

Evaluation of an insulin zinc suspension for control of naturally occurring diabetes mellitus in dogs

B HORN^a and RW MITTEN

Veterinary Clinic and Hospital, University of Melbourne, Werribee, Victoria 3030

Objective To evaluate duration of action of an insulin zinc suspension (Caninsulin, Intervet) in spontaneously occurring cases of canine diabetes mellitus and suitability of its use as a once daily administered insulin for treatment of this disease.

Design Eight client-owned canine diabetics were included in a prospective pilot study. All dogs had been treated with Caninsulin for a minimum of 2 months and were considered on clinical grounds to be adequately stabilised.

Procedure Dogs were hospitalised for 24 h and blood collected every 2 h via indwelling venous catheters for blood glucose determination.

Results Once daily Caninsulin administration failed to maintain glycaemic control for greater than 13 h in five of eight dogs, but acceptable blood glucose concentrations were maintained for 22 h and greater than 24 h in two others. One dog became distressed during hospitalisation and the blood glucose curve did not show an identifiable response to the insulin.

Conclusion Most diabetic dogs may require twice daily administration of Caninsulin for satisfactory glycaemic control, but once daily administration may be adequate in some animals. More comprehensive investigation into duration of activity of Caninsulin is warranted.

Aust Vet J 2000;78:831-834

Key words: Dog, diabetes mellitus, insulin, insulin zinc suspension.

SC Subcutaneously

Insulin therapy has remained the mainstay of treatment for canine diabetes mellitus.¹ The aim is to alleviate signs of polyuria, polydipsia, polyphagia and weight loss.² This is achieved by maintaining blood glucose concentrations below the renal threshold (10 to 13 mmol/L) for the majority of the time.² Concurrently, any complications brought about by nonenzymatic glycosylation of structural and nonstructural proteins must be minimised and the potentially serious problem of hypoglycaemia should be avoided.

The veterinary clinician must, therefore, select an insulin with a duration of activity approaching 24 h, if given once daily, or 12 h, if given twice daily. If the activity of insulin is not sustained for the appropriate duration, the type of insulin or frequency of administration should be changed. For instance, if insulin is given once daily but appears to be effective for only 15 to 20 h then changing to a longer-acting insulin should be considered. Alternatively, continuing with the same insulin but giving it twice daily at two-thirds of the dose may be suitable.² If duration of activity is closer to 14 h, then changing to twice daily administration might be more appropriate, as changing to longer-acting types usually only extends activity by 6 to 8 h at best.³

Given that many different insulin injection regimens have been advocated,^{2,4-6} it would appear that no one insulin is ideal

in all diabetic dogs. In addition, although different insulins can be classified as having short, intermediate or prolonged action, there is potentially great variation in the duration of a particular insulin preparation between individuals. Therefore, the insulin regimen must be tailored for each patient. The owner preference for once daily regimens and the attending clinician's experience and preference for a particular insulin must also be taken into consideration.

An insulin zinc suspension specifically targeted for use in canine patients became available in 1988 in Australia, Canada and the UK. Caninsulin (Intervet, Australia) is an aqueous suspension of highly purified porcine insulin, consisting of 30 % amorphous and 70 % crystalline zinc insulin. It has been marketed for use once daily in the majority of diabetic dogs. It is claimed that the amorphous fraction will reach peak activity approximately 3 h following SC administration and has a total useful effect for about 8 h. Thereafter, the therapeutic effect is produced by the crystalline fraction, which has slower onset, with a maximum effect at 8 to 14 h and a duration of approximately 24 h.

In one study of the pharmacokinetics of Caninsulin, plasma insulin and glucose concentrations were monitored every 2 h in 10 naturally occurring diabetic dogs given Caninsulin SC once daily.⁷ Peak plasma insulin concentrations occurred at 2 to 6 h and at 8 to 14 h. Insulin concentration remained above pre-injection concentrations for 14 to 24 h: that is, for 14 h in two dogs; 16 h in five; 18 h in one, and greater than 24 h in two. Whether an insulin with duration of action of 14 to 16 h (in 7 of 10 dogs) is able to maintain glycaemic control for 24 h is uncertain.

