Use of insulin glargine in dogs with diabetes mellitus

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The objective of this study was to evaluate the safety and efficacy of insulin glargine in dogs with diabetes mellitus (DM). Twelve client-owned dogs with DM were included. All dogs received insulin glargine every 12 hours for at least six months, re-evaluations were performed after one, two, four, eight, 12 and 24 weeks and included clinical signs, blood glucose curves (BGCs) and measurement of serum fructosamine concentrations. Mean blood glucose concentrations were significantly lower after two weeks of treatment and remained significantly lower for the duration of the study. By week 24, polyuria/polydipsia had improved in 91 per cent of the dogs. No clinical signs that could have been caused by hypoglycaemia were observed. Based on BGCs and remission of the clinical signs for judging the success of the treatment, 58, 33 and 8 per cent of the dogs attained good, moderate and poor glycaemic control by week 24 of the study, respectively. Insulin glargine administered subcutaneously twice daily is a possible and safe method of treatment for dogs with naturally occurring DM. Although only a few studies are available on the use of other types of insulin in dogs, their success rate is somewhat greater than that with insulin glargine.

Materials and methods

Inclusion criteria
Client-owned dogs with DM were used if the owners were willing to return to the hospital for six re-evaluations over a 24-week period. The diagnosis of DM was based on characteristic clinical signs, fasting hyperglycaemia, glucosuria and increased serum fructosamine levels (>340 μmol/l). Dogs could be newly diagnosed or previously diagnosed but considered poorly regulated on their current insulin regimen. Dogs with a serious concurrent disease (e.g., hypothyroidism, hyperadrenocorticism, neoplasia, renal insufficiency) and dogs that had received glucocorticoids or progestagens within the previous 60 days were not enrolled.

Dogs with ketoacidosis requiring aggressive management were used if their condition had been stabilised by medical treatment, including regular insulin therapy. Dogs with lower urinary tract infections with little likelihood of a substantial impact on diabetic management were also included.

Study design
This was a prospective, uncontrolled clinical trial that was conducted over 24 weeks. On the initial visit (day 0), patient histories and bodyweights were recorded, physical examinations and urine analyses were performed, and complete blood counts, serum biochemical profiles, serum fructosamine concentrations and urine cultures were obtained. All dogs received insulin glargine (Lantus; Aventis Pharmaceuticals) every 12 hours. The starting dose was 0.25 to 0.5 U/kg subcutaneously (SQ) every 12 hours. Dogs were fed with a complete and balanced diet without simple sugars every 12 hours at the time of insulin administration. Re-evaluations were performed one, two, four, eight, 12 and 24 weeks after and included an assessment of clinical signs, blood glucose curves (BGC) and serum fructosamine concentrations. Usually, insulin and food were given in the hospital and glucose concentrations were measured before the insulin injection and every two hours thereafter for 12 hours. For dogs that were reluctant to eat in the hospital, insulin and food were administered at home and the BGC was started as soon as they arrived at the hospital (no more than two hours after the insulin injection). The insulin dose was adjusted by 5 to 20 per cent at each evaluation until the blood glucose nadir for the
Comparison with the results on day 0, the T-bars represent the main body of the data. (*) p <0.05, box represents the interquartile range (ie, 25 to 75% range). The blood glucose concentration before treatment (week 0) was obtained from the fasting blood glucose concentration. The blood glucose concentration before therapy with insulin glargine is expressed as the median (range) of the fasting blood glucose concentrations of the 12 dogs, and after therapy is expressed as the mean (±sd) blood glucose concentrations in the blood glucose curves.

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FIG 1: Box plots of mean blood glucose concentrations in the blood glucose curves for 12 dogs with naturally acquired diabetes mellitus treated by insulin glargine administration twice daily for 24 weeks. The blood glucose concentration before treatment (week 0) was obtained from the fasting blood glucose concentration. The box represents the interquartile range (ie, 25 to 75% range). The horizontal bar in the box represents the median value. For each box plot, the T-bars represent the main body of the data. (*) p <0.05, compared with the results on day 0.

BGC was 5 to 10 mmol/l. The proportional adjustment of the insulin dose was decided upon by the managing clinician for each dog, based on the dog’s bodyweight, response to the previous insulin dose on the glucose curve, serum fructosamine concentration and clinical signs of response to therapy, or evidence of hypoglycaemic episodes.

