrist) non-weight bearing pads of dogs and the metacarpal pad of cats have been also used and validated as alternative sampling sites for blood glucose testing (Zeugswetter et al., 2010; Borin-Crivellenti et al., 2012; Ford and Lynch, 2013). Sampling may become easier if the site is pre-warmed and lightly pinched with two fingers for a few seconds before and after puncture until an adequate blood drop forms (Ford and Lynch, 2013) (Fig. 3). Irrespectively of the site, puncture is typically performed with commercially available lancing devices with lancets of 25-32 gauge, depending on the patient (Ford and Lynch, 2013); some lancing devices create negative pressure which may facilitate ear pinna capillary blood sampling (Rios and Ward, 2008). One exception is for capillary blood sampling from the carpal pad in dogs, where a hypodermic needle may be more appropriate (Borin-Crivellenti et al., 2012).

The presence of contaminants at the site of sampling, even in trace amounts, can significantly raise blood glucose, as it has been shown in human medicine (Ginsberg, 2009a). Hand washing is suggested prior to blood sampling in humans and cleaning of the sampling site in dogs and cats can be proposed. However, the use of any disinfectant should be avoided, until any possible effect of the substance on glucose measurement is excluded.

The effect of three anticoagulants (EDTA, lithium heparin and fluoride) on glucose measurements in canine blood was determined for five different PBGMs. While no difference was found among EDTA, lithium heparin and blood without anticoagulant, the concentration of glucose in fluoride-anticoagulated blood was underestimated by one of the devices whose manufacturer advised against the

use of anticoagulants (Wess and Reusch, 2000b). However, in a different study evaluating feline blood samples without anticoagulant, with lithium heparin and with fluoride oxalate, no effect on glucose concentration was witnessed for six different human PBGMs (Dobromylskyj and Sparkes, 2010). This topic remains controversial but it is of minor clinical importance because PBGMs readings are typically obtained immediately after collection of capillary or venous blood samples without anticoagulant.

The haematocrit can influence blood glucose measurements by PMBGs. These devices measure glucose in a complex mixture of whole blood and plasma, while plasma glucose is used for their calibration during the manufacturing process. The effect of haematocrit is related, not only to the presence of glucose in red blood cells, but also to the influence of erythrocytes on various mechanisms operating during PBGM measurement, such as the diffusion of plasma to the test strip reagent pad, the enzymatic reaction and the proper function of the electrodes of the device (Wiener, 1991). For example, haemoconcentration and the associated increased blood viscosity may lead to obstruction of the holes of the strip filter and to a decreased plasma diffusion rate and consequently, to falsely lower glucose readings, whereas artificially increased glucose concentrations are expected in anaemic patients (Tang et al., 2000). In general, PBGMs employing glucose dehydrogenase are suggested for human patients with abnormal haematocrit values and efforts have been made to apply mathematical equations for the correction of glucose readings for haematocrit values (Musholt et al., 2011).

The effect of the haematocrit is expected to also apply in dogs and cats (Paul et al., 2011) and perhaps to become more important with the use of human PBGMs which have been evaluated within the narrower haematocrit range of humans. Although, a veterinary PBGM was more accurate than a human one in canine blood samples with normal or increased haematocrit, the reverse was true in anaemic samples (Paul et al., 2011). In any case, PBGMs seem to consistently overestimate blood glucose concentration in

anaemic dogs (Johnson et al., 2009) and cats (Wess and Reusch, 2000a), although a recent study did not reveal an effect of haematocrit on the accuracy of six different human PBGMs in feline blood samples (Dobromylskyj and Sparkes, 2010).

Plasma concentration of certain metabolites (i.e. triglycerides, creatinine, uric acid, proteins) have also been reported to influence the measurement of blood glucose in humans with PBGMs (Ervin and Kiser, 1999), more notably when their concentration is altered during certain diseases states. For example, high triglyceride concentration may decrease the amount of glucose in the capillary blood, whereas, uric acid at extreme concentrations erroneously increases glucose values (Ginsberg, 2009a). However, the latter effect is unlikely to be of clinical significance in companion animals due to the differences in purine metabolism compared to humans, and the effects of the other metabolites on the accuracy of PBGMs in dogs and cats have not been studied.

Various drugs, such as paracetamol, L-dopa, tolazamide, and ascorbic acid, interact with the electrode of PBGMs employing the electrochemical glucose oxidase system and alter glucose readings in human blood (Ginsberg, 2009a). To the author's knowledge similar data are not available in the veterinary literature.

The importance of following manufacturer's instructions by the operator is fundamental for the accurate measurement of blood glucose concentration with PBGMs in both the hospital and the home setting. Operator error can become a major source of misleading results and therefore adequate training of operators is of great importance.

CONCLUDING REMARKS

Due to several factors, including the analytical method employed, results from different PBGMs are likely to differ. Even though such differences

are not always clinically relevant and may not affect diagnostic or therapeutic decisions (Johnson et al., 2009), veterinary PBGMs should be preferred over human devices. If this is not feasible, human PBGMs that have been shown to be characterized by a systemic bias when used in dogs and cats must be selected. Furthermore, it is always a good idea to compare the results of any newly purchased PBGM with that of the previous device and/or of a chemical analyser.

Moreover, the clinician must be aware of the potential limitations of PBGMs and the factors that can interfere with the result, like storage conditions, altitude, temperature, blood sampling method, haematocrit etc. For this reason, when consecutive measurements of blood glucose are necessary, as in diabetic animals, all these factors should remain as constant as possible so that any influence on the measured blood glucose concentration would be uniform.

Since the advent of the PBGMs, home monitoring of diabetic dogs and cats has become feasible. Also, the immediate availability of the results offers an important advantage of PBGMs over automated chemistry analysers in the critical care setting. In any case, training of the operator, being either a pet owner or a health care provider, is essential to obtain reliable results. Finally, as with all diagnostic tests and devices, the clinician should combine clinical judgment and experience with the measurement of blood glucose concentration by PBGMs and interpret the result in light of all the information available.

CONFLICT OF INTEREST

None of the authors of this article has a financial or personal relationship with other people or organizations that could inappropriately influence or bias the content of this paper. ■

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