Differential Effects of Human and Animal Insulin on the Responses to Hypoglycemia in Elderly Patients With NIDDM

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Recent studies suggest that insulin-dependent diabetes mellitus patients switched from animal to human insulin may have decreased awareness of hypoglycemic warning symptoms. The risk of severe or fatal hypoglycemia associated with the treatment of diabetes increases with age. We conducted these studies to determine if awareness of hypoglycemic warning symptoms was greater with animal than with human insulin in elderly patients with diabetes. Nonobese elderly patients with non-insulin-dependent diabetes mellitus (NIDDM) (n = 13; age, 74 ± 1 years; body mass index, 26.6 ± 0.7 kg/m^2) underwent paired hyperinsulinemic glucose clamp studies (insulin infusion rate 60 mU · m^{-2} · min) in random order. In one study, regular human insulin was infused, and in the other study, regular beef/pork insulin was infused. In all studies, plasma glucose was decreased from fasting levels to 5 mmol/l during the first 60 min and was then allowed to fall to 4.4, 3.8, 3.3, and 2.8 mmol/l in each subsequent hour. Subjects were blinded as to which study they were undergoing. In each study, a hypoglycemic symptom checklist was administered, and counterregulatory hormones were measured every 15 min. Neuropsychological tests were performed every hour. Counterregulatory hormone responses to the two insulin preparations were similar. Autonomic ($P < 0.05$) and neuroglycopenic ($P < 0.01$) symptom scores were significantly higher during the beef/pork insulin studies. The responses on the neuropsychological tests were not significantly different. We conclude that beef/pork insulin results in greater awareness of hypoglycemic warning symptoms than does human insulin in elderly patients with NIDDM. *Diabetes* 44:272–277, 1995

In recent years, there has been a trend to treat patients with diabetes of all ages with human insulin rather than animal insulin. There has been extensive debate in the literature as to whether younger insulin-dependent diabetes mellitus (IDDM) patients treated with human insulin have a decreased awareness of hypoglycemic warning symptoms and an increased risk of hypoglycemia compared with similar patients treated with animal insulin (1–4). Since elderly patients with diabetes already have an increased frequency of hypoglycemic reactions (5–8), it would be advantageous to use the insulin preparation associated with the lowest likelihood of hypoglycemia in these patients.

We conducted the following studies to determine whether there was any difference between animal and human insulin in the intensity of hypoglycemic warning symptoms in elderly patients with diabetes. We also assessed the effects of the two insulins on counterregulatory hormone responses to hypoglycemia and the effects of hypoglycemia on psychomotor performance.

RESEARCH DESIGN AND METHODS

These studies were performed on elderly patients with non-insulin-dependent diabetes mellitus (NIDDM) (n = 13; age, 74 ± 1 years, range 65–85; 10 men, 3 women; body mass index, 26.6 ± 0.7 kg/m^2). The data from the hypoglycemic study with human insulin in 10 of these subjects have been reported elsewhere (9). Elderly patients with NIDDM had a disease duration of 6 ± 1 years and were being treated with diet (1 patient) or oral agents (12 patients) but not insulin. Nine patients were being treated with glyburide, one with gliclazide, one with tolbutamide, and one with metformin. Their mean HbA_{1c} value was 7.2 ± 0.3% (normal <6.4%). Seven of the patients were being treated with angiotensin-converting enzyme inhibitors or calcium channel blockers for hypertension. None of the patients had clinical evidence of cardiovascular disease (other than hypertension) or symptomatic peripheral neuropathy. No patients had experienced symptomatic hypoglycemic reactions within at least 8 weeks of the studies. All patients had glucometer readings at 1600 and 2200 the day before the study that were >5 mmol/l. In addition, all patients had a snack at 2100 the night before each study to minimize the risk of nocturnal hypoglycemia. All patients had normal renal function, no proteinuria, and no evidence of significant diabetic retinopathy when examined by an ophthalmologist. This study was approved by the Committee on Human Investigation at the University of British Columbia. All subjects gave written informed consent before participation.

Each subject underwent paired hyperinsulinemic glucose clamp studies, in random order, according to the method of Andres et al. (10). In the case of the 10 subjects whose data have been previously published (9), the animal insulin studies were performed concurrently and in random order with the human insulin hypoglycemic and control studies. By chance, eight subjects participated in the human insulin study first, and five participated in the animal insulin study first.

Studies were separated by at least 4 weeks. The longest interval between studies was 12 weeks. All studies commenced at 0730 in our clinical research center after an overnight fast. Oral agents were discontinued 24 h before each study. In all studies, intravenous lines were inserted into an antecubital vein for infusion of glucose and into a contralateral hand vein for the sampling of arterialized venous blood (11). Three blood samples were taken at 10-min intervals from −20 to 0 min to measure basal glucose, insulin, and counterregulatory hormones. At time 0, glucose clamp studies were started and continued for 300 min. In one study, regular human insulin (Lilly, Indianapolis, IN) was infused at a rate of 60 mU · m^{-2} · min. In the other study, regular beef/pork

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ANOVA, analysis of variance; IDDM, insulin-dependent diabetes mellitus; NIDDM, non-insulin-dependent diabetes mellitus.

