Comparison of the Soluble Basal Insulin Analog Insulin Detemir With NPH Insulin

A randomized open crossover trial in type 1 diabetic subjects on basal-bolus therapy

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OBJECTIVE — Insulin detemir (NN304) is a soluble basal insulin analog developed to cover basal insulin requirements. This trial aimed to compare the blood glucose–lowering effect of insulin detemir with that of NPH insulin (NPH) and to evaluate the two treatments with regard to intrasubject variation of fasting blood glucose, incidence of hypoglycemia, dose requirements, and safety.

RESEARCH DESIGN AND METHODS — This multicenter open randomized crossover trial in 59 type 1 diabetic subjects comprised a 2-week run-in period on a basal-bolus regimen with NPH insulin once daily, followed by two 6-week periods of optimized basal-bolus therapy with either once-daily insulin detemir or NPH insulin.

RESULTS — The area under the curve, in the time interval 23:00–8:00, derived from 24-h serum glucose profiles, was not statistically significantly different for the two treatment periods (insulin detemir:NPH ratio 89.2:83.5, $P = 0.59$). The intrasubject variation in fasting blood glucose during the last 4 days of treatment was lower for insulin detemir compared with NPH ($P < 0.001$). Mean dose requirements of insulin detemir were 2.35 times higher (95% CI 2.22–2.48) compared with NPH. During the last week of treatment, fewer subjects experienced hypoglycemic episodes on insulin detemir (60%) compared with NPH treatment (77%) ($P = 0.049$).

CONCLUSIONS — Insulin detemir was as effective as NPH in maintaining glycemic control when administered at a higher molar dose. The results indicate that insulin detemir may provide more predictable fasting blood glucose with lower intrasubject variation and reduced risk of hypoglycemia compared with NPH.

Diabetes Care 24:296–301, 2001

Tight metabolic control is important to prevent and delay the development of late complications in subjects with type 1 diabetes (1,2). However, one of the barriers for optimal control is that low and relatively constant basal insulin levels between meals and at night are difficult to obtain with the basal insulin formulations currently available, such as NPH, which is associated with a high day-to-day variation in insulin absorption of 20–30% (3). In addition, bedtime administration of NPH can lead to peak insulin levels 3–8 h after administration and, potentially, nocturnal hypoglycemia. As a consequence, insulin doses sufficient to maintain normal morning blood glucose are difficult to obtain because of the potential for hypoglycemia during the night. Moreover, the basal insulin preparations currently used are all formulated as insulin suspensions, and the insulin concentration may change considerably between each administration as a result of inadequate agitation and homogenization before injection (4,5).

Insulin detemir [Lys^{B29}(N^{ε}-tetradecanoyl) des(B30) human insulin] is a new soluble insulin analog developed to ensure appropriate basal insulin supply. This analog exists in the presence of zinc and phenol, like native insulins, predominantly in the hexameric state. The fatty acid sidechain contributes to provide aggregation of hexamers, which can contribute to delay hexamer dissociation and absorption (6). In the monomeric state, the 14-C fatty acid chain attached to position B29 binds to binding sites on albumin. Because only the free fraction of insulin detemir is biologically active, albumin binding and the ensuing slow dissociation of the analog from the albumin further prolong the blood glucose–lowering action (7,8). The soluble formulation ensures a homogenous concentration, with no need for agitation before administration.

Clinical trials in healthy subjects suggest that insulin detemir has a less-pronounced peak of action (9) and lower intrasubject variation in pharmacokinetic parameters compared with NPH (10). Thus, insulin detemir may provide more consistent insulin levels and more predictable glucose control than NPH because of lower absorption variability. The dose requirement of insulin detemir appears to be somewhat higher than that of NPH when compared on a molar basis (11).

The aim of this trial was to compare the blood glucose–lowering effect of insulin detemir with that of NPH in terms of metabolic control, intrasubject variation in fast-

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Received for publication 16 June 2000 and accepted in revised form 20 October 2000.

