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Efficacy of Protamine Zinc Recombinant Human Insulin for Controlling Hyperglycemia in Dogs with Diabetes Mellitus

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Background: Alternative insulin preparations are needed when NPH insulin is ineffective in diabetic dogs. This study evaluated the efficacy of recombinant human protamine zinc insulin (rhPZI) for treating diabetic dogs.

Hypothesis: rhPZI is effective for treating diabetic dogs.

Animals: Six newly diagnosed and 11 insulin-treated diabetic dogs.

Methods: Prospective clinical trial. Dogs were treated with rhPZI for 60 days. Control of glycemia was assessed on days 7, 14, 30, and 60 by evaluation of history, physical examination, body weight, serum fructosamine concentration, and blood glucose concentrations measured before and 2, 4, 6, 8, and 10 hours after rhPZI administration. Adjustments in dosage of rhPZI were made as needed to control glycemia.

Results: rhPZI administration resulted in a significant decrease in 10-hour mean blood glucose (MBG_{10h}; 299 ± 115 versus 457 ± 38 mg/dL, X ± SD, P = .0003) and serum fructosamine (478 ± 83 versus 557 ± 104 μmol/L, P = .006) concentration at day 60, compared with day 1, respectively. By day 60, polyuria and polydipsia had improved in 14, body weight was stable or increased in 16, MBG_{10h} had decreased in 16, and serum fructosamine concentration had decreased in 11 of 17 dogs, compared with day 1. Hypoglycemia (<80 mg/dL) was the only consistent adverse event.

Conclusions and Clinical Relevance: rhPZI is effective in diabetic dogs and can be considered as an alternative treatment in diabetic dogs that are poorly controlled using other insulin preparations.

Key words: Fructosamine; Glucose; Hypoglycemia; Pancreas.

Treatment of diabetes mellitus in dogs requires daily insulin administration to achieve control of hyperglycemia. Commonly used insulin preparations for diabetes in dogs include recombinant human neutral protamine Hagedorn (NPH) insulin and purified pork source lente insulin.[1,2] After FDA approval in 2004, purified pork source lente insulin became very popular for treating diabetes in dogs in the United States. The withdrawal of purified pork source lente insulin by the FDA in November 2009 left the veterinary community searching for an alternative insulin replacement. Beef/pork source and recombinant human NPH insulin have been used successfully to treat diabetes in dogs when administered twice daily.[2] In our experience, control of the diabetic state is difficult in some dogs because of short duration of NPH effect despite twice daily administration. Studies evaluating the efficacy of longer acting insulin preparations such as protamine zinc insulin (PZI), insulin glargine, and insulin detemir in diabetic dogs are lacking. In our experience, insulin detemir improved glycemic control in a small number of diabetic dogs poorly controlled on other insulin preparations but problems with hypoglycemia and potency of the insulin may limit its use in diabetic dogs.[3] Beef/pork source PZI was a longer acting insulin preparation commonly used in diabetic cats[4] before its withdrawal from the market in 2008. Preliminary information on the actions of beef/pork source PZI in healthy dogs[a] suggested that PZI was effective in decreasing blood glucose concentration and had a prolonged duration of effect, but the efficacy of beef/pork source PZI for managing diabetic dogs was not reported. One study[5] documented anti-insulin antibody production in a group of diabetic dogs treated with a beef PZI preparation, but information on diabetic regulation of the dogs was minimal. A recombinant human PZI preparation (rhPZI) was approved by the FDA in 2009 for managing diabetes in cats.[6] Because a recombinant human insulin analog preparation is a more attractive insulin option than a beef/pork source insulin preparation in dogs,[5] the recent availability of rhPZI may provide another viable insulin option for treating diabetic dogs. The purpose of this study was to evaluate the efficacy of rhPZI for controlling hyperglycemia in newly diagnosed and previously insulin-treated diabetic dogs.

Abbreviations:

rhPZI	recombinant human PZI preparation
PZI	protamine zinc insulin
NPH	recombinant human Neutral Protamine Hagedorn
MBG_{10h}	10-hour mean blood glucose concentration
LBG_{10h}	lowest blood glucose concentration during the 10-hour evaluation

Materials and Methods

One university teaching hospital and 1 referral practice participated in the study. The study was conducted with approval from the University of California Clinical Trial Review Board.