Analytical methods
Blood samples for the BGCs were collected from all animals using the Vaculance method, from the inner pinna, using a Microlet Vaculance lanceting device (Bayer Diagnostics), as previously described (Wess and Reusch 2000a). Serial whole blood glucose concentrations were measured using a hand-held glucometer (Ascensia ELITE, Bayer HealthCare or Optium Xceed, Abbott Animal Health) validated for dogs (Wess and Reusch 2000b, Fragiacco and others 2009). Haematology, serum biochemical profiles and urinalesys were performed by standard laboratory methods. Fructosamine analyses were performed using a Cobas Integra 700 (Roche) analyser and a commercial reagent (fructosamine; Roche); reference range was set at 207 to 340 μmol/l.

Assessment of efficacy
The blood parameters used to assess the control of the disease included mean blood glucose concentrations during the BGCs, the blood glucose nadir and the serum fructosamine concentration. Control of glycaemia was classified as good, moderate or poor according to the following criteria: mean blood glucose concentrations during BGCs <13 mmol/l were considered good, 13 to 17 mmol/l moderate and >17 mmol/l poor; and serum fructosamine concentrations <450 μmol/l were considered good, 450 to 550 μmol/l moderate and

TABLE 1: Blood glucose concentrations, blood glucose nadir concentrations, serum fructosamine concentrations and insulin doses (median and range or mean±sd) before and 1, 2, 4, 6, 8, 12 and 24 weeks after the beginning of insulin glargine therapy in 12 dogs with naturally acquired diabetes mellitus

| Parameters | Before | 1 week | 2 weeks | 4 weeks | 8 weeks | 12 weeks | 24 weeks |
|------------|--------|--------|---------|---------|---------|----------|---------|
| Blood glucose concentration (mmol/l) | 26.40 (9.6-33.1) | 18.50 (8.4-30.5) | 13.2±5.6 (4.8-23.9) | 12.3±6.9 (4.0-21.8) | 11.2±4.4 (3.6-15.9) | 12.1±4.0 (6.3-18.5) | 12.4±3.6 (5.2-17.6) |
| Glucose nadir (mmol/l) | Not determined | 15.7 (3.5-31.8) | 10.1 (3.2-22.6) | 8.4 (2.2-18.9) | 8.5 (1.8-14.9) | 6.5 (3.1-16.2) | 7.0 (3.3-12.3) |
| Fructosamine (μmol/l) | 611 (320-815) | 593 (359-842) | 503 (418-748) | 540 (385-663) | 588 (359-597) | 532 (461-584) | 484 (342-691) |
| Insulin (U/kg) every 12 hours | 0.27 (0.18-0.53) | 0.37 (0.22-0.53) | 0.25 (0.18-0.41) | 0.45 (0.25-0.67) | 0.47 (0.15-1.09) | 0.56 (0.22-1.25) | 0.60 (0.11-0.67) |

Data analysis
The blood glucose concentrations used to generate the BGCs were reported as mean±sd, whereas, all other data are given as ranges and medians. Data recorded before and after insulin glargine therapy were compared with a Wilcoxon matched pairs test and with the Mann-Whitney U test. Data were analysed using a commercially available software program (GraphPad Prism). The level of significance was set at p<0.05.

Results

Animals and results upon admission
Twelve dogs (eight females, three entire, five neutered; and four entire males) with DM fulfilled the inclusion criteria and were enrolled. The entire female dogs were neutered within three weeks after enrolment in the study. The median age of the study population was 11 years (range 5 to 15), the median bodyweight was 9.5 kg (range 3.7 to 45) and the median body condition score was 5/9 (range 3/9 to 8/9). Nine dogs were newly diagnosed with DM and three had been previously treated with an insulin zinc suspension with poor control of the disease. Three dogs with urinary tract infection were treated with antibiotics.

Outcome
The median insulin dose was 0.27 U/kg (range 0.18 to 0.53) every 12 hours upon admission and increased significantly (p<0.05) to a medi-
an of 0.60 U/kg (range 0.11 to 1.07) every 12 hours after 24 weeks of therapy (Table 1). All dogs displayed subjective improvements in their clinical signs throughout the study period. Polyuria/polydipsia was present in 92 per cent upon admission and in 8 per cent of the dogs after 24 weeks of therapy. Bodyweight was not different after six months of therapy. The mean (±sd) BGC blood glucose concentrations in the median (range) glucose nadir values and the median (range) fructosamine concentrations before and one, two, four, eight, 12 and 24 weeks after the beginning of insulin glargine therapy are reported in Table 1. The mean (±sd) BGC glucose concentrations were significantly lower after weeks two, four, eight and 12 as well as at week 24 of insulin glargine therapy compared with the fasting glucose before treatment (Fig 1).