TABLE 1
Basal glucose and hormone values

	Human insulin	Beef/pork insulin
Glucose (mmol/l)	7.7 ± 0.3	7.7 ± 0.4
Insulin (pmol/l)	113 ± 12	94 ± 6
Glucagon (ng/l)	155 ± 10	164 ± 10
Norepinephrine (nmol/l)	1.47 ± 0.12	1.45 ± 0.12
Epinephrine (pmol/l)	682 ± 60	650 ± 38
Growth hormone (μg/l)	1.7 ± 0.7	1.6 ± 0.4
Cortisol (nmol/l)	430 ± 52	326 ± 36*

Data are means ± SE; n = 13. *P < 0.05 human vs. beef/pork.

insulin (Connaught, Toronto, Ontario, Canada) was infused at the same rate. In all studies, plasma glucose was decreased from fasting levels to 5 mmol/l during the first 60 min. It was then allowed to fall to 4.4, 3.8, 3.3, and 2.8 mmol/l in each subsequent hour.

Blood samples were taken at 5-min intervals in each study to measure plasma glucose and at 15-min intervals to measure insulin and counterregulatory hormones. The coefficient of variation of plasma glucose did not exceed 5% in any study. Every 15 min during each study, a symptom questionnaire was administered to assess awareness of hypoglycemia. Neuropsychological tests were administered at 45, 105, 165, 225, and 285 min in each study. Blood pressure and pulse were measured at baseline and every 15 min. The patient, nurse, technician, and the psychologist administering the symptom questionnaire and neuropsychological tests were blinded as to which study the patient was undergoing.

Symptom questionnaire. The symptom questionnaire we used was similar to one previously described in the literature (12,13). The autonomic symptoms were sweating, shaking, hunger, nervousness, tingling, and pounding of the heart. The neuroglycopenic symptoms were blurred vision, weakness, tiredness, dizziness, difficulty in thinking, and faintness. The appropriateness of this classification of symptoms is supported by modification of hypoglycemic symptoms in studies using panautonomic blockade (14). Subjects were asked to rate the severity of each symptom on a visual analog scale from 0 (absent) to 10 (severe). The sum of the scores for all six symptoms constituted the autonomic or neuroglycopenic score at each point.

Neuropsychological tests. During the last 15 min of each hypoglycemic interval, subjects underwent a battery of neuropsychological tests, as previously described (9).

Analytical methods. Plasma glucose was measured immediately in all studies by the glucose oxidase method in a glucose analyzer (YSI, Yellow Springs, OH). The remaining blood was placed in prechilled test tubes containing aprotonin (400 KIU) and EDTA (1.5 mg/ml) and centrifuged at 4°C. All samples from each subject were analyzed in the same assay. Assays of glucagon were performed as previously described (15). Insulin assays were performed using a kit from Linco Research (St. Louis, MO). Cortisol and growth hormone assays were performed using kits from Immunocorp (Montreal, Quebec, Canada). Catecholamines were analyzed using high-performance liquid chromatography with electrochemical detection, as previously described (16).

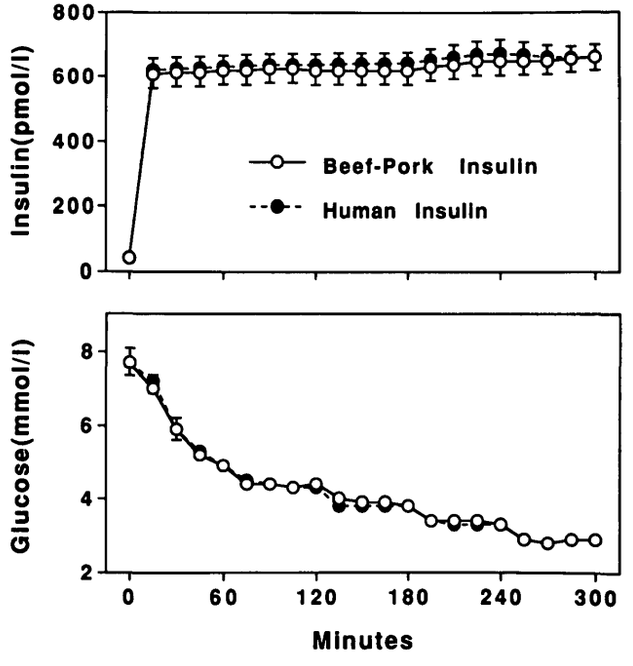

FIG. 1. Insulin and glucose values during the beef/pork and human insulin studies.

