Insulin Treatment and Type 1 Diabetes Topics

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This is the fifth in a series of articles on presentations at the American Diabetes Association Annual Meeting, San Diego, California, 10–14 June 2005.

Insulin treatment

Basal insulin. Malcolm Nattrass (Birmingham, U.K.) discussed basal insulin analogs, noting that the ideal approach will lead to "a flat profile that is reproducible." The development of basal insulin began with protamine zinc insulin in 1936, followed by NPH insulin in 1946 and zinc insulins lente, semilente, and ultralente in 1951. All of these preparations have high variability, making them less than optimal for treatment. Natrass reviewed studies of persons given four injections of 0.4 units/kg NPH in the thigh, with measurement of the glucose infusion rate required to maintain euglycemia. Great variability in biologic activity was shown. Early strategies pursued with insulin analogs included changes in the isoelectric point leading to precipitation at pH 7.4 with NovoSol Basal (1) and strengthening hexamer formation with Co(III)-hexamer insulin. Neither strategy led to successful development of a commercial product. Subsequent research led to insulin glargine, which is stable and soluble in acidic solution, precipitating following injection into subcutaneous tissues. Compared with NPH, there is major improvement in variability comparable to that with continuous subcutaneous insulin infusion (CSII) and a considerably "flatter" action profile than seen with NPH. Another method involves acylation of the insulin molecule with hydrophobic residues. Insulin detemir has a myristic acid fatty acid side chain that strengthens self-association, leading to increased hexamer formation and to albumin binding at the injection site and in the circulation. The binding to albumin may buffer insulin action.

Nattrass reviewed a number of studies of glargine and of detemir. In a study of 619 persons with type 1 diabetes, insulin glargine given at bedtime was associated with lower fasting glucose and with lower variability in fasting glucose level than NPH insulin given either once or twice daily (2). A similar 28-week study of 534 persons with type 1 diabetes showed reduction in HbA_{1c} (A1C) from 7.7 to 7.5% for both insulin glargine and NPH, with greater fall in fasting glucose and with 40 vs. 49%, respectively, having nocturnal hypoglycemia with the agents (3). In a study of 394 type 1 diabetic persons receiving insulin glargine at bedtime or NPH insulin twice daily, fasting glucose again showed greater decrease with glargine, with 73 vs. 82% having at least one glucose <50 mg/dl and 36 vs. 46% having at least one glucose <36 mg/dl (4). In 756 type 2 diabetic persons, the "treat-to-target" approach was associated with similar lowering of fasting glucose to ~120 mg/dl with both glargine and NPH, but again hypoglycemia was more frequent with NPH (5).

Comparing NPH with detemir, mean glucose levels are similar but variability is less with the latter agent. Detemir also is associated with less weight gain and, in persons with type 1 diabetes, with weight loss, when compared with NPH. In a study of 54 persons with type 1 diabetes receiving four doses of NPH, glargine, or detemir, with 24-h glucose infusion, the coefficients of variation of glucose required to maintain euglycemia were 68, 48, and 27%, respectively, suggesting that the detemir may have the most predictable glucose-lowering effect. Nattrass commented that with regular insulin, "the tail of the conventional [regular] insulin lasted much longer and conceivably made a contribution to the basal insulin," which is particularly a problem with long periods between meals, so that it may become difficult to "get away with a single injection" of basal insulin in persons receiving the shorter-acting insulin analogs, and many patients with type 1 diabetes require two basal doses.