The number of hours after insulin injection at which the glucose nadir occurred varied between the BGCs, ranging from zero to 12 hours. Taking into consideration all of the BGCs, the nadirs were most commonly observed after six, eight and 10 hours (24, 19 and 16 per cent of all BGCs, respectively) (Fig 2). Serum fructosamine concentrations were found to be significantly lower after eight and 24 weeks of insulin glargine therapy compared with the concentrations before treatment.

Where the assessment of glycaemic control was solely based on the owners’ opinions, 11 (92 per cent) dogs were well and one dog (8 per cent) was poorly controlled by week 24 of the study. Where the assessment was based on clinical parameters obtained in the hospital (results of the physical examination and bodyweight stability) and results of the BGCs, seven (58 per cent), four (33 per cent) and one (8 per cent) of the dogs were, respectively, well, moderately and poorly controlled, respectively. The assessment based on fructosamine concentrations resulted in good, moderate and poor control in three (25 per cent), seven (58 per cent) and two (17 per cent) of the dogs.

**Adverse effects and hypoglycaemia**

No reactions at the site of glargine administration were reported. Despite a blood glucose nadir of <5 mmol/l being identified in 15 per cent of the BGCs, symptomatic hypoglycaemia was not observed by the clinicians or reported by the owners.

**Discussion**

The mean blood glucose concentrations were significantly reduced after two weeks of insulin glargine treatment and remained significantly lower for the duration of the study. No adverse events associated with insulin glargine administration were observed. These results suggest that insulin glargine administered subcutaneously twice daily is a possible method of treatment for naturally occurring DM in dogs, and that it is an alternative to other insulin preparations that have been shown to be effective in the treatment of canine DM (Church 1981, Horn and Mitten 2000, Monroe and others 2005, Palm and others 2009).

Insulin glargine is commonly used in human beings as a once-daily basal insulin therapy, often supplemented with ultrashort-acting insulin analogues given at mealtimes (Garg and others 2010). In the present study, the authors chose to evaluate twice-daily insulin glargine therapy. Although an experimental study in dogs showed that insulin glargine has a duration of action of approximately 18 to 24 hours with a pronounced peak of action at seven hours (Mori and others 2005), previous studies showed that once-daily insulin administration in dogs results in very high insulin doses and an increased risk of hypoglycaemia (Hess and Ward 2000). Moreover, most dogs achieve better control of the disease when treated with twice-daily insulin administration (Horn and Mitten 2000, Monroe and others 2005).

In terms of the resolution of clinical signs that was achieved with insulin glargine, the results were similar to those of isophane insulin in a study of 54 dogs (Lorenzen 1992) and a porcine insulin zinc suspension in a study of 53 dogs (Monroe and others 2005). The median insulin glargine dose at the end of the study (0.60 U/kg every 12 hours) was comparable with doses in other studies where insulin was administered every 12 hours (Lorenzen 1992, Monroe and others 2005).

Although most of the dogs included in this study had lower serum fructosamine concentrations after 24 weeks compared with their initial values, the concentrations of fructosamine in seven and two of the 12 dogs were indicative of moderate and poor control of the disease, respectively. Fructosamine is continuously formed in the body as the result of a non-enzymatic reaction between glucose and plasma proteins and the concentration mainly depends on blood glucose levels (Reusch and Haberer 2001). Discrepancies between the fructosamine concentration and the clinical picture of the dogs are more common when the result of blood glucose concentrations, can occur in some dogs with diabetes (Nelson 2010). In a study in which dogs were treated with isophane insulin every 12 hours for six months, the fructosamine concentration in four out of eight dogs was >450 μmol/l, despite the good control of clinical signs in all dogs (Thoresen and Lorenzen 1997).