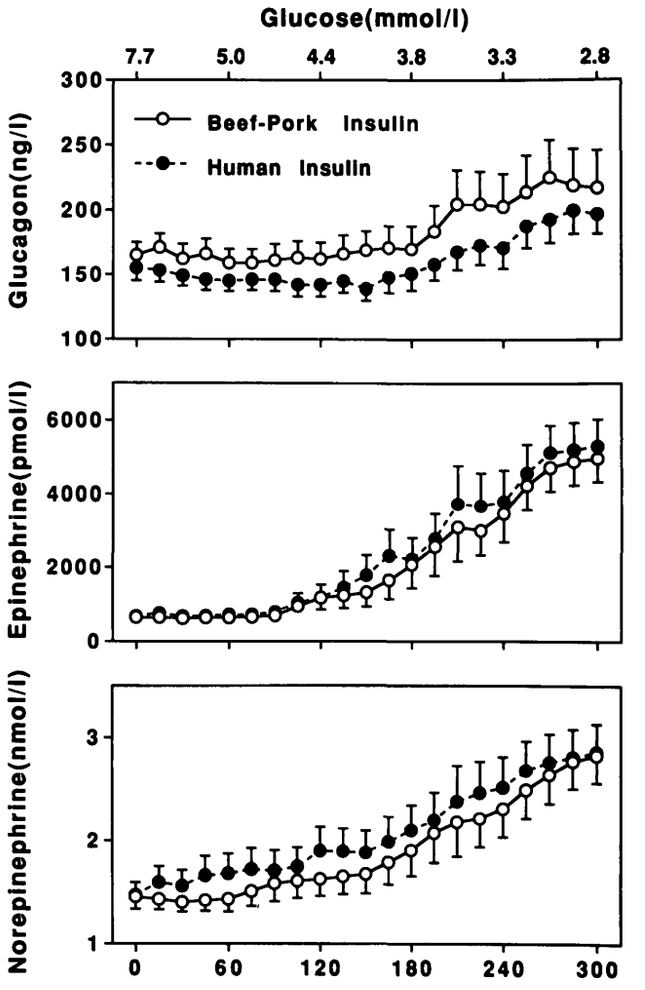

FIG. 2. Glucagon and catecholamine values during beef/pork and human insulin studies.

Statistical analysis. All results were presented as means ± SE. Differences between studies were determined using paired Student's t tests, and repeated measures of analysis of variance (ANOVA) were determined using the program Super ANOVA (Abacus Concepts, Berkeley, CA). The last two symptom questionnaire scores in each hour were averaged to calculate the mean score for that hour. Glucose thresholds for the onset of autonomic and neuroglycopenic symptoms were calculated as previously described (17). P < 0.05 was considered significant in all analyses.

RESULTS

Mean baseline values in patients are shown in Table 1. Basal cortisol values were higher before the human insulin study, but otherwise basal values were similar in the two studies. Glucose and insulin values during the studies are shown in Fig. 1 and were similar in the beef/pork and human insulin studies. Counterregulatory hormone levels during the study are shown in Figs. 2 and 3. There was no difference in counterregulatory hormone responses between the studies, as determined by repeated measures ANOVA.

Hypoglycemic symptom scores are shown in Fig. 4. Auto-

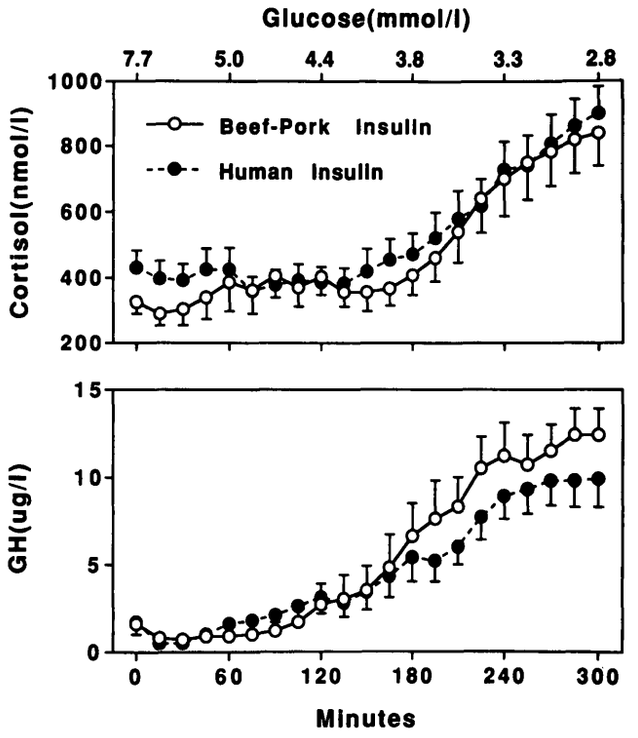

FIG. 3. Cortisol and growth hormone values during the beef/pork and human insulin studies.