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Institutional Animal Care and Use Committee (IACUC) guidelines were followed for blood sampling, urine sampling, and imaging. Written owner consent was obtained before entry of each dog into the study.

Inclusion Criteria

All dogs were diagnosed with diabetes mellitus based on the presence of polyuria, polydipsia, polyphagia with or without weight loss, persistent fasting hyperglycemia (blood glucose concentration >250 mg/dL), and persistent glycosuria. Dogs could be newly diagnosed but naïve to treatment, previously diagnosed but considered poorly regulated on their current insulin treatment regimen, or previously diagnosed and considered well regulated on pork source lente insulin.[b] Dogs with a prior history of diabetic ketoacidosis could be enrolled once ketoacidosis had resolved using regular crystalline insulin.[c] Dogs could be thin, ideal body weight, or obese. Dogs had to complete the study, had to be treated with rhPZI[d] throughout the study, and had to be fed the same diet throughout the study to be considered for inclusion. Dogs previously treated with beef/pork source PZI[e] were excluded from the study. Dogs with suspected concurrent conditions causing insulin resistance such as hyperadrenocorticism or dogs with concurrent potentially life-threatening disease such as neoplasia or renal failure that decreased the likelihood of completing the study were not enrolled. Dogs treated with glucocorticoids within the past 30 days were not enrolled.

Study Design

This was a prospective clinical trial. The study was conducted over a period of 60 days. On day 1, a history was obtained, a physical examination performed, and blood and serum collected for evaluation of a CBC, serum biochemical analysis, canine pancreatic-specific lipase (Spec cPL), and serum fructosamine concentration. Urine was collected by antepubic cystocentesis for urinalysis and bacterial culture and an abdominal ultrasound examination was performed. In newly diagnosed untreated diabetic dogs, a blood glucose concentration was determined twice during a 2-hour interval to confirm hyperglycemia. Treatment with rhPZI was started on day 1 and blood glucose concentrations were determined before and 2, 4, 6, 8, and 10 hours after insulin administration. The initial recommended insulin dosage range was 0.25–0.5 U/kg administered every 12 hours. The veterinarian determined the insulin dose after reviewing the history and results of the physical examination, body weight, and blood glucose concentrations measured on day 0. Diet at entry into the study was determined by the owner and veterinarian; the diet and quantity consumed were kept constant throughout the study. Owners were asked to divide the daily caloric intake in half and feed one-half each time rhPZI was administered. A dosing diary was provided to each owner on days 1,7, 14, and 30 to record date, time, and dose of rhPZI. Adverse events and health related problems such as lethargy, weakness, inappetence, vomiting, and inappropriate urination also were recorded.

Dogs were evaluated on days 7, 14, 30, and 60 after entry into the study. On the morning of each scheduled in-hospital evaluation, owners were asked to feed their dog within 30 minutes before arrival at the hospital. Blood for glucose measurement was obtained immediately upon arrival, and rhPZI administered immediately after blood collection. The dosing diary was reviewed, and a complete history was obtained, including the owner's perception of changes (increased, decreased, or no change) in their dog's frequency of urination, water consumption, and appetite compared with day 0, as well as any adverse events they had observed. At each visit, a complete physical examination was performed and abnormalities and body weight recorded. Blood glucose concentrations were determined before insulin administration, and 2, 4, 6, 8, and 10 hours after administration of rhPZI. On days 30 and 60, blood and urine were obtained for evaluation of a CBC, serum biochemical analysis, serum fructosamine concentration, and urinalysis. In addition, on day 60, a spec cPL and bacterial urine culture were performed. The dosing diary was collected and reviewed at each visit and a new dosing diary provided to the owner. Adjustments in insulin dose were made based on the owner's perception of presence and severity of clinical signs of diabetes, results of the physical examination, change in body weight, and blood glucose measurements, with the intent to maintain the blood glucose concentration between 80 and 300 mg/dL and the blood glucose nadir between 80 and 150 mg/dL. Changes in the insulin dose ultimately were at the veterinarian's discretion.