A number of studies of basal insulin analogs were presented at the ADA meeting. Hermansen and Tamer (abstract 271) analyzed results of treatment of 475 insulin-naïve patients with type 2 diabetes with insulin detemir versus NPH twice daily, showing a BMI-related decrease in weight gain with detemir but not with NPH, with particular benefit of detemir in obese patients. Garber et al. (abstract 479) evaluated response to insulin detemir versus NPH among 418 and 890 persons aged ≥65 and <65 years, showing no difference in effect on A1C or fasting glucose but less weight gain and less variability of fasting glucose with insulin detemir. The likelihood of hypoglycemia was 40 and 23% lower with detemir than with NPH among the older and younger groups, respectively. Heller and Kim (abstract 487) and Kolendorf and Kim (abstract 489) compared hypoglycemia in 1,180 persons treated with detemir versus 810 receiving NPH insulin, showing that at every level of A1C, the frequency of hypoglycemia was lower with detemir versus NPH, ~36 vs. 48% at A1C 7% and 26 vs. 34% at A1C 8%, with an overall reduction in risk of hypoglycemia of 39%.

A number of studies addressed aspects of treatment with insulin glargine. Becker et al. (abstract 586) used the Biostator-supported euglycemic clamp in 24 nondiabetic men who were administered two preparations of 0.4 units/kg body wt glargine, finding that the coefficient of variation of the glucose infusion requirement was 18 and 32%. It would be interesting to see the degree of variability of detemir using this approach. Gerstein et al. (abstract 273) randomized 405 persons with type 2 diabetes and A1C >7.5% on up to two oral agents to either the addition of insulin glargine or to optimization of the oral regimen, showing more rapid and greater improvement in A1C and greater reduction in triglyceride and non-HDL cholesterol levels with glargine. Fiallo-Scharer et al. (abstract 1879) compared 45 children mixing insulin glargine with a rapid-acting insulin

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Abbreviations: CSII, continuous subcutaneous insulin infusion; DLCO, carbon monoxide–diffusing capacity; FEV1, forced expiratory volume in 1 s.

analog with 45 using separate injections. They showed no difference in A1C or in the percentage of blood glucose values at, above, or below 70–180 mg/dl, suggesting mixing as an option for children for whom the use of multiple injections is an issue.

Several studies addressed the question of whether insulin glargine acts for 24 h in all patients. Porcellati et al. (abstract 524) described an "afternoon phenomenon" of persons with type 1 diabetes treated with insulin glargine before dinner experiencing an increase in glucose levels from postlunch to predinner despite administration of a rapid-acting insulin analog before each meal. They defined the phenomenon by an increase in glucose by at least 50 mg/dl from 2-h postlunch to before dinner, when the 2-h postlunch glucose is <150 mg/dl, and with fasting glucose<110 mg/dl as evidence of optimized glargine dosage. Of a group of 143 persons with type 1 diabetes studied in this manner, 56 exhibited the phenomenon, suggesting that a substantial minority of type 1 diabetic persons have a duration of action of insulin glargine of <24 h. The pattern occurred on multiple occasions in most of the patients and was associated with A1C 7.3 vs. 7.1% in those not exhibiting the abnormality. Rossetti et al. (abstract 274) studied 60 patients exhibiting this pattern randomized to continue the same regimen, change the prelunch bolus to regular insulin, change the glargine to twice daily (50% with each dose), or add 1–4 units of rapid analog insulin 3 h after lunch. Improvements in late afternoon glycemia, as well as resulting decreases in A1C, were only observed for the latter approach. The authors believe that the phenomenon is not due to a <24-h duration of action of glargine but instead represents insufficient insulin replacement with rapid-acting analogs for the lunch meal. However, Ashwell et al. (abstract 483) randomized 20 people with type 1 diabetes to once-daily insulin glargine injection at dinnertime or twice-daily injection at breakfast and dinnertime, showing lower postbreakfast, postlunch, predinner, and mean 24-h self-monitored glucose levels, with lower within-day variability, although overnight glucose levels were higher with the twice-daily regimen, leading these authors to suggest that there indeed is a phenomenon of "waning preinjection plasma insulin levels" in persons with type 1 diabetes taking insulin glargine once daily.