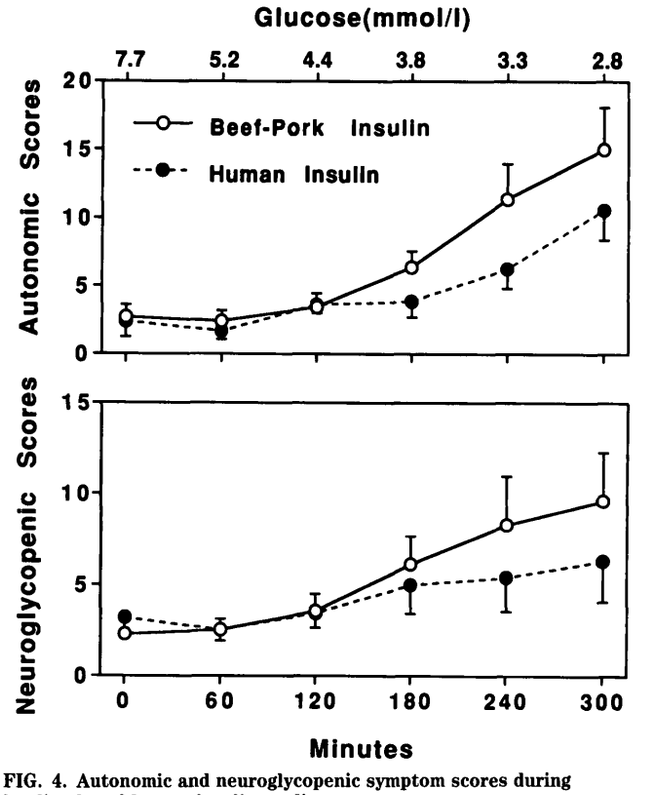

FIG. 4. Autonomic and neuroglycopenic symptom scores during beef/pork and human insulin studies.

nomic scores were significantly higher during the beef/pork insulin studies (F = 3.54, df = 12, $P < 0.05$). Neuroglycopenic symptom scores were also higher during the beef/pork insulin study (F = 4.72, df = 12, $P < 0.01$). The glucose threshold for awareness of autonomic symptoms was 3.3 ± 0.1 mmol/l for human insulin and 3.6 ± 0.1 mmol/l for animal insulin ($P < 0.05$). The glucose threshold for neuroglycopenic symptoms was 3.0 ± 0.1 mmol/l for human insulin and 3.3 ± 0.1 mmol/l for animal insulin ($P < 0.05$).

There was no significant difference between the beef/pork and human insulin studies in blood pressure and pulse values. The data are not shown.

Neuropsychological test results are shown in Table 2.

TABLE 2
Neuropsychological test results

	Glucose					
	Baseline, 7.7 mmol/l	60 min, 5.2 mmol/l	120 min, 4.4 mmol/l	180 min, 3.8 mmol/l	240 min, 3.3 mmol/l	300 min, 2.8 mmol/l
Simple reaction time (ms)						
Beef/pork	607 ± 30	673 ± 39	647 ± 42	642 ± 40	652 ± 43	694 ± 51*
Human	575 ± 32	612 ± 33	606 ± 33	624 ± 41	648 ± 38	691 ± 39†
Choice reaction time (ms)						
Beef/pork	736 ± 35	773 ± 44	780 ± 56	787 ± 46	900 ± 59	1,000 ± 84†
Human	725 ± 35	744 ± 40	782 ± 50	791 ± 46	848 ± 48	912 ± 43†
Digit symbol						
Beef/pork	40 ± 3	37 ± 3	38 ± 4	38 ± 4	36 ± 3	29 ± 2†
Human	43 ± 2	39 ± 2	39 ± 3	39 ± 3	35 ± 2	34 ± 2†
Digits backwards						
Beef/pork	6 ± 1	6 ± 1	6 ± 1	5 ± 1	5 ± 1	5 ± 1
Human	6 ± 1	7 ± 1	7 ± 1	6 ± 1	6 ± 1	5 ± 1
Stroop color (s)						
Beef/pork	15 ± 1	14 ± 1	16 ± 2	16 ± 1	15 ± 1	18 ± 1*
Human	15 ± 1	14 ± 1	14 ± 1	15 ± 1	16 ± 1	18 ± 1*
Stroop word (s)						
Beef/pork	29 ± 3	28 ± 3	27 ± 3	29 ± 2	28 ± 3	37 ± 3
Human	29 ± 3	26 ± 2	27 ± 3	30 ± 4	28 ± 2	32 ± 3
Story						
Beef/pork	9 ± 1	8 ± 1	7 ± 1	7 ± 1	6 ± 1	6 ± 2
Human	10 ± 1	9 ± 1	7 ± 1	8 ± 1	8 ± 1	9 ± 1
Pegboard (s)						
Beef/pork	96 ± 7	93 ± 5	103 ± 11	107 ± 12	116 ± 19	114 ± 8
Human	89 ± 6	90 ± 5	93 ± 6	92 ± 5	92 ± 5	97 ± 4

Data are means ± SE. *$P < 0.05$; †$P < 0.01$, baseline vs. hypoglycemia.