Analytical Methods

Blood glucose concentrations were determined by use of a handheld portable blood glucose monitor[f] previously evaluated for use in diabetic dogs.[7] Detectable blood glucose concentrations ranged from 20 to 500 mg/dL. Blood glucose concentrations <20 mg/dL and >500 mg/dL registered as "LO" and "HI" on the portable blood glucose monitor and were arbitrarily given the value of 19 and 501 mg/dL, respectively. Quality control procedures recommended by the manufacturer were followed to assure accurate functioning of the monitor. Serum fructosamine concentration was determined by means of the nitroblue tetrazolium reduction method.[8] Glycosuria was confirmed with urine reagent test strips.[g] Other analyses carried out on blood, urine, and serum samples were performed at central reference laboratories.[h,i] Serum was evaluated using a commercial assay for canine Spec cPL[i,9]

Assessment of Efficacy

Blood parameters used to assess control of glycemia included the mean of the 6 blood glucose concentrations measured before and every 2 hours for a 10-hour period after administration of rhPZI (10-hour mean blood glucose concentration [MBG_{10h}]), lowest blood glucose concentration during the 10-hour evaluation (LBG_{10h}), time from rhPZI administration to the LBG_{10h}, and serum fructosamine concentration. Control of glycemia was classified as good, moderate, or poor according to the following criteria: MBG_{10h} concentrations < 250 mg/dL were considered good, 250–300 mg/dL moderate, and >300 mg/dL poor; LBG_{10h} concentrations between 80 and 150 mg/dL were considered good, 150–200 mg/dL moderate, and >200 mg/dL poor; and serum fructosamine concentrations <450 μmol/L were considered good, 450–500 μmol/L moderate, and >500 μmol/L poor. If the blood glucose concentration was <80 mg/dL at any time during the 10-hour blood glucose evaluation period, the dog was classified as having experienced an insulin overdosage. Clinical parameters used to assess efficacy of treatment included owner's subjective assessment of polyuria and polydipsia, findings on physical examination, and stability of body weight. For purposes of this study, treatment with rhPZI was considered successful if the owner observed improvement of polyuria and polydipsia, the veterinarian believed the dog was in good body condition on physical examination, body weight was stable or increasing, and there was improvement in the MBG_{10h}, serum fructosamine concentration, or both when day 1 and day 60 results were compared.

Data Analysis

Summary statistics were calculated for all data points showing the number of observations, mean, standard deviation, minimum,

and maximum. All analyses were performed by a statistical software program.[j] For analyses conducted between days 1 and 60, continuous endpoints (insulin dosage, MBG_{10h}, LBG_{10h}, and serum fructosamine concentration) were analyzed by 2-way repeated measures analysis of variance (ANOVA). *P*-values < .05 were considered statistically significant. Body weight was compared on day 0 and day 60 using a paired *t*-test.

Results

Twenty diabetic dogs met the inclusion criteria for entry into the study. Three of the dogs subsequently were removed from the study because of fractious behavior (1 dog at day 7), a diagnosis of hyperadrenocorticism (1 dog at day 7), and exocrine pancreatic insufficiency (1 dog at day 30). Seventeen diabetic dogs completed the study. Breeds included Labrador Retriever (n = 4), West Highland White Terrier (2), and 1 each of Chow Chow, Corgi, Beagle, Bichon Frise, Shih Tzu, Miniature Poodle, Chihuahua, Yorkshire Terrier, Blue Heeler, Brittany, and Cairn Terrier. Six dogs were castrated males and 11 were spayed females. Age ranged from 4 to 16 years (median, 10 years) and body weight ranged from 5.2 to 44.5 kg (mean, 17.5 kg). Six of 17 dogs were naïve to treatment and 11 dogs had been treated with either pork source lente[b] (9) or recombinant human NPH[k] (2) insulin at entry into the study. Diabetic control was considered good and poor in 3 and 8 insulin-treated dogs, respectively, based on the history and results of physical examination and serum fructosamine measurements at entry into the study. Dogs were fed a variety of diets including a mixture of commercial canned and dry (n = 9), commercial dry maintenance (4), high fiber (2), low fat (1), and home cooked (1) diets.