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Abbreviations: ANOVA, analysis of variance; AUC, area under the curve; ELISA, enzyme-linked immunosorbent assay; HSI, human soluble insulin.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

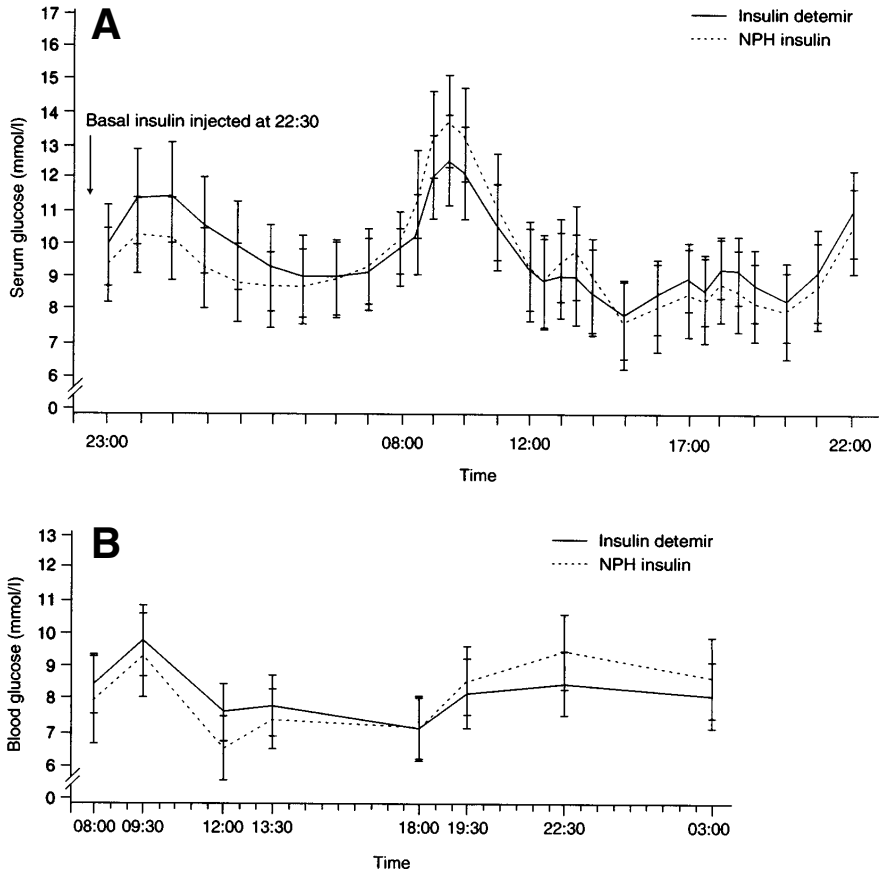

Figure 1—*Blood glucose profiles. A: Mean 24-h serum glucose profiles. B: Home-monitored mean eight-point blood glucose profiles during the last week of treatment. Values are means ± 2 × SEM.*

ing blood glucose, dose requirement, and safety in type 1 diabetic subjects treated with basal-bolus therapy.

RESEARCH DESIGN AND METHODS

Subjects

Subjects included in the trial had received once-daily (evening) NPH in combination with meal-related human soluble insulin (HSI) for at least 6 months and had documented type 1 diabetes for >2 years. All were Caucasians aged 18–55 years with a BMI <27.5 kg/m^2. Only subjects with HbA_{1c} ≤8.7%, glucagon-stimulated C-peptide ≤0.1 nmol/l or fasting C-peptide ≤0.04 nmol/l, and an NPH dosage <40 IU/day were included. Selection criteria excluded subjects with proliferative retinopathy; impaired hepatic function (aspartate aminotransferase and/or alkaline phosphatase at least twice the upper normal level); impaired renal function (creatinine ≥150 μmol/l); decompensated heart failure; unstable angina pectoris; myocardial infarction within the last year; hypertension (systolic and/or diastolic blood pressure ≥180 and 100 mmHg, respectively); hypoglycemic unawareness; recurrent major hypoglycemia; or allergy to insulin or any compositional component, as well as those who abused alcohol or narcotics; used systemic corticosteroids, β-blockers, or hormones within the past month; or were pregnant, breast-feeding, or using inadequate contraceptive measures. Subjects treated with other investigational products within the last 3 months or previously treated with insulin detemir were also excluded. The trial was carried out in accordance with Good Clinical Practice and the Declaration of Helsinki and was approved by the local ethics committees and health authorities according to local regulations. Informed written consent was obtained from each subject before trial entry.