There was no significant difference between the studies in test scores at baseline. Hypoglycemia resulted in a significant impairment in performance on the simple and choice reaction time, digit symbol, and Stroop color tests, but there was no significant difference in performance between the human and animal insulin studies.

DISCUSSION

We conducted these studies to determine if there was any potential advantage of animal over human insulin for elderly patients with diabetes. We found that, although counterregulatory hormone responses to hypoglycemia induced by the two insulins were similar, animal insulin resulted in greater awareness of hypoglycemic warning symptoms and a higher glucose threshold for awareness of symptoms.

The literature regarding differences between human and animal insulin is conflicting. Studies comparing hypoglycemia induced with animal and human insulin in healthy young IDDM patients have found reduced (18–25) or unchanged (26–36) counterregulatory hormone responses to human insulin. Awareness of hypoglycemic symptoms has been found to be reduced (18,21,22,27,32,33,37) or unchanged (25,31,34–37) with human insulin. Our data are consistent with studies that report no differences in counterregulatory hormone responses and reduced awareness of hypoglycemia with human insulin. In our study, we found reduced awareness of both autonomic and neuroglycopenic symptoms of hypoglycemia. This is in contrast to the work of Egger et al. (38), who found that human insulin resulted in reduced awareness of autonomic but not neuroglycopenic symptoms of hypoglycemia.

If animal insulin results in greater awareness of hypoglycemic warning symptoms, what is the mechanism for the effect? In particular, why were there similar catecholamine and heart rate responses to hypoglycemia with the two insulin preparations, but reduced autonomic symptom responses with human insulin? We do not have a definitive explanation for the separation between hormone and symptom responses, although this is certainly consistent with data from other investigators (22,32). It is likely that animal insulins cross the blood-brain barrier more readily than human insulins since they are more lipophilic (39). It has been shown in animals that insulin stimulates glucose uptake in the hypothalamus (40) and that direct stimulation of hypothalamic insulin receptors evokes behaviors consistent with hypoglycemia (41). If hormonal responses are mediated primarily by the glucose levels in the central nervous system and the symptom responses are mediated, at least in part, by the direct effect of insulin on neurons, this could be an explanation for the differential effect of the two insulins.

Even though some experimental studies have demonstrated reduced hypoglycemic awareness with human insulin, is this of any significance in clinical practice? Again, the literature is conflicting. Surveys of IDDM patients switched from animal to human insulins and randomized controlled trials of animal and human insulin have found either reduced awareness of hypoglycemic symptoms and increased frequency of hyperglycemia with human insulin (38,42–48) or no difference between the insulin preparations (4,36, 49–55). The most compelling study in this area is by Colagiuri et al. (53). These investigators performed a randomized controlled trial of human and animal insulin in 67 IDDM patients who had reported hypoglycemic unawareness when changing from porcine to animal insulin. They found no difference in the frequency of symptomatic or asymptomatic hypoglycemia with the two insulins, suggesting that any differences are not clinically meaningful, at least in younger IDDM patients. There is little information about elderly patients treated with different insulin preparations. Burden (56) reports frequent hypoglycemic events in an elderly woman switched from animal to human insulin. Berger (4) followed 75 elderly diabetic patients receiving human and animal insulin for 2 years and found no difference in the frequency of hypoglycemic events between the two preparations. Berger's results do not mean that there is no clinically significant difference between human and animal insulins in the elderly. This study was not a randomized controlled trial and is subject to all the biases of a descriptive study. In addition, blood glucose values were not rigorously monitored. It is known that symptomatic awareness of hypoglycemia is reduced in the elderly (9,57). In addition, randomized controlled trials of oral hypoglycemic agents in the elderly, in which blood glucose values were monitored regularly, have found that low glucose values are frequently asymptomatic in this population (58). Thus, while the number of symptomatic events may have been similar with human and animal insulin in Berger's study, the number of asymptomatic events could conceivably have been greater with human insulin.

By chance, eight subjects underwent the study with human insulin first. Since these subjects may have been less anxious with the second study, their symptom responses could have been altered. We studied a group of elderly diabetic patients whose diabetes was well controlled with oral agents, who were free from complications, and who would not normally be candidates for insulin therapy. It may be argued that it is difficult to extrapolate our results to clinical practice. We have been responsible for the care of large numbers of elderly patients with diabetes, and it has been our experience that elderly patients receiving insulin generally have disease of longer duration, more complications, and poorer metabolic control than do patients receiving small doses of oral agents. We felt that by studying a relatively homogeneous group of patients with good control, we would reduce the number of factors that would interfere with the interpretation of our results. We were also inducing relatively severe hypoglycemia in these studies, and we were concerned how this would be tolerated by frail elderly patients who were receiving insulin and who had multiple complications. Now that we have established the safety of our protocol, it would be appropriate to perform future studies on elderly patients who are treated with insulin. Nonetheless, we believe that the current studies are an important first step in defining the role of different insulin species in the care of the elderly patient with diabetes.