Preliminary clinical experience with Caninsulin at the University of Melbourne (unpublished) suggested that once daily administration did not effectively control hyperglycaemia in all diabetic dogs. A review of diabetic cases seen between 1988 and 1998 was performed and identified 28 dogs that had received Caninsulin: 20 dogs were treated twice daily and 8 dogs were treated once daily (unpublished).

The aim of this pilot study was to further evaluate the efficacy of once daily Caninsulin in maintaining glycaemic control in naturally occurring canine diabetics. Our hypothesis was that because other 30% amorphous, 70% crystalline zinc insulin suspensions such as Monotard MC (Novo Nordisk) and Monotard HM (Novo Nordisk) have required twice daily administration in most dogs, Caninsulin should also require twice daily administration. This information was considered to be of clinical relevance.

Materials and methods

Patient selection

Eight client-owned dogs previously diagnosed with naturally occurring uncomplicated diabetes mellitus were included in this study. All dogs had been receiving Caninsulin once daily for at

least 2 months. Four dogs were patients of the University of Melbourne Veterinary Clinic and Hospital and four were identified by contacting Melbourne veterinary practices that had purchased Caninsulin and requesting permission to contact owners of suitable study candidates. Dogs were included if there had been reduction (but not necessarily resolution) of clinical signs of diabetes since initiating insulin therapy and if blood glucose monitoring suggested that an adequate dose of insulin was being given, as assessed by the primary care veterinarian. However, only one dog had undergone previous 24 h blood glucose monitoring. Seven were sexed females and one was an entire male. Ages were 4 to 16 years (mean 9.1). One dog was being treated concurrently for atopic dermatitis with alternate day prednisolone orally at 0.4 mg/kg.

Data collection

All owners completed a standard questionnaire under the guidance of the chief examiner^a in order to provide information regarding the dog's daily exercise, insulin and feeding regimens so these could be reproduced during hospitalisation. In addition, information about persistence of clinical signs of diabetes mellitus was obtained. Owners were questioned regarding their dog's water intake and appetite, whether body weight and insulin dose had remained stable and whether syncopal episodes had occurred in the last 2 months. Clinical examination was performed and any abnormalities were noted. In particular, presence or absence of hepatomegaly, cataracts or dermatoses, such as pyoderma, were noted. Blood cell counts, serum biochemical profiles and urinalyses were not performed because of the preliminary nature of this study and limited funding. Dogs were hospitalised for 24 h and blood was collected from indwelling catheters every 2 h for glucose determination. Dogs that were likely to become distressed by separation from owners, as assessed by owners or referring veterinarians, were hospitalised overnight prior to beginning blood sampling.

At initiation of monitoring, an 18 gauge jugular catheter was placed in each dog with the exception that cephalic catheters (16 to 18 gauge) were used in two dogs weighing over 20 kg. Catheters were maintained by flushing with heparinised saline. Blood collected into lithium heparin tubes was centrifuged within 2 h of collection and plasma harvested. Glucose was measured by a Roche, Cobas MIRA biochemical analyser using the hexokinase method.

Blood glucose curves for each animal were plotted. On the basis of these curves, dogs were divided into three groups: 24 h blood glucose control (group 1), partial blood glucose control (group 2) and poor blood glucose control (group 3). Twenty-four h control was arbitrarily defined as maintenance of blood glucose within the ideal range for a diabetic (5 to 13 mmol/L) for greater than or equal to 20 h, or in animals where only insulin dose adjustment would be expected to maintain blood glucose concentrations within this range. Partial blood glucose control was defined as maintenance of blood glucose concentrations in the 5 to 13 mmol/L range for less than 20 h. Poor blood glucose control was identified when there did not appear to be any obvious relationship between insulin administration and the blood glucose curve obtained. Statistical analysis was not performed due to the small sample size of the study group.