Hypoglycaemia is the most serious condition seen in insulin-treated dogs and should always be avoided. Asymptomatic hypoglycaemia was observed during the BGCs. A possible explanation for the identification of asymptomatic hypoglycaemia could be related to the bias of the portable blood glucose monitoring devices. Despite the fact that the hand-held glucometers used in the present study were validated for dogs, they tended to underestimate the blood glucose concentration when compared with the hexokinase reference results measured from the same blood sample (Wess and Reusch 2000b, Fracassi and others 2009). The paucity of clinical signs compatible with hypoglycaemia seen in the dogs in this study, coupled with the results reported in human beings (Fulcher and others 2005, Garg and others 2010) and cats (Marshall and others 2009), indicate that glargine is safe for the treatment of canine DM.

In human medicine, glargine is a peakless insulin (Heinemann and others 2000). A clear nadir was identified in almost all of the BGCs of the present study, indicating that the pharmacokinetics and pharmacodynamics of insulin glargine are not the same in dogs as they are in human beings. The time to glucose nadir was extremely unpredictable, ranging from before the insulin injection to 12 hours after insulin administration. A possible explanation for this variability could be differences in the duration of insulin glargine action, suggesting an overlap of the effects of the two injections administered every 12 hours. Another contributing factor could be the lack of standardisation of the diet. Some dogs received a mix of dry and moist formulation and it is possible that the ratio between the two components was not kept constant during the entire study period. A further possible explanation for the time to nadir variability could be variable gastric emptying and/or postprandial variations in blood glucose via variable absorption of the calories consumed as well as the potential for the variation in the glycaemic indices for the food consumed. A large variation with regard to the time of the glucose nadir can have substantial consequences for the management of the disease and thus monitoring with BGCs is mandatory for the dogs with diabetes treated with insulin glargine.

The present study had a number of limitations, such as the small number of dogs included. However, the study was only intended to be a pilot study to demonstrate that glargine is a safe, alternative, twice-daily treatment option to stabilise the majority of dogs diagnosed with DM. Another limitation of the study is that blood samples for the BGCs were only taken every two hours for the first 12 hours and not for 24 hours. This limited the ability to determine the true duration of insulin action in dogs, in which the action of insulin lasts for longer than 12 hours. Because of this, determination of true nadir values was also difficult, and may have prevented the identification of some dogs with low-night-time blood glucose levels. However, 12-hour BGCs are generally considered reliable for assessing the adequacy of diabetic control in most clinical patients (Feldman and Nelson 2004).

Other limitations of the present study were that the diet and exercise were not standardised in the study population, which are factors that may have influenced control of the disease. Although the owners were permitted to feed their dogs any commercial diet that the owner considered appropriate, it is possible that the ratio between the two components was not kept constant. A diet high in fibre and low in simple carbohydrates was recommended.

Although veterinarians use a variety of insulin products, in most countries only a porcine lente product (a porcine insulin zinc suspension) has been approved for use in dogs. In fact, this U-40 insulin preparation was recommended as a first-choice treatment for dogs diagnosed with diabetes (Rucinsky and others 2010). However, according to the Food and Drug Administration (FDA), porcine lente...
Because the insulin glargine was not compared with other insulin products in this study, a direct comparison of the efficacy of insulin glargine versus other treatment options is difficult to perform. However, based on mean blood glucose concentration during 12-hour BGcs, 58 per cent of dogs in the present study obtained good control of hyperglycaemia, which is less than the reported 75 per cent of dogs with good control in a similar study where a porcine insulin zinc suspension was used (Monroe and others 2005). It is the authors’ opinion that insulin glargine appears to be safer than the porcine insulin zinc suspension in terms of episodes of iatrogenic hyperglycaemia, even though its disease control success rate appears to be lower.

References

CHURCH, D. B. (1981) The blood glucose response to three prolonged duration insulins in canine diabetes mellitus. Journal of Small Animal Practice 23, 301-310

FELDMAN, E. C. & NELSON, R. W. (2004) Canine diabetes mellitus. In Canine and Feline Endocrinology and Reproduction. 3rd edn. Eds E. C. Feldman, R. W. Nelson. Saunders Elsevier. pp 486-538

FRACASSI, F., HADAR, G. S., PIETRA, M. & FAMIGLI-BERGAMINI, P. (2009) Assessment of two portable blood glucose meters for use in dogs and cats. Journal of Veterinary Clinical Sciences 2, 100-121

FULCHER, C. R., GILBERT, R. E. & YUE, D. K. (2005) Glargine is superior to neutral protamine Hagedorn for improving glycated haemoglobin and fasting blood glucose levels during intensive insulin therapy. Internal Medicine Journal 35, 586-542