Design

A total of seven investigation sites participated in the trial, which consisted of a run-in period of 2 weeks followed by two 6-week treatment periods. During the run-in phase, subjects administered an evening NPH injection in addition to HSI before each meal. Subjects were randomized symmetrically in blocks of four to a treatment sequence, starting with insulin detemir followed by NPH or vice versa.

Procedures

Subjects were instructed to administer either insulin detemir (100 U/ml, 100 U = 600 nmol) or NPH (Insulatard 100 IU/ml; Novo Nordisk A/S, Gentofte, Denmark) between 21:00 and 23:00 and HSI (Actrapid 100 IU/ml, Novo Nordisk A/S) 30 min before each main meal as subcutaneous injections. Meal-related insulin was administered in the abdominal region and basal insulin in the thigh. Subjects visited the investigation centers at 2-week intervals, and telephone contacts were made midway between visits for adjustment of insulin doses.

Subjects were informed of blood glucose targets (fasting, 4–7 mmol/l; postprandial, 5–9 mmol/l; 03:00, 4–7 mmol/l), instructed in the use of the NovoPen 1.5 device and in the calibration and use of glucose meters (One Touch II; LifeScan). Subjects recorded all data in diaries and were encouraged to measure blood glucose whenever symptoms of hypoglycemia were experienced. Hypoglycemia was defined as blood glucose <3 mmol/l with or without symptoms. Episodes were classified as minor if the subjects dealt with the episode themselves and as major if help from a third party or intravenous glucose or glucagon treatment was required.

Subjects performed two blood glucose profiles each week (before each meal, 90 min after each meal, at bedtime, and one profile at 03:00) and recorded administered insulin doses. In addition, subjects measured fasting blood glucose during the last 4 days of each treatment period. Subjects randomized to NPH continued on the NPH dose used at the end of the run-in period, whereas the starting dose of insulin detemir was twice the run-in dose of NPH on a molar basis. Subjects requiring doses of insulin detemir >40 U (240 nmol) administered the dose as two injections. If the insulin detemir dose was >120 U (720 nmol), the subject was withdrawn. Subjects were hospitalized overnight after the first insulin detemir dose, and blood glucose was monitored closely. The dose of insulin detemir was titrated according to blood glucose measurements in a stepwise fashion. During the first 2 weeks, meal-

related insulin doses remained unchanged, whereas basal insulin doses were optimized. During the following weeks, the distribution ratio of meal-related versus basal insulin doses was optimized. Subjects were asked to keep insulin doses unchanged during the last week of treatment.

On the last day of each treatment period, subjects were hospitalized at 21:00, and 24-h blood sampling for determination of serum concentrations of glucose, insulin detemir, and human insulin was performed. Food intake and treatment were kept as identical as possible between the two 24-h profiles. Subjects fasted from 22:00 until breakfast, and basal insulin was administered at 22:30. Blood samples were taken hourly from 22:00 and every 30 min for the first 2 h after each meal. Sampling was stopped at 22:00 on the next day, and subjects were discharged.

Methods

Serum glucose was determined by an enzymatic glucose oxidase method (12), whereas serum insulin was measured using an enzyme-linked immunosorbent assay (ELISA) (code no. K6219; Dako, Cambridgeshire, U.K.). Serum levels of total insulin determir were quantified by an ELISA method, developed by Novo Nordisk A/S (9). Fructosamine was measured at baseline and after each treatment period using a photometric method (13). Measurement of HbA_{1c} (normal range of assay, 4.5–5.7%) was performed using an immunological method (14).

Safety parameters included recording of adverse events, weight, physical examination, blood pressure, electrocardiogram, fundoscopy/fundophotography, laboratory tests for hematology, biochemistry, and lipids, and a urine screening.