Concurrent diseases identified in the 17 dogs after review of their histories and results of physical examinations, CBC, serum biochemical analyses, urinalyses, and urine cultures at entry into the study included diabetic ketosis (n = 6), atopic dermatitis (4), bacterial cystitis (2), otitis externa (1), and keratoconjunctivitis sicca (1). Diabetic ketosis resolved in all dogs after initiating rhPZI treatment. Atopic dermatitis was managed with flea control and diet; none of the dogs were treated with glucocorticoids. Dogs with bacterial cystitis were treated with antimicrobials; urine cultures were negative for bacterial growth after 2 weeks of treatment. Serum Spec cPL results were within the reference interval in 12 dogs and increased in 5 dogs (282, 557, 642, 915, and 1000 μg/L; reference interval, 0–200 μg/L)[9] at entry into the study. None of the 17 dogs had clinical signs suggestive of pancreatitis at any time during the study.

Abnormalities identified on abdominal ultrasound examination were consistent with diabetes mellitus and included hepatomegaly (n = 9) and a hyperechoic liver (6). Ultrasonographic abnormalities suggestive of pancreatitis were not identified in any dog, including the 5 dogs with increased serum Spec cPL. At day 60, serum Spec cPL was within the reference interval in 16 dogs and was still increased (354 μg/L) in 1 dog that was healthy and had good diabetic control.

Abnormalities identified on the CBC, serum biochemical analyses, and urinalyses at entry into the study were consistent with diabetes mellitus and included, in addition to hyperglycemia, glucosuria, and ketonuria, increased serum alanine aminotransferase (6 dogs; median, 180 IU/L; range, 148–303 IU/L; reference interval, 5–107 IU/L) and alkaline phosphatase (14 dogs; median, 543 IU/L; range, 194–2192 IU/L; reference interval, 10–170 IU/L) activity and serum cholesterol (7 dogs; median, 401 mg/dL; range, 367–509 mg/dL; reference interval, 112–367 mg/dL) concentration. On day 60, serum alanine aminotransferase (median, 153 IU/L; range 110–595 IU/L) and alkaline phosphatase (median, 367 IU/L; range, 176–1415 IU/L) activity were increased in 5 and 10 dogs, respectively, and serum cholesterol (median, 427 mg/dL; range, 375–468 mg/dL) concentration was increased in 5 dogs. None of the abnormalities identified on blood and urine tests had any predictive value regarding response to rhPZI or classification of glycemic control on day 60.

There was a significant increase in the median dosage of rhPZI ($P = .0001$) and a corresponding significant decrease in MBG_{10h} ($P = .0003$) and serum fructosamine concentration ($P = .0061$) by day 60, compared with day 1 (Table 1; Figs 1–2). There was no difference in median insulin dosage ($P = .76$), MBG_{10h} ($P = .57$), LBG_{10h} ($P = .93$), or serum fructosamine concentration ($P = .70$) in newly diagnosed untreated diabetic dogs, compared with previously insulin-treated dogs on days 1 and 60.

Results of the LBG_{10h} were between 80 and 150 m/dL in 4 dogs, >200 mg/dL in 11 dogs, and <80 mg/dL in 2 dogs at day 60 of the study. There was no significant change in mean LBG_{10h} ($P = .058$) by day 60, compared with day 1. LBG_{10h} results varied unpredictably for individual dogs during the study. Only 2 dogs had LBG_{10h} results consistent with poor control at every evaluation day, and no dog had results consistent with good or moderate control at every evaluation day.

The LBG_{10h} was > 200 mg/dL in 42 of 68 blood glucose curves evaluated during the study and was < 200 mg/dL in 15 of 17 dogs at 1 or more of the 4 evaluation days. The time interval from rhPZI administration to LBG_{10h} also was variable and often this variability occurred at the beginning (12 hours after rhPZI administration the previous evening) or at the end of the 10-hour blood sampling interval. The median interval from rhPZI administration to LBG_{10h} was 8 or 10 hours for each of the 4 evaluation days in the study (Table 1). The LBG_{10h} occurred at the start or the end of the 10-hour blood glucose curve in 54% of 68 blood glucose curves evaluated during the study and occurred on 1 or more of the 4 evaluation days in 16 of 17 dogs. When comparing consistency of the timing of the LBG_{10h} among blood glucose curves obtained for each diabetic dog, the LBG_{10h} occurred at 2, 3, and 4 different times of the day during the study in 6, 9, and 2 dogs, respectively.