Rapid-acting insulin analogs. Tim Heise (Neuss, Germany) asked whether all the rapid-acting analogs are the same, noting that all are produced by modification of insulin's B28 amino acid to weaken self-association of monomers into hexamers, leading to faster onset and shorter duration of action. Insulin aspart and lispro use zinc, but glulisine requires a zinc-free solution to reduce its strength of aggregation. Comparison of aspart with lispro in 14 persons with type 1 diabetes showed similar time to peak with slightly longer subsequent duration of glucose-lowering action with aspart, although no differences were seen in blood glucose patterns (6). Furthermore, another study of 24 persons with type 1 diabetes stabilized glucose levels first and showed virtually identical serum insulin levels and glucose-lowering effects with the two agents (7). Comparing glulisine with lispro, Heise showed evidence of more rapid onset of action with glulisine in obese persons, although he was uncertain whether these different patterns have any clinical consequence, with a study of ~600 persons receiving insulin glargine once daily and glulisine versus lispro showing no difference in hypoglycemia or in the rapidity of fall in A1C, suggesting on balance that all three analogs may be regarded as equal.

Premixed insulin. Phillip Raskin (Dallas, TX) asked, "Are premixed insulins more than just a compromise?" He noted that the majority of his patients are from economically disadvantaged ethnic minority groups and would not be likely to follow a basal bolus regimen. On the other hand, he stressed the importance of the ADA goal of A1C <7% and premeal glucose between 90 and 130 mg/dl. Few of his patients actually follow a diet regimen, and the use of multiple oral agents may also be complex, so that an effective approach to treatment of type 2 diabetes is that of premixed insulin plus a sensitizer. He suggested that this is a potentially synergistic regimen leading to better patient acceptability, in part due to less complexity and fewer injections, with more favorable lipid changes, and less expense. Raskin reviewed a 24-week study of 43 insulin-treated type 2 diabetic patients, 40% receiving premixed 70/30 insulin, randomized to addition of metformin versus placebo, with A1C falling from 9% by 2.5% vs. 1.5% and with weight gain 0.5 vs. 3.2 kg (8). A similar study of 28 insulin-treated persons with type 2 diabetes compared insulin plus metformin versus insulin plus troglitazone for 4 months, with subsequent combined insulin, metformin, and troglitazone treatment of all participants for an additional 4 months. A1C decreased from 8.5 to 7% with the addition of metformin and to 6.2% with the addition of troglitazone, without further decrease using both sensitizers, but body weight showed a 0- vs. 4.4-kg increase comparing addition of metformin versus troglitazone, with metformin followed by troglitazone appearing to prevent weight gain as well (9). He suggested that insulin be given to type 2 diabetic patients with significant hyperglycemia at presentation, particularly when associated with weight loss, as well as those who remain hyperglycemic despite diet, exercise, and maximal-dose oral agents. He often starts with two daily injections of a mix of intermediate- and short-acting insulin, often with 70/30, initially adding metformin and then changing the insulin dose. If A1C fails to reach goal, a thiazolidinedione is then added. Reviewing a 28-week study of 233 insulin-naïve type 2 diabetic persons with A1C >8% on oral agents, including metformin, in a dose exceeding 1,000 mg daily, administration of glargine at bedtime versus 70/30 aspart twice daily led to a decrease in A1C of 2.4 vs. 2.8%, with particular benefit of premixed insulin for those with initial A1C >8.5% who fell 2.6 vs. 3.1% (10), leading Raskin to suggest that "70/30 is more than a compromise."

Barratt et al. (abstract 673) randomized 24 type 2 diabetic persons within 4 weeks of starting insulin to control versus "lifestyle clinic" for 6 months. A weight gain of 4.1 kg versus a loss of 0.4 kg was observed, as well as an increase versus decrease in blood pressure, suggesting the benefit of this approach when persons with type 2 diabetes begin insulin treatment. Ligthelm et al. (abstract 496) compared three prandial insulin aspart doses plus NPH at bedtime with biphasic aspart 70/30 three times daily in 196 and 198 persons with type 2 diabetes, respectively, showing a similar decrease in A1C from 9.1 to 7.8% with both approaches. Brod et al. (abstract 461) randomized 233 persons with type 2 diabetes to insulin glargine once daily versus biphasic aspart 70/30 twice daily and reported 28 vs. 42% likelihood of achieving A1C ≤6.5% over 28 weeks, with both groups reporting similar satisfaction with treatment, suggesting that multiple dose approaches can be accepted by patients. Jain et al. (abstract 278) described an approach to