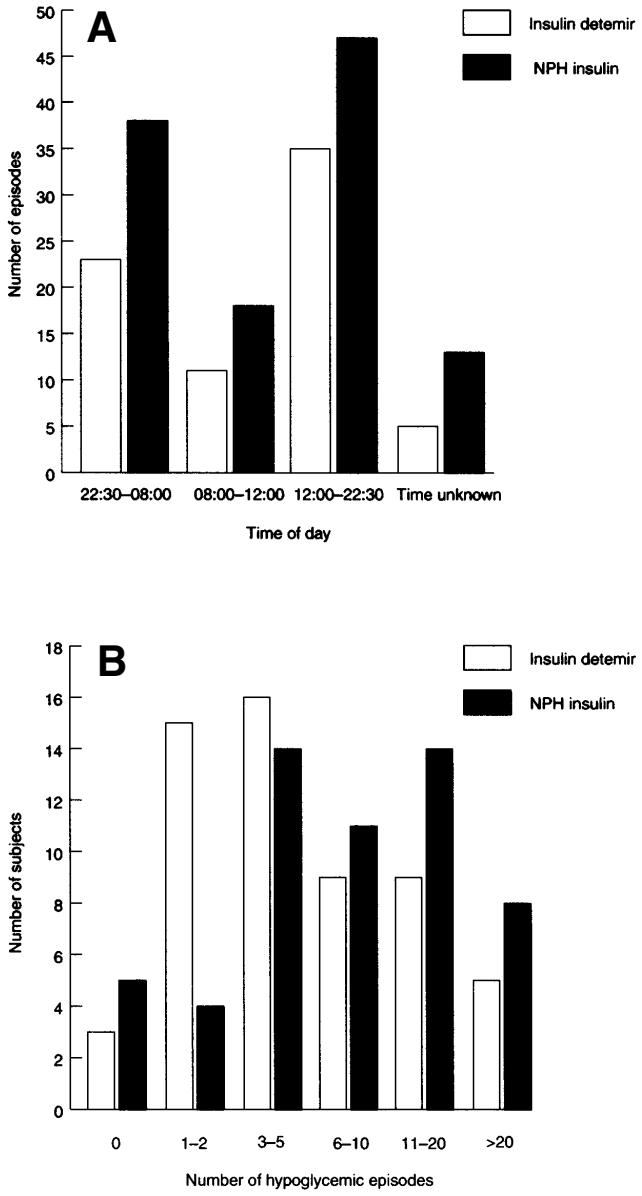

Figure 2—*Hypoglycemic episodes during the last week of treatment versus time of day* (A) *and distribution by treatment during the 6-week treatment period* (B).

Statistical analyses

To obtain a power of 80%, 49 subjects were needed to detect a difference in the primary end point of 10% with a significance level of 5%. The primary end point was the area under the serum glucose curve in the time interval from 23:00 to 08:00 [AUC(glu 23–08)]. The area under the curve (AUC) was calculated by the trapezoidal method and logarithmically transformed before analysis. The null hypothesis (equal AUC after treatment with insulin detemir and NPH) against the alternative hypothesis (AUC would be unequal) was analyzed by an analysis of variance (ANOVA) model including subject as a random effect and treatment period, center, and sex as fixed effects.

The hypothesis that time profiles for serum glucose concentration in the time interval 23:00–08:00 were parallel was evaluated by repeated measurements ANOVA with treatment, time, and treatment × time as fixed effects and subject, subject × treatment, subject × time, and error as random effects. The same method was used to examine the eight-point blood glucose profiles obtained during the last week of treatment. The intrasubject variation in fasting blood glucose was estimated in an ANOVA model with treatment, center, and sex as fixed effects and subject and error as variance components. The dose requirement of insulin detemir relative to NPH was estimated as a contrast in an ANOVA model of logarithmically transformed insulin doses, including treatment and treatment nested within subject as fixed effects. Analyses of all other end points were performed similarly to that of the primary end point (AUC). Fructosamine was analyzed with correction for baseline levels. The number of subjects having hypoglycemia during the last week of each treatment period was compared using the McNemar test. To minimize bias,

Table 1—*Hypoglycemic episodes by treatment and trial period*

	Subjects administered insulin detemir (*n* = 57)			Subjects administered NPH (*n* = 56)		
	Subjects with hypoglycemic episodes (*n*)	Proportion of subjects with an episode (%)	Hypoglycemic episodes (*n*)	Subjects with hypoglycemic episodes (*n*)	Proportion of subjects with an episode (%)	Hypoglycemic episodes (*n*)
Hypoglycemic episodes recorded during the last week of each treatment period						
Minor	34	59.6	74	42	75.0	113
Major	0	0	0	3	5.4	3
Total	34*	59.6	74	43	76.8	116
Hypoglycemic episodes recorded during the 6-week treatment period						
Minor	53	93.0	428	51	91.1	566
Major	4	7.0	4	7	12.5	11
Total	54	94.7	432	51	91.1	577

*$P = 0.049$.

personnel responsible for statistical analysis were kept blinded until the randomization code was broken.