Insulin and feeding regime

All dogs received insulin once daily between 0 and 1 h after feeding 50% of the daily caloric food requirement (KJ). The second meal was fed 6.5 to 9 h later. The dose of insulin was 0.7 to 2.3 IU/kg (Table 1).

Results

A summary of clinical signs and abnormal findings is provided in Table 2. Results of questioning owners suggested that clinical signs of diabetes mellitus (polydipsia, polyphagia, weight loss) were not completely controlled in all dogs. However, findings on physical examination of dogs were generally unremarkable except for bilateral mature cataracts in five dogs and substantial weight loss in one. Dog 5 also had a grade III left apical systolic murmur.

Two of eight dogs met the criterion for inclusion in Group 1 (Figure 1), having experienced a duration of insulin action greater than 20 h. Although blood glucose concentrations for dog 1 were not below 13 mmol/L for greater than 20 h, a simple increase in the dose would have achieved this. Group 2 (partial blood glucose control) included five dogs: an initial drop in blood glucose was seen in all these dogs, but glucose began to rise 6 to 8 h following insulin administration. This rise was sometimes seen shortly after feeding of the afternoon meal, as in dog 3. The effect of Caninsulin appeared to last 9 to 13 h in these dogs (Figure 2). Blood glucose concentrations in the one dog in group 3 failed to show any obvious response to insulin and remained inappropriately high for the entire 24 h

Table 1. Caninsulin doses and feeding regimens in eight dogs with naturally occurring diabetes mellitus.

Dog	Insulin dose IU/kg	Feeding (hours postinsulin)	
		1st meal	2nd meal
1	1.2	0	6.5
2	1.9	1	9
3	1.0	0	7.5
4	1.0	0	8
5	2.3	0	7
6	1.4	0	8.5
7	0.7	0	8
8	1.0	0	9

Table 2. Clinical signs (obtained from owner questionnaire) and physical examination findings in eight dogs with naturally occurring diabetes mellitus.

Clinical signs and observed variables in individual dogs ^a	Decreased	Normal	Increased
Water intake ^b		1, 3, 5, 6, 7	2, 4, 8
Appetite		1, 2, 3, 4, 6, 8	5, 7
Weight	1 ^c , 5	3, 4, 6	2, 7, 8
	Absent		Present
Syncopal episodes	1, 2, 3, 4, 5, 7, 8		6
Hepatomegaly	1, 2, 3, 4, 5, 6, 7, 8		
Cataracts	3, 7		1, 2, 4, 5, 6, 8
Skin Abnormalities	1, 2, 3, 4, 5, 6, 7, 8		

^aNumbers shown identify individual dogs (refer Table 1)

^bWater intake was accurately measured for only two of eight dogs and was estimated in remaining dogs

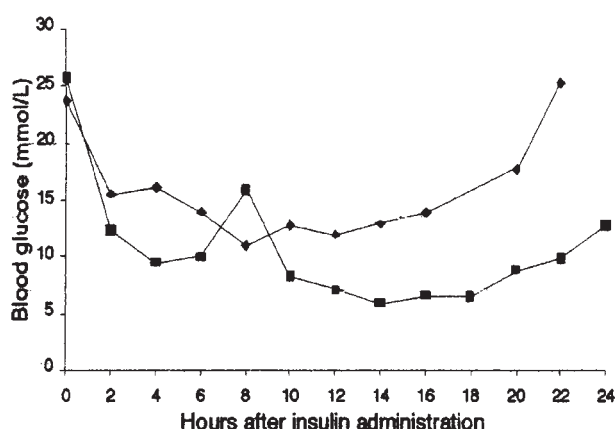


Figure 1. Blood glucose curves for two dogs with naturally occurring diabetes mellitus showing greater than or equal to 20 h blood glucose control: (♦) dog 1, (■) dog 2.