Results of serum fructosamine concentration were consistent with good glycemic control throughout the

study (n = 2) or had improved from moderate or poor to good control (5) by day 60 in 7 dogs. Glycemic control remained poor in 4 dogs for the duration of the study based on serum fructosamine concentration. Discordant interpretation of results of MBG_{10h} and serum fructosamine concentration occurred in 6 dogs at day 60. In 4 of the 6 dogs, serum fructosamine concentration decreased (median, 145 μmol/L; range, 72–283 μmol/L) and glycemic control improved from moderate (2) or poor (2) to good control by day 60, but glycemic control based on results of MBG_{10h} was poor despite a decrease in serum fructosamine concentration. In 2 of the 6 dogs, serum fructosamine concentration increased 68 and 102 μmol/L, glycemic control based on serum fructosamine concentration changed from good to moderate and good to poor, but MBG_{10h} was 208 and 82 mg/dL, respectively, at day 60. Both dogs were being treated with porcine lente insulin at entry into the study.

At completion of the study, 14 (82%) of 17 owners had observed improvement in the severity of their dog's polyuria, polydipsia, and polyphagia. All 14 dogs had stable or increased body weight and 10 of 14 dogs had improvement in MBG_{10h} and serum fructosamine concentration. Owners of the remaining 3 dogs believed their dogs' diabetes was well controlled at entry into the study and did not report any change in water consumption, frequency of urination, or appetite during the study. Two of 3 dogs had stable or increased body weight, and MBG_{10h} and serum fructosamine concentration improved in 2 of the 3 dogs. Mean body weight was similar (P = .33) at day 1 and day 60, body weight increased or remained stable in 16 (94%) dogs during the study, and all dogs were determined to be in good body condition based on the veterinarian's clinical assessment at day 60. In 1 dog, body weight decreased from 38 to 34 kg and diabetic control improved from poor to moderate control during the study.

Hypoglycemia (blood glucose <80 mg/dL) was the only consistent adverse event identified with rhPZI. Asymptomatic hypoglycemia was identified in 4 of 68 blood glucose curves in 2 diabetic dogs. Clinical signs consistent with hypoglycemia, including lethargy, exercise intolerance, weakness, and abnormal gait, were reported by owners of 3 dogs. In 2 of the dogs, hypoglycemia was confirmed after taking the dog to the veterinary hospital shortly after observing clinical signs. Clinical signs resolved with feeding and a

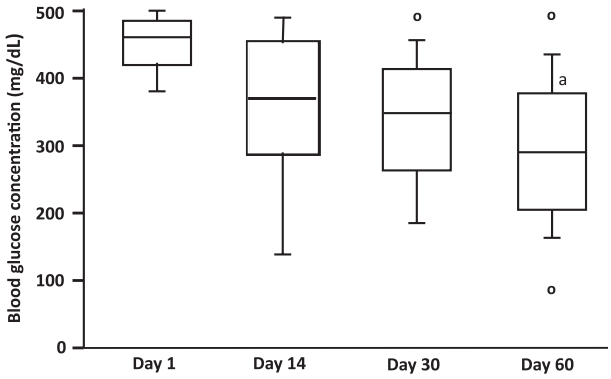

Fig 1. Box plots of the 10-hour mean blood glucose concentrations in 17 dogs with naturally acquired diabetes mellitus treated by administration of various doses of recombinant human protamine zinc insulin twice daily for 60 days. The 10-hour mean blood glucose concentration is the mean of the 6 blood glucose concentrations measured during a 10-hour period. The horizontal line represents the median, the box represents the interquartile range (ie, 25–75%), the T-bars represent the main body of data, and the circles represent the outliers. (a) $P = .0003$, compared with results on day 1.

Table 1. Variables (mean ± SD [range] or median [range]) used to assess control of glycemia in 17 dogs with naturally acquired diabetes mellitus treated with recombinant human protamine zinc insulin twice daily for 60 days.