biphasic aspart 70/30 insulin treatment of 100 persons with type 2 diabetes whose initial A1C averaged 8.6%, in which 21% attained A1C <6.5% with one daily insulin dose before dinner. Forty-one percent of remaining subjects attained A1C <6.5% with insulin before breakfast and before dinner, and 32% of those failing to attain goal with two doses attained A1C <6.5% with insulin administration before each meal. Lund et al. (abstract 511) reported that with a similar protocol, moving from one to two to three daily doses of biphasic aspart 70/30 in the same fashion, 37% attained A1C <6.5% at 12 months. Woerle et al. (abstract 310) treated 164 persons with type 2 diabetes and mean A1C 8.7% to goal <7%, achieving a mean of 6.5%. Comparing those who achieved A1C <7% (mean 6.2%) with those who did not (mean 7.6%), fasting glucose was 105 vs. 107 mg/dl but daylong glycemia on self-measured seven-point glucose profiles was 122 vs. 143 mg/dl, suggesting the importance of postprandial glucose excursions in achieving or not achieving strict glycemic goals. Shankhdhar et al. (abstract 453) described an interesting approach in which 30 type 2 diabetic patients failing to achieve goal with oral agents were randomized to premixed 70/30 human insulin with or without acarbose. A1C decreased from 11.5 to 7.2% versus decreasing from 11.8 to 8.7% with the two approaches, with 27 vs. 0% achieving A1C <7%. Acarbose presumably acted to improve postprandial glycemia. Uwe et al. (abstract 583), however, compared 130 type 2 diabetic persons aged ≥65 years with baseline A1C 8.9% who were randomized to human 70/30 NPH/regular insulin twice daily versus continued oral agents plus insulin glargine once daily. They showed that A1C decreased 1.4 vs. 1.9%, fasting glucose decreased 40 vs. 57 mg/dl, and 30 vs. 55% achieved A1C <7% without nocturnal hypoglycemia. In a study of insulin pen therapy, Asakura and Seino (abstract 525) measured the pressure required to depress the plunger of a variety of insulin delivery devices, finding it to be lowest with the InnoLet device, approximately twice as great for the NovoPen 3, FlexPen, and HumaPen Ergo, and more than three times as great for the Humalog/Humulin Pen and OptiPen Pro 1, suggesting that the latter devices "may be difficult for some patients due to high injection pressure."

Insulin pump therapy. Svoren et al. (abstract 1007) compared acute adverse event rates among 299 children aged 7–16 years with type 1 diabetes in 1997 vs. 152 in 2002, 65 vs. 85% taking more than two insulin injections per day, including 0 vs. 23% using CSII and 76 vs. 88% testing blood glucose more than twice daily. A1C was 8.7 vs. 8.4%, and there were 29 vs. 22 emergency room visits and 55 vs. 29 hypoglycemic episodes · 100 patients^{-1} · year^{-1}, suggesting that intensification of therapy, with multiple insulin doses or pump therapy and use of more frequent glucose testing, has improved glycemic control and reduced emergency room use and hypoglycemia rates