GARC, S., MOSER, E., DAIN, M. P. & RODIONOVA, A. (2010) Clinical experience with insulin glargine in type 1 diabetes. Diabetes Technology & Therapeutics 12, 833-846

HEINEMANN, L., LINKESCHOVA, R., RAVE, K., HOMPESCH, B., SEDLAK, M. & HEISE, T. (2000) Time-action profile of the long-acting insulin analog insulin glargine (HOE901) in comparison with those of NPH insulin and placebo. Diabetes Care 23, 644-649

HESS, R. S. & WARD, C. R. (2000) Effect of insulin dosage on glycemic response in dogs with diabetes mellitus: 221 cases (1993-1998). Journal of the American Veterinary Medical Association 216, 217-221

HORN, B. & MITTEN, R. W. (2000) Evaluation of an insulin zinc suspension for control of naturally occurring diabetes mellitus in dogs. Australian Veterinary Journal 78, 831-834

LORENZEN, F. H. (1992) The use of subcutaneous insulin for the control of diabetes mellitus in dogs. Acta Veterinaria Scandinavica 33, 219-227

MONROE, W. E., LAXTON, D., FALLIN, E. A., RICHTER, K. P., SANTEN, D. R., PANCIERA, D. L., TOWELL, T. L., WILLIAMS, K. A., HART, J. R., HILL, S., FINKLER, M. B. & SHINN, J. S. (2005) Efficacy and safety of a purified porcine insulin zinc suspension for managing diabetes mellitus in dogs. Journal of Veterinary Internal Medicine 19, 675-682

MOU, A., SAKO, T., LEE, P., MOTOIKE, T., IWASE, K., KANAYA, Y., FUKUTA, H., MIZUTANI, H. & ARAI, T. (2008) Comparison of time-action profiles of insulin glargine and NPH insulin in normal and diabetic dogs. Veterinary Research Communications 32, 563-573

MARSHALL, R. D., RAND, J. S. & MORTON, J. M. (2009) Treatment of newly diagnosed diabetic cats with glargine insulin improves glycaemic control and results in higher probability of remission than protamine zinc and lente insulin. Journal of Feline Medicine and Surgery 11, 683-691

NELSON, R. W. (2010) Canine diabetes mellitus. In Textbook of Veterinary Internal Medicine. 7th edn. Eds S. J. Ettinger, E. C. Feldman. WB Saunders. pp 1782-1796

PALM, C. A., BOSTON, R. C., REFAL, K. R. & HESS, R. S. (2009) An investigation of the action of Neutral Protamine Hagedorn human analogue insulin in dogs with naturally occurring diabetes mellitus. Journal of Veterinary Internal Medicine 23, 50-55

REUSCH, C. E. & HABERER, B. (2001) Evaluation of fructosamine in dogs and cats with hypo- or hyperproteinaemia, azotaemia, hyperlipidaemia and hyperbilirubinaemia. Veterinary Record 148, 370-376

REUSCH, C. E., ROBBEN, J. H. & KOOISTRA, H. S. (2010) Endocrine pancreas. In Clinical Endocrinology of Dogs and Cats. 2nd edn. Eds A. Rijnberk, H. S. Kooistra. Schiltersche. pp 159-185

RUCINSKY, R., COOK, A., HALEY, S., NELSON, R. & ZORAN, D. L. (2010) AAHA diabetes management guidelines for dogs and cats. Journal of the American Animal Hospital Association 46, 215-224

THORESEN, S. I. & LORENZEN, F. H. (1997) Treatment of diabetes mellitus in dogs using isophane insulin penfills and the use of serum fructosamine assays to diagnose and monitor the disease. Acta Veterinaria Scandinavica 38, 137-146

WESS, G. & REUSCH, C. (2000a) Capillary blood sampling from the ear of dogs and cats. Veterinary Record 146, 50-55

WESS, G. & REUSCH, C. (2000b) Evaluation of five portable blood glucose meters for use in dogs. Journal of the American Veterinary Medical Association 216, 203-209

WEAVER, K. E., ROZANSKI, E. A., MAHONY, O. M., CHAN, D. L. & FREEMAN, L. M. (2006) Use of glargine and lente insulins in cats with diabetes mellitus. Journal of Veterinary Internal Medicine 20, 234-238