Since glyburide has a duration of action of at least 24 h in some patients, it is possible that several patients experienced nocturnal hypoglycemia the night before the study, which could have affected the results. We think that this is unlikely, since all patients had an evening snack and evening glucometer readings were normal, but we cannot exclude this possibility entirely because glucometer readings were not performed in the middle of the night. It is possible that short-term fluctuations in metabolic control between the studies could have altered the results. Although we do not have fructosamine values to definitively exclude this possi-

bility, we think it is unlikely because our patients' conditions were stable and their HbA_{1c} values were essentially similar in the 6 months before and the 6 months after the study.

The neuropsychological tests we used were selected because they test a variety of cognitive domains and because previous investigators (12,17) found that performance on these tests is impaired at glucose levels similar to those used in this study. Consistent with previous work (9,12,17,57), we found that hypoglycemia impaired performance on several neuropsychological tests, although there was no difference between insulins. The lack of difference between insulins does not mean that there was no difference in the effects of these insulins on cognitive function during hypoglycemia. Rather, the tests may not have been sensitive enough to detect subtle differences. Kern et al. (27,59) found that there was a greater deterioration in auditory evoked potentials when hypoglycemia was induced with pork insulin rather than human insulin in normal young subjects. A potential mechanism for greater deterioration in neuropsychological function with animal insulin is the lipid solubility of the insulin preparations, as discussed above.

We conclude that, when compared with human insulin, beef/pork insulin may result in higher scores on a symptom questionnaire designed to assess hypoglycemic warning symptoms. Our results should not be interpreted to imply that elderly patients be treated exclusively with animal insulin or that human insulin results in hypoglycemia unawareness. Insulin was given intravenously in this study and in high doses. We believe that additional studies should be performed to confirm our data, using different routes of insulin administration and different doses of insulin. If future studies confirm our data, then a randomized control trial of different insulin preparations should be performed in community-dwelling elderly patients with NIDDM to determine if animal insulins have any advantage in clinical practice.