Danne et al. (abstract 1887) collected continuous CSII programming history with centralized laboratory A1C measurement in 1,041 children aged 12 years. They found that those taking more than five versus four or less boluses per day had A1C 7.8 vs. 8.7% and that those with less than versus half or more of their total daily insulin dose administered as basal insulin had A1C 7.8 vs. 8.5%, suggesting the optimal approach to be one with less basal insulin and more than five daily boluses. Jorgensen and Solbeck (abstract 416) reported stability of insulin aspart with the use of NPH-diluting medium for children using low-dose insulin for CSII treatment. Siegmund et al. (abstract 427) randomized 20 persons to CSII with insulin lispro versus aspart in a 4-week cross-over study. They reported that aspart showed significantly less burning, inflammation, and dermal redness, with 68% of patients preferring aspart to lispro. However, in another randomized study of 18 persons on CSII, Block et al. (abstract 2064) found no difference in duration of stable control between the two insulin analogs and found that patients correctly guessed the insulin they were using on only 53% of occasions. Davidson et al. (abstract 442) reported that use of an automated algorithm suggesting adjustments in both basal rates and insulin boluses was associated with a 0.4% decrease in A1C in 157 patients receiving CSII, while no change was seen in 297 patients concurrently treated at the same center without use of this program. Reichel et al. (abstract 434) compared 40 type 1 diabetic persons starting CSII with stimulated C-peptide <0.1 nmol/l versus 10 persons with C-peptide 0.1–0.3 nmol/l. Over a 3-year period, A1C decreased from 8.3 to 7.6 vs. 7.3%, and hypoglycemia occurred approximately half as frequently in the patients with endogenous insulin. Heptulla et al. (abstract 447) administered pramlintide either as a subcutaneous bolus or a square wave in eight persons with type 1 diabetes treated with CSII. The immediate postprandial hypoglycemia seen with bolus administration did not occur with more gradual infusion, with lesser glucagon suppression and delay in gastric emptying. Von Dobeln et al. (abstract 495) administered a partial basal replacement dose of insulin glargine to seven persons with type 1 diabetes treated with CSII, showing prevention of the nocturnal ketosis otherwise occurring when CSII was discontinued at 7:00 P.M. Herman et al. (abstract 504) randomized 48 adults with type 2 diabetes aged ≥60 years to CSII versus 50 similar patients to glargine plus multiple-dose lispro subcutaneous insulin, showing a similar fall in mean A1C from 8.4 to 6.8% over 6 months with stabilization through 12 months. Both approaches were associated with similar frequency of mild hypoglycemia, with three versus six persons in the two groups experiencing an episode of severe hypoglycemia.

Inhaled insulin. Jay Skyler (Miami, FL) reviewed developments with inhaled insulin, which was first studied in 1925, so that perhaps we should not "hold our breath" in waiting for these agents. The inhalation of systemically active pharmaceuticals allows absorption across the 100- to 140-m^2 alveolar surface area, equivalent in size to a tennis court. The efficiency of delivery varies with the device studied, with between 20 and 40% reaching the lungs and between 8 and 15% reaching the systemic circulation. Dry powder formulations have been employed by Pfizer, Sanofi Aventis, and Nektar (Exubra), MannKind (Technosphere), Lilly/Alkermes, and Bristol-Myers Squibb/Qdose. Generex is studying a dry powder for buccal administration. Liquid formulations are being studied by Novo Nordisk/Aradigm (using the AERx insulin diabetes management system electronically powered inhaler) and Kos, with a breath-actuated inhaler containing a multidose canister. Other companies that have explored inhaled insulin formulations include Aerogen, Dura ("Spiros" system), AstraZeneca, and Coremed Alveair, although development of these products appears to have been suspended.

Pulmonary administration of insulin may enhance nonhepatic glucose clearance in comparison to the effect of insulin

administered intravenously (11). The first publication describing use of the Exubra formulation of inhaled insulin in dry powder form in 2001 described a similar action profile to that of rapid-acting insulin analogs, with a somewhat longer duration of action (12). Around the same time, the AERx liquid formulation was shown to exhibit a dose-response effect (13). Skyler reviewed data showing the time-action profile of inhaled insulins, suggesting that the Technosphere formulation has more rapid onset, while the other agents are similar to lispro.