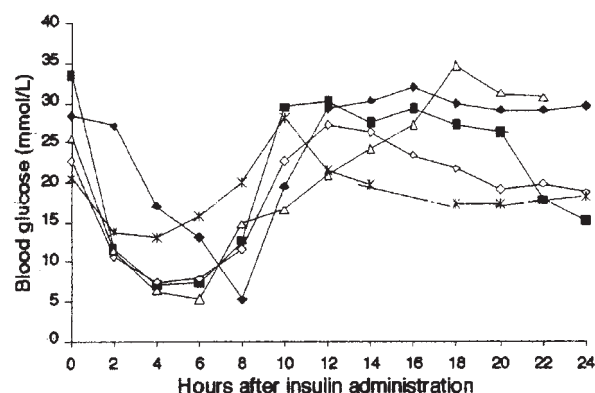


Figure 2. Blood glucose curves for five dogs with naturally occurring diabetes mellitus showing partial (less than 20 h) blood glucose control. (♦) dog 3, (■) dog 4, (Δ) dog 5, (◇) dog 6, (*) dog 7.

period (range 19.5 to 28.7; mean 23.7 mmol/L). This dog displayed restlessness and anxiety during hospitalisation and developed anorexia and haemorrhagic diarrhoea that resolved within 24 h after discharge from hospital.

Discussion

Results of this study suggest that once daily Caninsulin administration did not maintain glycaemic control for longer than 13 h in five of eight dogs, although acceptable blood glucose concentrations were maintained for 22 h and greater than 24 h in dog 1 and dog 2 respectively. Although blood glucose concentrations for dog 1 were not below 13 mmol/L for greater than 20 h, the dog was still included in group 1. The time of peak action of insulin in this dog was 8 to 12 h following administration, which is appropriate for a once daily administered insulin. However the lowest blood glucose (glucose nadir) of 11 mmol/L was higher than the ideal nadir of 5.5 to 7.0 mmol/L. Hence, by simply increasing the insulin dose, the expected effect would be to shift the blood glucose curve downwards. The result would be that the blood glucose values would fall within the ideal range for a diabetic dog for greater than 20 hours. In one other dog that did not appear to respond to the administered insulin, it is possible that stress and / or excitement and the consequent release of counter-regulatory hormones antagonised the effects of insulin. Unfortunately repeat testing was considered unlikely to yield more reliable results and was also not considered to be in the animal's welfare.

It was of interest that the results of 24 h blood glucose monitoring could not be reliably predicted from the owners' accounts of persistence or absence of clinical signs of diabetes mellitus. This suggests that more reliable information would be obtained if owners accurately measured water intake during a 24 h period rather than making a subjective assessment. Accurate monitoring of the dog's weight may also be helpful. In addition, it suggests that a spectrum of 'diabetic control' exists and varies with the owner's and the primary care veterinarian's expectations of what may be achieved. All owners agreed that their dogs' clinical signs had improved since initiating insulin therapy. The degree to which these owners were satisfied by the

clinical improvement may have varied from that of another group of owners. In addition, assessment by veterinary practitioners had indicated that some degree of control had been achieved. However, without 24 h monitoring of the blood glucose curve, an accurate assessment of the duration of activity of an insulin administered on a once daily basis cannot be made.

Caninsulin is the only insulin registered for veterinary use in Australia. However, there are several other insulin zinc suspensions for human use that have been used extensively in dogs and cats. Several studies have examined the pharmacodynamics (onset of activity and duration of activity) of these human preparations in the dog to determine the appropriate frequency of administration in this species. Lente insulin (Novo Nordisk) is composed of bovine insulin and presented as a 100 IU/mL formulation. Its duration of activity in dogs has been stated as being 8^{3,7} to 14⁸ or 24 h.^{3,7} As it is composed of beef insulin, it would be expected to have longer activity than its more soluble porcine equivalents.^{4,9,10} Monotard is another commonly used insulin zinc suspension. Monotard HM (Novo Nordisk) is composed of human insulin, derived through genetic engineering. In the past, Monotard MC (Novo Nordisk, also known as IZS-P) was manufactured from highly purified porcine insulin⁴ and was composed of 30 % amorphous and 70 % crystalline insulin. Duration of activity of Monotard MC in dogs was reported as 14 to 16 h¹¹ with relatively predictable peak activity at 4 to 8 h.⁴ It was, therefore, recommended that Monotard MC be given twice daily.⁴ Human studies have demonstrated that porcine and human insulin have similar kinetics of activity in vivo.¹² With discontinuation of Monotard MC production and introduction of Monotard HM, it became widespread practice to substitute Monotard MC with Monotard HM at equivalent dose rates and frequency of administration.