Variables	Day 1	Day 7	Day 14	Day 30	Day 60
Median insulin dosage (U/kg/injection)	0.6 (0.2–0.8)	0.6 (0.2–0.8)	0.7 (0.2–1.1)	0.8 (0.3–1.2)	1.0[a] (0.4–1.5)
Body weight (kg)	17.5 ± 13 (5.2–44.5)	17.5 ± 12 (5.2–43.9)	17.5 ±12 (5.3–43.9)	17.8 ± 12 (5.3–44)	17.8 ± 11.8 (5.3–44.3)
10-hour mean blood glucose concentration (mg/dL)[d]	457 ± 3 (393–500)	457 ± 3 (382–500)	378 ±140 (107–501)	341 ± 93 (181–481)	299 ± 115[b] (82–442)
Median lowest blood glucose (mg/dL)	397 (254–491)	377 (79–467)	279 (81–484)	243 (97–438)	204 (56–478)
Time of the lowest blood glucose (h)[e]	10 (2–12)	10 (2–12)	10 (6–12)	8 (2–12)	10 (2–12)
Serum fructosamine (μmol/L)	557 ± 104 (415–748)	NA	NA	512 ± 98 (411–725)	478 ± 83[c] (362–660)

[a] $P = .0001$ significantly different from the value obtained on day 1.
[b] $P = .0003$ significantly different from the value obtained on day 1.
[c] $P = .0061$ significantly different from the value obtained on day 1.
[d] Values were calculated from blood glucose concentrations determined prior to and 2, 4, 6, 8, and 10 hours after administration of insulin.
[e] LBG at 12 hours after insulin injection was at the start of the blood glucose curve, prior to the next dose of insulin. NA, not available.

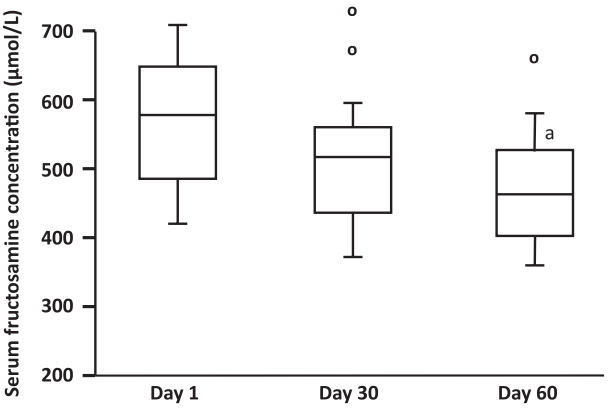

Fig 2. Box plots of serum fructosamine concentrations in 17 dogs with naturally acquired diabetes mellitus treated by administration of various dosages of recombinant human protamine zinc insulin twice daily for 60 days. The horizontal line represents the median, the box represents the interquartile range (ie, 25–75%), the T-bars represent the main body of data, and circles represent the outliers. (a) Significant $P = .006$, compared with results on day 1.

reduction in the dose of rhPZI in the 3 dogs. Reactions at the site of rhPZI administration were not reported by any owner or veterinarian.

Discussion

Administration of rhPZI was effective in improving or maintaining control of glycemia in the majority of diabetic dogs enrolled in the study. Owner compliance was excellent and all dogs remained healthy throughout the study. For most dogs, body weight remained stable or increased, owners reported improvement in clinical signs, and MBG_{10h} and serum fructosamine concentrations decreased significantly during the study. The only adverse event associated with rhPZI administration was hypoglycemia.