RESULTS — A total of 59 type 1 diabetic subjects were randomized and exposed to trial products. Two subjects discontinued prematurely because of excessive C-peptide levels at trial inclusion, and the third was concerned over the volume of injected insulin detemir. In total, 56 subjects completed the trial and were included in the efficacy analyses. These subjects included 46 men and 10 women between 19 and 52 years of age (mean age 34.5) with a mean duration of diabetes of 14.8 years (range 2.6–47.8) and an average HbA_{1c} of 7.9% (range 5.7–8.7). The mean weight and BMI were 77.1 ± 8.9 kg and 23.8 ± 2.0 kg/m^2, respectively.

Serum glucose and glycemic control

The AUC(glu 23–08) was derived from the 24-h serum glucose profile obtained on the last day of each treatment period. The size of the AUC(glu 23–08) was comparable between the two periods ($P = 0.59$ with an insulin detemir/NPH AUC ratio of 1.04; 95% CI 0.90–1.21). During the next day from 08:00 to 12:00, the AUC ratio was 0.94, and maximal blood glucose levels were observed with both insulins in this period (Fig. 1*A*). Between 12:00 and 22:00, the ratio was 1.05. Mean total insulin detemir serum levels (albumin bound + free) reached 2,028 pmol/l ~5 h after injection and was ~10 times higher than the corresponding level of human insulin.

Analyses of the mean serum glucose profiles (Fig. 1*A*) showed that they were not parallel ($P = 0.02$). During the night, serum glucose with insulin detemir was higher than with NPH. The difference between the curves tended to increase until ~02:00, indicating a somewhat delayed onset of action for insulin detemir. The maximal glucose concentration and the range of the serum glucose concentrations were comparable between treatments.

Analysis of the eight-point blood glucose profiles obtained during the last week of each treatment period (Fig. 1*B*) showed that the AUC(glu 08–03) was not significantly different between the two treatments ($P = 0.32$) and that the profiles were parallel ($P = 0.41$). Mean blood glucose during this period was 8.08 ± 1.71 mmol/l for insulin detemir and 8.18 ± 1.76 mmol/l for NPH.

During the last 4 days of each treatment period, mean levels of home-monitored fasting blood glucose were 8.32 ± 3.58 and 8.75 ± 4.16 mmol/l for insulin detemir and NPH, respectively. A reduced intrasubject variability in fasting blood glucose was demonstrated for insulin detemir compared with NPH, with estimated coefficients of variation of 35 and 43%, respectively ($P < 0.001$).

At screening, the mean fructosamine level was 361 ± 49.4 µmol/l. The level decreased, and at the end of each treatment period, it did not differ significantly, being 345 ± 57.4 and 338 ± 56.3 µmol/l for insulin detemir and NPH, respectively ($P = 0.078$).

Insulin doses

The need for insulin detemir relative to NPH to obtain comparable blood glucose levels was evaluated during the last 8 days of treatment. The mean requirement for insulin detemir was 2.35 times higher than that for NPH (95% CI 2.22–2.48), ranging from 1.40 to 4 times the molar NPH dose. During this period, basal insulin use was relatively constant with an intrasubject coefficient of variation of 4.3%. A total of 36 subjects required an insulin detemir dose >40 U and applied two injections. While the mean daily NPH dose was relatively constant over time (0.28 IU/kg), the mean daily dose of insulin detemir increased the first 3–4 weeks during dose titration and remained stable over the rest of the treatment period (0.67 U/kg). The mean daily dose of meal-related insulin was ~0.45 IU/kg and remained constant over time regardless of treatment.