In clinical studies of persons with type 1 diabetes, inhaled insulin has been shown to have similar effect to that of subcutaneous insulin (14,15), with Skyler mentioning that the Technosphere preparation may have lower variability than subcutaneous insulin. Studies in type 2 diabetic persons suggest a similar effect to subcutaneous insulin (16), with some evidence of less weight gain and hypoglycemia (17). Interestingly, studies comparing type 2 diabetic persons treated with once-daily glargine versus preprandial inhaled insulin suggest similar improvement in glycemia.

There are a number of potential issues related to the effects of pulmonary disorders on inhaled insulin and possible effects of inhaled insulin on lung function. Cigarette smoking increases absorption of inhaled insulin by increasing permeability, while asthma decreases absorption, although without evidence of change in pulmonary flow rate after acute dosing in asthmatic persons (18). No change in insulin pharmacokinetics has been reported with acute upper-respiratory infection. A modest increase is seen in IgG insulin antibodies after inhaled insulin administration, although not to the levels seen in the past with impure animal insulin preparations, and there is no relationship of antibody levels to change in insulin action or in pulmonary function. Skyler concluded that inhaled insulin is effective, that "there are no unique safety issues," and that the approach should meet regulatory requirements for approval, noting that diabetes control, quality of life, cost-benefit, use in children, and effects of smoking and of exercise require further study.

A number of studies of inhaled insulin were presented at the ADA meeting. Petersen et al. (abstract 410) reported the effect of high versus normal tidal volume ventilation in rabbits administered inhaled human insulin (5 units) via a nebulizer system, showing a 49% increase in total insulin absorption with the former, suggesting the need for caution with inhaled insulin during a change in breathing pattern, such as may occur with physical exercise. Dumas et al. (abstract 355) randomized 226 persons with type 1 diabetes receiving once- or twice-daily intermediate- or long-acting insulin to subcutaneous short-acting versus Exubera inhaled insulin before meals for 24 weeks. A1C decreased similarly from 7.5 to 7.0 vs. 7.1%, with 24% more minor hypoglycemia but 48% less severe hypoglycemia with inhaled insulin. At 2 weeks, the forced expiratory volume in 1 s (FEV1) and carbon monoxide–diffusing capacity (DLCO) decreased −0.070 l and −0.973 ml · min^{-1} · mmHg^{-1} for Exubera and −0.027 l and −0.246 min^{-1} · mmHg^{-1} for subcutaneous, respectively, with the decrease remaining stable through the study period, although it was resolved within 2 weeks after inhaled insulin was of discontinued (normal DLCO is usually in the range of 20 to 30 ml · min^{-1} · mmHg^{-1}). Cough was reported in 30 vs. 8% of patients, typically occurring after dosing. Cefalu et al. (abstract 356) reported on the use of the same inhaled insulin preparation in 158 type 2 diabetic persons versus 146 not receiving inhaled insulin. At 2 years, FEV1 decreased 0.08 vs. 0.07 l/year and DLCO decreased 0.7 ml · min^{-1} · mmHg^{-1} · year^{-1} in both groups. Rosenstock et al. (abstract 357) randomized 42 vs. 48 type 2 diabetic persons to placebo versus Technosphere pulmonary insulin, reporting A1C to decrease 0.3 vs. 0.8% from baseline 7.8%, without a difference in FEV1 or DLCO. Petersen et al. (abstract 359) reported comparison of AERx inhaled insulin with subcutaneous regular and aspart insulin, with the inhaled preparation showing similar pharmacodynamics to insulin aspart. Similar findings were reported by Rave et al. (abstract 360) with the Lilly/Alkermes inhaled insulin system. Garg et al. (abstract 361) compared the latter inhaled insulin versus subcutaneous insulin in 133 vs. 126 persons with type 1 diabetes over 12 weeks, showing similar A1C and hypoglycemia rate, with DLCO significantly lower at 25.3 vs. 26.3 ml · min^{-1} · mmHg^{-1} in the inhaled insulin group. Thus, there appears to be little doubt that the pulmonary route is effective in insulin delivery, although one may question whether this approach has truly been shown to be safe, given the consistent mean 3–5% reduction in DLCO, a measure of alveolar volume. It would be important to know the degree of variation of this effect. If a subset of persons exhibit a greater decrease, it might be wise to follow pulmonary function parameters in persons receiving this treatment.