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REFERENCES

1. Pickup J: Human insulin. *Br Med J* 299:991–993, 1989
2. Cryer PE: Human insulin and hypoglycemia unawareness. *Diabetes Care* 13:536–538, 1990
3. Gale EAM: Hypoglycemia and human insulin. *Lancet* ii:1264–1266, 1989
4. Berger M: Human insulin: much ado about hypoglycaemia (un)awareness. *Diabetologia* 30:829–833, 1987
5. Asplund K, Wiholm BE, Lithner F: Glibenclamide-associated hypoglycaemia: a report on 57 cases. *Diabetologia* 24:412–417, 1983
6. Selzer HS: Drug-induced hypoglycemia: a review based on 1418 cases. *Endocrinol Metab Clin North Am* 18:163–183, 1989
7. Sonnenblick A, Shilo S: Glibenclamide-induced prolonged hypoglycaemia. *Age Ageing* 15:185–189, 1986
8. Rump A, Stahl M, Caduff F, Berger W: 173 Cases of insulin-induced hypoglycaemia admitted to the hospital. *Deutsch Med Wochenschr* 112: 1110–1116, 1987
9. Meneilly GS, Cheung E, Tuokko H: Counterregulatory hormone responses to hypoglycemia in the elderly patient with diabetes. *Diabetes* 43:403–410, 1994
10. Andres R, Swerdloff R, Pozefsky T, Coleman D: Manual feedback technique for control of glucose concentration. In *Automation in Analytic Chemistry*. Skeggs LT Jr, Ed. New York, Medaid, 1966, p. 456–501
11. McGuire EAH, Tobin JD, Berman M, Andres R: Effects of arterial vs. venous sampling on analysis of glucose kinetics in man. *J Appl Physiol* 41:565–573, 1976
12. Mitrakou A, Ryan C, Veneman T, Mokan M, Jenssen T, Kiss I, Durrant J, Cryer P, Gerich J: Hierarchy of glycemic thresholds for counterregulatory hormone secretion, symptoms, and cerebral dysfunction. *Am J Physiol* 23:E67–E74, 1991
13. Schwartz NA, Clutter WE, Shah SD, Cryer PE: Glycemic thresholds for activation of glucose counterregulatory systems are higher than the threshold for symptoms. *J Clin Invest* 79:777–781, 1987
14. Towler DA, Havlin CE, Craft S, Cryer P: Mechanism of awareness of hypoglycemia: perception of neurogenic (predominantly cholinergic) rather than neuroglycopenic symptoms. *Diabetes* 42:1791–1798, 1993
15. Meneilly GS, Elahi D, Minaker KL, Sclater AL, Rowe JW: Impairment of non-insulin-mediated glucose disposal in the elderly. *J Clin Endocrinol Metab* 63:566–571, 1989
16. Meneilly GS, Minaker KL, Young JB, Landsberg L, Rowe JW: Counterregulatory responses to insulin-induced glucose reduction in the elderly. *J Clin Endocrinol Metab* 61:178–182, 1985
17. Widom B, Simonson DC: Glycemic control and neuropsychological function during hypoglycemia in patients with insulin-dependent diabetes mellitus. *Ann Intern Med* 112:904–912, 1990
18. Heine RJ, Van der Heyden EAP, Van der Veen EA: Responses to human and porcine insulin in healthy subjects. *Lancet* ii:946–948, 1989
19. Petersen KG, Schluter KJ, Kerp L: Less pronounced changes in serum potassium and epinephrine during hypoglycemia induced by human insulin (recombinant DNA). *Diabetes Care* 5:90–92, 1982
20. Rosak C, Althoff PH, Enzmann F, Schoffling K: Comparative studies on intermediary metabolism and hormonal counterregulation following human insulin (recombinant DNA) and purified pork insulin in man. *Diabetes Care* 5:82–89, 1982
21. Schluter KJ, Petersen KG, Sontheimer J, Enzmann F, Kerp L: Different counterregulatory responses to human insulin (recombinant DNA) and purified pork insulin. *Diabetes Care* 5:78–81, 1982
22. Landgraf-Leurs MMC, Brugelmann I, Kammerer S, Lorenz R, Landgraf R: Counterregulatory hormone release after human and porcine insulin in healthy subjects and patients with pituitary disorders. *Klin Wochenschr* 2:659–668, 1984
23. Sjobom NE, Lins PE, Adamsen U, Theodoressen E: A comparative study of the hormonal responses to insulin-induced hypoglycaemia using semi-synthetic human insulin and pork insulin in patients with type I diabetes mellitus. *Diabetic Med* 7:775–779, 1990
24. Kerr D, MacDonald I, Hillier J, Tattersall R: Adrenaline response to hypoglycemia and insulin species (Letter). *Lancet* i:836, 1989
25. Fisher BM, Gray CE, Beastall GH, Frier BM: Responses to acute insulin-induced hypoglycaemia in diabetic patients: a comparison of short-acting human and porcine insulins. *Diabetes Res* 1:1–8, 1988
26. Jones TW, Caprio S, Diamond MP, Hallarman L, Boulware SD, Sherwin RS, Tamborlane WV: Does insulin species modify counterregulatory hormone response to hypoglycemia? *Diabetes Care* 14:728–730, 1991
27. Kern W, Lieb K, Kerner W, Born J, Fehm HL: Differential effects of human and pork insulin-induced hypoglycemia on neuronal functions in humans. *Diabetes* 39:1091–1098, 1990
28. Fernandez RP, Cananeuva FF, Devesa J, Cabezas-Cerrato J: Metabolic and hormonal parameters after insulin-induced hypoglycemia in man: comparison between biosynthetic human insulin and purified pork insulin. *Horm Metab Res* 17:351–354, 1985
29. Le Roith D, Shemer J, Pickens W, Leslie N, Sperling M, Berelowitz M: Comparison of the counterregulatory hormone response to semi-synthetic human insulin and purified porcine insulin in normal subjects and patients with type I diabetes mellitus. *Clin Ther* 13:613–636, 1991
30. Owens DR, Vora JP, Tronier B, Keller U, Luzio S, Turkes A: Hormonal counterregulatory responses to human (semi-synthetic and recumbent DNA) and porcine insulin-induced hypoglycaemia. *Diabetes Res* 8:17–20, 1988
31. Bendtson I, Binder C: Counterregulatory hormonal response to insulin-induced hypoglycaemia in insulin-dependent diabetic patients: a comparison of equimolar amounts of porcine and semi-synthetic human insulin. *J Intern Med* 229:293–306, 1991
32. Lingenfelser T, Renn W, Plonz C, Radjaipour M, Eggstein M, Jakober B: Catecholamine response during human and pork insulin-induced hypoglycemia in IDDM patients. *Diabetes Care* 15:261–264, 1992
33. Lingenfelser T, Bueltner UW, Plonz C, Steffen J, Eggstein M, Jakober B: Hormonal counterregulation symptom awareness and neuropsychologic function in type I diabetes during insulin-induced hypoglycaemia. *Diabetic Med* 9:528–535, 1992
34. Ferrer JP, Esmatjes E, Gonzalez-Clemente JM, Goday A, Conget I,