Hypoglycemic episodes

During the last week of treatment, when subjects were asked not to change their insulin doses, fewer subjects had hypoglycemic episodes on insulin detemir (60%) than on NPH (77%) ($P = 0.049$). The lower incidence on insulin detemir was observed regardless of the time of day (Fig. 2*A*), and, in contrast to the NPH treatment period, no major hypoglycemic episodes were recorded

with insulin detemir (Table 1). When the entire treatment period was compared, a similar proportion of subjects experienced hypoglycemic episodes (Table 1). However, the occurrence of clustered recurrent hypoglycemic episodes was slightly more common during NPH treatment (Fig. 2*B*). During insulin detemir treatment, 23 hypoglycemic episodes occurred at night compared with 38 during NPH therapy, based on home blood glucose measurements during the last week of each treatment period. On the first day of insulin detemir treatment, where subjects were transferred to twice the run-in dose of NPH, six subjects had seven hypoglycemic episodes, one of which was rated as major. In comparison, nine subjects had 13 episodes on NPH, all of which were minor. The subject who had a major hypoglycemic episode experienced 2 minor episodes during the remaining treatment period on insulin detemir compared with 10 on NPH.

Adverse events

Approximately 30% of subjects had adverse events during either treatment period. There were no withdrawals because of adverse events, but two major hypoglycemic episodes were recorded as serious adverse events during the insulin detemir treatment period. The overall pattern of events was similar between treatments, and the majority was mild and considered unrelated to trial products.

CONCLUSIONS — This trial compared the blood glucose–lowering effect of insulin detemir and NPH in type 1 diabetic subjects on basal-bolus therapy. An open trial design was chosen because the two basal insulins were easily distinguishable (insulin determir being a solution and NPH a cloudy suspension), and the difference in dose requirements made it necessary to inject a larger volume of insulin detemir compared with NPH. The potential risk of a carry-over effect between the two treatments was believed to be limited, because the dynamic effect of the two insulins has fully disappeared within a 24-h period (11,15).

Analyses of the AUC(glu 23–08) of the glucose concentration time curves showed that once-daily administration of insulin detemir at bedtime could lower serum glucose to the same extent as NPH with equal requirements for meal-related insulin. This was supported by the overall similarity between the home-monitored eight-point blood glucose profiles obtained during the last week of each treatment period. Furthermore, no significant difference was observed in fructosamine levels between the two treatments, indicating that long-term therapy with insulin detemir was as effective as NPH in maintaining metabolic control.

Consistent with earlier findings (11), a higher molar dose of insulin detemir was needed to obtain similar blood glucose control compared with NPH. Although the doubling of the insulin detemir dose required a doubling of the injection volume, this caused no safety concerns. The impact of the difference in administered volume is not known, and, in general, it is difficult to compare the absorption of the two insulins because of their different modes of protraction. Earlier studies indicate that the rate of NPH absorption decreases with increasing volume and dose (16), which may be similar for insulin detemir, although pharmacokinetic studies in healthy subjects showed only marginal differences in time to maximal concentration between doses of 0.3 and 0.6 U/kg (9,11). However, to reduce the uncertainties concerning the impact of volume on insulin absorption, a more concentrated formulation of insulin detemir is currently under development.

The high individual variation in absorption rate is one of the major drawbacks related to current therapy with NPH (3,16). A smaller intrasubject variability in fasting blood glucose was observed with insulin detemir, although 65% of the subjects had to apply two injections. The lower variability together with the reduced action peak (9) could be an important advantage and facilitate strict metabolic control. More reproducible and predictable glucose levels throughout the night would minimize the risk of hypoglycemia and provide better glucose control in the early morning hours. Because treatment with insulin detemir resulted in higher mean serum glucose levels during the time interval 23:00–06:00, earlier administration and/or higher dosing might optimize therapy further.

A reduction in the number of subjects with hypoglycemia was observed during the last week of insulin detemir treatment. Because insulin doses remained unchanged during this period, the reduction was not a result of the decreased doses of meal-related insulin. The number of nocturnal hypoglycemic episodes was too limited for valid risk assessment. However, the higher serum glucose levels observed with insulin detemir during the night makes it unlikely that this risk would increase. As with all new insulins, careful monitoring of blood glucose should be performed in the initial days of treatment. Although one subject had a major hypoglycemic episode during the first day of insulin detemir treatment, hypoglycemia was less common when switching to the analog. It is recommended that NPH insulin pens are inverted a minimum of 20 times to ensure adequate resuspension (4). Introduction of soluble insulin detemir eliminates the inconvenience of adequate mixing before administration. In conclusion, insulin detemir appears to provide more predictable fasting blood glucose levels with lower intrasubject variation and to induce fewer hypoglycemic incidents than NPH.