Prolonged duration of rhPZI effect was 1 problem identified in the study. The lowest blood glucose concentration occurred at the beginning of the blood glucose curve 12 hours after the evening administration of rhPZI by the owner or at the last blood sampling time 10 hours after the morning administration of rhPZI administration in 54% of the blood glucose curves evaluated and was identified on 1 or more blood glucose curves obtained in 16 of 17 dogs. The actual blood glucose nadir and time from rhPZI administration to the blood glucose nadir could not be identified in these blood glucose curves. Regardless, results of the study suggest a 10-hour or longer duration of effect of rhPZI may be common in diabetic dogs. Problems with hypoglycemia and initiation of the Somogyi response may occur when an insulin preparation with a duration of effect longer than 12 hours is administered every 12 hours.[10] Although absorption kinetics were not performed, day-to-day variability in the pharmacokinetics of rhPZI in individual dogs would result in variable blood insulin and glucose concentrations from day to day. Prolonged duration of rhPZI effect combined with the effects of the Somogyi response and pharmacokinetic variability of rhPZI could explain, in part, the inconsistency in blood glucose results that occurred among evaluation days in individual dogs. This inconsistency made interpretation of blood glucose curves difficult in some dogs and created a bias toward reliance on history, findings on physical examination, stability of body weight, and results of serum fructosamine concentrations when assessing control of glycemia. Efficacy of once daily treatment was not evaluated in the study. However, results suggest once daily administration of rhPZI may be effective in maintaining control of glycemia in some diabetic dogs and should be considered in those dogs where the blood glucose nadir consistently occurs 12 hours or longer after administration of rhPZI, especially if hypoglycemia, the Somogyi response and poor diabetic control become recurring problems.[10,11]

The starting dosage of rhPZI insulin increased as the study progressed and the median insulin dosage at day 60 was higher than anticipated based on our experience with other insulin preparations in diabetic dogs. Control of glycemia usually can be attained with NPH or lente insulin dosages ranging from 0.25 to 1.0 U/kg (median, 0.5 U/kg) in diabetic dogs,[2,12–14] although the upper range for porcine lente insulin was 1.4 U/kg in 1 study.[1] Initially, the starting dosage of rhPZI insulin in our study was kept conservative (approximately 0.25 U/kg) to avoid hypoglycemia. As the study progressed, it became evident 0.25 U/kg was ineffective as a starting dosage and by the end of the study we were routinely starting dogs at approximately 0.5 U/kg. At the end of the study, the median rhPZI dosage was 0.9 U/kg, with a range of 0.4–1.5 U/kg. The starting dosage of insulin did not predict classification of glycemic control at day 60.

The relatively higher than anticipated rhPZI dosage requirements in the study may be due, in part, to individual dog variability in responsiveness to rhPZI and to use of a longer acting basal insulin preparation rather than intermediate-acting insulins (NPH, lente) commonly used in diabetic dogs. Basal insulin preparations typically have a slower rate of absorption and lower peak blood insulin concentration, compared with intermediate-acting insulins.[15] These properties may result in higher rhPZI dosage requirements to attain comparable glycemic control versus NPH or lente insulin. The longer duration of rhPZI effect in some diabetic dogs also may have inadvertently affected insulin dosage requirements secondary to initiation of the Somogyi response.

Concurrent insulin resistance frequently affects insulin dosage requirements.[16,17] Four dogs remained poorly controlled throughout the study based on results of MBG_{10h} and serum fructosamine concentrations, suggesting the possibility of insulin resistance in these dogs. However, an identifiable disorder causing insulin resistance was not evident in any dog enrolled in the study, including the 4 poorly controlled diabetic dogs. Abnormalities identified on physical examination

and results of diagnostic tests performed at entry and at the end of the study were typical for diabetic dogs and most abnormalities identified at entry had improved or resolved by the end of the study. Although serum Spec cPL results were increased in 5 dogs at entry into the study, none of these dogs had clinical signs, laboratory abnormalities, or abdominal ultrasound findings suggestive of pancreatitis, the dogs remained healthy throughout the study, and serum Spec cPL results were within the reference range in 4 dogs and had decreased substantially in 1 dog at the end of the study. None of the 5 dogs were considered poorly controlled at the end of the study.

Although there was a significant decrease in MBG_{10h} and serum fructosamine concentration by day 60 of the study, MBG_{10h} and serum fructosamine results were both consistent with good glycemic control in only 7 dogs at day 60. Discordant interpretation of status of glycemic control based on MBG_{10h} and serum fructosamine concentration occurred in 6 dogs at day 60. The Somogyi response was suspected in 1 of 6 dogs that had a MBG_{10h} of 82 mg/dL and serum fructosamine concentration of 538 μmol/L. In 1 dog, serum fructosamine concentration decreased 161 μmol/L by day 60 but was still greater than 500 μmol/L despite a MBG_{10h} of 282 mg/dL. In 4 of 6 dogs, serum fructosamine concentration decreased 72 to 283 μmol/L by day 60 and was < 450 μmol/L in all dogs despite MBG_{10h} remaining > 300 mg/dL. Presumably, blood glucose results obtained on the day of the blood glucose curve were not representative of blood glucose concentrations during the prior 2–3 weeks.