Although Caninsulin would appear similar to Monotard HM and Monotard MC, consideration of the physiochemical nature of the individual preparations is required before pharmacodynamics can be compared. Formulations of insulin zinc suspensions can potentially be varied in a number of ways. For example, variations in zinc concentration, crystal size and

shape, pH, temperature during manufacture, presence of halides or other divalent cations and species of origin of insulin can all influence pharmacodynamics.⁹ Information on how different manufacturers vary their insulin formulations is generally not available. However, because of the numerous possibilities, some guidelines have been set by the British Pharmacopoeia to limit variation between insulins.¹³ The result is that all 30% amorphous and 70% crystalline zinc insulin suspensions of the same species of origin have similar properties and behave in predictable ways. Human and porcine insulin behave similarly, so that a 30% amorphous and 70% crystalline zinc insulin made from porcine or human insulins is expected to have similar kinetics of activity. Caninsulin, as a veterinary registered drug, does not have to comply with the British Pharmacopoeia. It has its own stringent manufacturing standards it must adhere to, but these are not readily available for review and, therefore, direct comparison of composition is not possible. Nevertheless, it would seem unlikely that the formulation guidelines for Caninsulin would differ much from the British Pharmacopoeia formulation guidelines.

Although not a difference in composition, Caninsulin is presented at 40 U/mL while Monotard contains 100 U/mL. While this may explain small differences in pharmacodynamics between the two, it is unlikely to result in substantial differences. In an unpublished study, comparison of Caninsulin and Monotard MC in three pancreatectomised dogs and two naturally occurring diabetics failed to show differences between the 40 U/mL formulation and an equivalent dose of 100 U/mL insulin (DB Church personal communication).

There are a number of limitations of this study, not least the small number of dogs included. However, it was intended as a pilot study and its preliminary results question the ability of Caninsulin to stabilise the majority of canine patients with diabetes mellitus on once daily administration. It may be that most dogs require twice daily administration. This should be further evaluated in a more comprehensive study.

Although criteria for patient selection included uncomplicated diabetes, one dog studied was subsequently found to have exocrine pancreatic insufficiency and hepatopathy. The owners had noted substantial weight loss over several months, but had considered this a sign of 'old age'. During hospitalisation, the dog also produced a large, pale stool. Investigators recommended a complete blood count, serum biochemical profile and trypsin-like immunoreactivity be performed by the referring veterinarian that led to the diagnosis of exocrine pancreatic insufficiency and hepatopathy. This dog was one of the four in Group 2. While hepatopathies may lead to altered glucose metabolism, this should be a general effect seen for the entire 24 h period. Hence, it would appear unlikely that a blood glucose curve showing initial blood glucose control that then failed to persist for more than 12 h would result. In addition, it is more likely that hepatopathy would result in prolonged insulin effects due to impaired insulin degradation¹⁴ rather than a shortened duration of action as seen here.

In addition, one dog was atopic and was on long-term low-dose corticosteroid administration. Although much has been written on the potential of corticosteroid administration to cause diabetes mellitus by producing peripheral insulin antagonism,¹⁵⁻¹⁷ one article concluded that anti-inflammatory doses of prednisolone given orally for 4 weeks probably do not alter insulin sensitivity or glucose tolerance in normal dogs.¹⁸ In any case, this atopic dog (dog 2) was one of the two animals that showed 24 h blood glucose control.

Finally, the study group is acknowledged as a biased sample. However, this bias is in favour of finding dogs that were stabilised on once daily Caninsulin, as only dogs that were receiving once daily Caninsulin and thought to be adequately stabilised were included.