Jimenez W, Gomis R, Rivera F, Vilardell E: Symptomatic and hormonal hypoglycaemic response to human and porcine insulin in patients with type I diabetes mellitus. *Diabetic Med* 9:522–527, 1992
35. Patrick AW, Bodmer CW, Tieszen KL, White MC, Williams G: Human insulin and awareness of acute hypoglycemic symptoms in insulin-dependent diabetes. *Lancet* 338:528–532, 1991
36. Maran A, Lemus J, Archibald H, MacDonald IA, Gale EAM, Amiel SA: Double-blind clinical and laboratory study of hypoglycaemia with human and porcine insulin in diabetic patients reporting hypoglycaemic unawareness after transferring to human insulin. *Br Med J* 3060:167–171, 1993
37. Jakober B, Lingenfelser T, Gluck H, Maassen T, Overkamp D, Renn W, Eggstein M: Symptoms of hypoglycaemia: a comparison between porcine and human insulin. *Klin Wochenschr* 68:447–453, 1990
38. Egger M, Smith GD, Teuscher AU, Teuscher A: Influence of human insulin on symptoms and awareness of hypoglycaemia: a randomized double-blind cross-over trial. *Br Med J* 303:617–621, 1991
39. Markussen J, Damgard U, Johansen NL, Sorgensen KM, Sorensen E, Thim L: Charkterisierung des aus Schweineinsulin praperierten Humaninsulin. *Aktuel Endokrinol* 1:104–114, 1981
40. Lucignani G, Namba H, Nehlig A, Porrino LJ, Kennedy C, Soliooff L: Effect of insulin on local cerebral glucose utilization in the rat. *J Cereb Blood Flow Metab* 7:309–314, 1987
41. Szabo AJ, Iguchi A, Burleson PD, Szabo O: Vagotomy or atropine blocks hypoglycemic effect of insulin-injected into ventromedial hypothalamic nucleus. *Am J Physiol* 244:E467–E471, 1983
42. Daneman D, Zinman B: Syndrome of hypoglycemia unawareness when changing insulin species. *Diabetes Care* 14:145–146, 1991
43. Teuscher A, Berger WG: Hypoglycemia unawareness in diabetics transferred from beef/porcine insulin to human insulin. *Lancet* ii:382–385, 1987
44. Berger WG, Althaus BU: Reduced awareness of hypoglycemia after changing from porcine to human insulin in IDDM. *Diabetes Care* 10:260–261, 1987
45. Hepburn DA, Eadington DW, Patrick AW, Colledge NR, Frier BM: Symptomatic awareness of hypoglycaemia: does it change on transfer from animal to human insulin? *Diabetic Med* 6:586–590, 1989
46. Egger M, Smith ED, Imboof H, Teuscher A: Risk of severe hypoglycaemia in insulin-treated diabetic patients transferred to human insulin: a case-control study. *Br Med J* 14:617–621, 1991
47. Berger W, Honegger B, Keller U, Jaeggi E: Warning symptoms of hypoglycemia during treatment with human and porcine insulin in diabetes mellitus. *Lancet* i:1041–1044, 1989
48. Peacock I, Taylor A, Tattersall RB, Douglas CA, Reeves WG: Effects of new insulins on insulin and C-peptide antibodies, insulin dose, and diabetic control. *Lancet* i:149–152, 1983
49. Muhlhauser I, Heinemann L, Fritsche E, von Lennep K, Berger M: Hypoglycemic symptoms and frequency of severe hypoglycemia in patients treated with human and animal insulin preparations. *Diabetes Care* 14:745–749, 1991
50. Orchard TJ, Maser RE, Becker DJ, Dorman JS, Drash AL: Human insulin use and hypoglycaemia: insights from the Pittsburgh Epidemiology of Diabetes Study. *Diabetic Med* 8:469–474, 1991
51. Gorden P: Human insulin and hypoglycemia. *N Engl J Med* 322:1007–1008, 1990
52. Home PD, Mann NP, Hutchison AS, Park P, Walford S, Murphy M, Reeves WG: A fifteen-month double-blind cross-over study of the efficacy and antigenicity of human and pork insulins. *Diabetic Med* 1:93–98, 1984
53. Colagiuri S, Miller JJ, Petocz P: Double-blind cross-over comparison of human and porcine insulins in patients reporting lack of hypoglycemia awareness. *Lancet* 339:1432–1435, 1992
54. Clark AJL, Knight G, Wiles PG, Keen H, Ward JD, Cauldwell JM, Adeniyi-Jones RO, Leiper JM, Jones RH, MacCuish AC, Watkins PJ, Glynne A, Scotton JB: Biosynthetic human insulin in the treatment of diabetes: a double-blind cross-over trial in established diabetic patients. *Lancet* ii:354–357, 1982
55. Sonnenberg GE, Chantelau E, Sundermann S, Hauff C, Berger M: Human and porcine regular insulins are equally effective in subcutaneous replacement therapy: results of a double-blind crossover study in type I diabetic patients with continuous subcutaneous insulin infusion. *Diabetes* 31:600–602, 1982
56. Burden AC: Awareness of hypoglycemia in diabetes (Letter). *Lancet* i:1267, 1987
57. Meneilly GS, Cheung E, Tuokko H: Altered response to hypoglycemia of healthy elderly people. *J Clin Endocrinol Metab* 78:1341–1348, 1994
58. Brodows RG: Benefits and risks of glyburide and glipizide in elderly NIDDM patients. *Diabetes Care* 15:75–80, 1992
59. Kern W, Schlosser C, Kerner W, Pietrowsky R, Born J, Fehm HL: Evidence for effects of insulin on sensory processing in humans. *Diabetes* 43:351–356, 1994