Interestingly, most owners were happy with the clinical response of their dog to rhPZI. Fourteen of 17 owners stated improvement in severity of their dog's polyuria and polydipsia at day 60 regardless of blood glucose and serum fructosamine results. Sixteen and 14 of 17 dogs, including 3 dogs that remained poorly controlled throughout the study, had a decrease in blood glucose and serum fructosamine concentrations, respectively, by day 60, a decrease that may explain owner perception of improvement in severity of polyuria and polydipsia in their dog. Conflicting results of the parameters used to assess control of glycemia (eg, results of the history and serum fructosamine concentration suggesting good and poor glycemic control, respectively) were evident in this study and emphasize the importance of relying on all available information when assessing glycemic control in diabetic dogs.

Although type of diet may affect control of glycemia in diabetic dogs,[18] feeding the same diet to all dogs was not attempted in this study because of owner reluctance to change the diet and concerns regarding owner compliance if consumption of a new diet designed for diabetic dogs became problematic during the study. Dogs consumed a variety of diets and only 2 dogs were fed a fiber-containing diet designed for diabetes mellitus. Each dog was fed the same diet throughout the study, thereby minimizing diet as a possible factor for improved control of glycemia in any given dog. Because a variety of diets were fed to the dogs, the impact of any specific diet on response of dogs to treatment with rhPZI could not be determined.

Amount and timing of exercise and time of day when insulin was administered also varied among dogs but was kept consistent for each dog throughout the study, thereby minimizing changes in daily schedule and exercise as possible factors for improved control of glycemia in any given dog. The frequency of evaluations and timing of blood glucose measurements during the blood glucose curve were designed to provide the information needed to assess the efficacy of rhPZI while minimizing demands on owners and behavioral changes in dogs. Although development of stress hyperglycemia cannot be ruled out, none of the dogs developed aggression, fear, or an uncooperative behavior on any evaluation day during the study.

In conclusion, results of this preliminary study suggest that rhPZI is effective for controlling hyperglycemia in newly diagnosed and previously insulin-treated diabetic dogs. Inconsistency in blood glucose results in individual diabetic dogs was common, may be a reflection of prolonged duration of rhPZI effect, day-to-day pharmacokinetic variability of rhPZI in individual dogs, or both, and supports reliance on other indicators such as history, physical examination, stability of body weight, and serum fructosamine concentrations when assessing control of glycemia. Based on the results of this study, use of rhPZI at a starting dose of 0.5 U/kg q12h should be considered in diabetic dogs that are poorly controlled because of short duration of NPH or lente insulin effect. Prolonged duration of rhPZI effect may result in problems with hypoglycemia and induction of the Somogyi response when rhPZI is administered twice a day in some diabetic dogs.

Footnotes

[a] Stenner VJ, Fleeman LM, Rand JS. Comparison of the pharmacodynamics and pharmacokinetics of subcutaneous glargine, protamine zinc, and lente insulin preparations in healthy dogs. J Vet Intern Med 2004;18:444 (abstract)
[b] Vetsulin, Intervet-Schering Plough, Summit, NJ
[c] Humulin-R, Eli Lilly and Co, Indianapolis, IN
[d] ProZinc, Boehringer-Ingelheim Vetmedica, St Joseph, MO
[e] PZI-VET, IDEXX Pharmaceuticals, Inc., Greensboro, NC
[f] Alpha Trak, Abbott Laboratories, Abbott Park, IL
[g] Diastix, Bayer, Elkhart, IN
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[i] IDEXX Laboratories, Westbrook, ME
[j] Stata 10.1/IC, StataCorp LP, College Station, TX
[k] Humulin-N, Eli Lilly and Co

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