Acknowledgments — This trial was sponsored by Novo Nordisk A/S, Denmark.

We would like to thank the personnel from the following investigational sites for their participation in the trial: Örebro Medical Center and University Hospital Lund, Sweden; King's College School of Medicine and Dentistry, London; and Taunton & Somerset Hospital, Taunton, U.K.

References

1. The Diabetes Control and Complications Trial Research Group: The effect of intensive treatment of diabetes on the development and progression of long-term complications in insulin-dependent diabetes mellitus. *N Engl J Med* 329:977–986, 1993
2. Reichard P, Nilsson BY, Rosenqvist U: The effect of long-term intensified insulin treatment on the development of microvascular complications of diabetes mellitus. *N Engl J Med* 329:304–309, 1993
3. Kølendorf K, Bojsen J, Deckert T: Clinical factors influencing the absorption of 125I-NPH insulin in diabetic patients. *Horm Metab Res* 15:274–278, 1983
4. Jehle PM, Micheler C, Jehle DR, Breitig D, Boehm BO: Inadequate suspension of neutral protamine Hagedorn (NPH) insulin in pens. *Lancet* 354:1604–1607, 1999
5. Brange J, Vølund Å: Insulin analogs with improved pharmacokinetic profiles. *Adv Drug Deliv Rev* 35:307–335, 1999
6. Whittingham JL, Havelund S, Jonassen I: Crystal structure of a prolonged-acting insulin with albumin-binding properties. *Biochemistry* 36:2826–2831, 1997
7. Markussen J, Havelund S, Kurtzhals P, Andersen AS, Halstrøm J, Hasselager E, Larsen UD, Ribel U, Schäffer L, Vad K, Jonassen I: Soluble, fatty acid acylated insulins bind to albumin and show protracted action in pigs. *Diabetologia* 39:281–288, 1996

8. Kurtzhals P, Havelund S, Jonassen I, Kiehr B, Larsen UD, Ribel U, Markussen J: Albumin binding of insulins acylated with fatty acids: characterization of the ligand-protein interaction and correlation between binding affinity and timing of the insulin effect in vivo. *Biochem J* 312:725–731, 1995
9. Heinemann L, Sinha K, Weyer C, Loftager M, Hirschberger S, Heise T: Time-action profile of the soluble, fatty acid acylated, long-acting insulin analogue NN304. *Diabet Med* 16:332–338, 1999
10. Strange P, McGill J, Mazzeo M: Reduced pharmacokinetic (PK) variability of a novel, long-acting insulin analog (Abstract). *Diabetes* 48 (Suppl. 1):A103, 1999
11. Brunner GA, Sendlhofer G, Wutte A, Ellmerer M, Søgaard B, Siebenhofer A, Hirschberger S, Krejs GJ, Pieber TR: Pharmacokinetic and pharmacodynamic properties of long-acting insulin analog NN304 in comparison to NPH insulin in humans. *Exp Clin Endocrinol Diabetes* 108:100–105, 2000
12. Trinder P: Determination of glucose in blood using glucose oxidase with an alternative oxygen acceptor. *Ann Clin Biochem* 6: 24–27, 1969
13. Johnson RN, Metcalf PA, Baker JR: Fructosamine: a new approach to the estimation of serum glycosylprotein: an index of diabetic control. *Clin Chim Acta* 127:87–95, 1983
14. Goldstein DE, Little RR, Wiedmeyer HM, England JD, McKenzie EM: Glycated hemoglobin: methodologies and clinical applications. *Clin Chem* 32 (Suppl. 10): B64–B70, 1986
15. Starke AA, Heinemann L, Hohmann A, Berger M: The action profiles of human NPH insulin preparations. *Diabet Med* 6: 239–244, 1989
16. Lauritzen T, Pramming S, Gale EA, Deckert T, Binder C: Absorption of isophane (NPH) insulin and its clinical implications. *Br Med J* 285:159–162, 1982