This prospective study suggests that the duration of action of Caninsulin in the majority of dogs does not extend to 24 h and that twice daily dosing may be necessary in these animals for adequate glycaemic control. Caninsulin potentially has an advantage for use in canine diabetics due to lower antigenicity and ease of administration in small doses, but it may still benefit from twice daily administration. The approach to using Caninsulin, like other insulins, should rely on assessing its duration of activity in each dog before deciding whether once daily or twice daily administration is appropriate. A controlled study performed by independent investigators that further evaluated the duration of activity of Caninsulin in a large group of clinical cases of canine diabetes mellitus is indicated.

Acknowledgments

We thank the staff of the Western Animal Emergency Centre for assistance in collecting samples and Debbie Presidente for laboratory assistance. The study could not have been performed without the support of several Melbourne veterinary practices and the consent of the pet owners.

References

- Greco DS, Broussard JD, Peterson ME. Insulin therapy. *Vet Clin North Am Small Anim Pract* 1995;25:677-689.
- Feldman EC, Nelson RW. Diabetes mellitus. In: Feldman EC, Nelson RW, editors. *Canine and feline endocrinology and reproduction*. 2nd edn. Saunders, Philadelphia, 1996: 339-391.
- Hardy RM. Diabetes mellitus in the dog. In: *Small animal internal medicine*. University of Sydney Post Graduate Committee in Veterinary Science, Proceedings 1992, (187):177-209.
- Church DB. The blood glucose response to three prolonged duration insulins in canine diabetes mellitus. *J Small Anim Pract* 1981;22:301-310.
- Hoenig M, Ferguson DC. Diabetes mellitus in the dog and cat. *Companion Anim Pract* 1989;19:12-16.
- Lorenzen FH. The use of isophane insulin for the control of diabetes mellitus in dogs. *Acta Vet Scand* 1992; 33:219-227.
- Graham PA, Nash AS, McKellar QA. Pharmacokinetics of a porcine insulin zinc suspension in diabetic dogs. *J Small Anim Pract* 1997;38:434-438.
- Hill FWG. The control of diabetes mellitus in dogs. In: Grunsell GSG, Hill FWG, editors. *Veterinary annual*. 21st issue. Scientechica, Bristol, 1982:179-182.
- Skelbaek-Pedersen B, Langkjaer L, Damgaard U et al. In: Brange J, editor. *Galenics of insulin - the physico-chemical and pharmaceutical aspects of insulin and insulin preparations*. Springer-Verlag, Berlin, 1987:17-42.
- Wolfsheimer KJ. Insulin therapy in dogs and cats. *Canine Pract* 1991;16:6-12.
- Church DB. Canine diabetes mellitus: some therapeutic considerations. In: Grunsell GSG, Hill FWG, editors. *Veterinary annual*. Scientechica, 22nd issue. Bristol, 1982:235-240.
- Chisolm DJ, Kraegen EW, Hewett MJ, Lazarus L. Comparison of potency of porcine insulin and semisynthetic human insulin at 3 dose levels using the euglycaemic clamp. *Horm Metab Res* 1983;15:415-418.
- British Pharmacopoeia Commission. Insulin preparations. *British pharmacopoeia* 1993, volume II. 1993:961-965.
- Nelson RW. Use of insulin in small animal medicine. *J Am Vet Med Assoc* 1984;185:105-109.
- Campbell KL, Latimer KS. Transient diabetes mellitus associated with prednisone therapy in a dog. *J Am Vet Med Assoc* 1984;185:299-301.
- Edwards F. Transient diabetes mellitus and ketoacidosis in a dog. *J Am Vet Med Assoc* 1982;180:68-70.
- Jeffers JG, Shanley KJ, Schick RO. Diabetes mellitus induced in a dog after administration of corticosteroids and methylprednisolone pulse therapy. *J Am Vet Med Assoc* 1991;199:77-80.
- Moore GE, Hoenig M. Effects of orally administered prednisone on glucose tolerance and insulin secretion in clinically normal dogs. *Am J Vet Res* 1993; 54:126-129.

(Accepted for publication 24 October 1999)