Home glucose monitoring

Hanauer et al. (abstract 406) reported on the use of automated reminders for glucose testing by adolescents with type 1 diabetes. Cell phone SMS (short message service) text messaging was superior to e-mailed reminders, with patients randomized to the former system submitting more than twice as many glucose tests. Martin et al. (abstract 303) followed 3,268 patients with type 2 diabetes diagnosed between 1995 and 1999. Over an average of 6.5 years, 7.2 vs. 10.4% of those performing versus not performing home blood glucose monitoring had nonfatal myocardial infarction, stroke, foot amputation, blindness, or hemodialysis, and 2.7 vs. 4.6% had fatal events, with benefit for both patients receiving and not receiving insulin therapy. Davidson et al. (abstract 408) studied 552 non–insulin-treated diabetic patients, showing that there was an inverse correlation between glucose testing frequency and A1C, with those in the highest versus lowest two quintiles of glucose testing having mean A1C of 6.7 vs. 7.2%. There appeared to be little additional benefit of performing more than two glucose tests per day in this population. Schwartz et al. (abstract 391) compared capillary glucose levels determined from samples taken from the palm and fingertip in 181 persons with type 1 and type 2 diabetes, showing ~96% agreement between the two sites, similar to that when glucose samples from two different fingers were compared.

Hypoglycemia

Rolf Gruetter (Minneapolis, MN) discussed the use of magnetic resonance spectroscopy, which allows measurement of the concentrations of specific compounds, including glucose, in specific areas of the brain. Brain glucose measurement either with ^{13}C or with proton spectroscopy allows determination of transport rates into and out of brain. Brain glucose levels are proportional to circulating glucose, allowing determination of glucose transport rates. Under circumstances of chronic hypoglycemia, glucose transport is upregulated. In a rat model with implanted insulin pellets leading to a 14-day period of hypoglycemia, brain glucose levels decrease and glucose trans-

port (via GLUT1) subsequently increases by 58%. Brain glycogen may be an important fuel source under these circumstances, with Morgenthaler et al. (abstract 633) reporting a ~50% increase in brain glycogen during either insulin or glucose infusion and an ~70% increase with combined glucose and insulin infusion. There may be a role of brain glycogen in glucose sensing. Human studies show that during mild hyperglycemia, glycogen levels are stable. Glycogen is present in astrocytes and provides energy to neurons under circumstances of decreased glucose availability. Glucose label appears in glutamate and then in glutamine, in neurons, and in astrocytes, with conversion of glutamate to glutamine playing a role in neurotransmission between these cell types. As excess glutamate is toxic, glial function may be crucial for neuronal health.

If "what matters" are organ-specific metabolite levels, then these approaches will be crucial to the understanding of brain metabolism. In studies from Gruetter's group, Criego et al. (abstract 630) measured brain glucose concentrations in nondiabetic persons following three episodes of hypoglycemia over 24 h, showing higher levels among persons whose glucagon, epinephrine, and norepinephrine responses were reduced during the third hypoglycemic episode. Oz et al. (abstract 639) measured brain glycogen in seven persons during glucose infusion, showing that levels increase continuously over 46 h, with evidence of persistent elevated levels for days, potentially contributing to hypoglycemia unawareness by providing additional substrate for brain glucose metabolism.

William Powers (St. Louis, MO) discussed the use of positron emission tomography methodology and potential applicability to diabetes research. He noted that positrons are forms of antimatter-emitting gamma rays after combination with electrons, with the gamma rays actually being measured to determine the radioactivity of specific tissue volumes. The positron emitter must be combined with specific chemicals with subsequent assessment of brain metabolism using mathematical physiologic models. This approach allows estimation of regional glucose transport and glucose metabolism. Deoxyglucose is phosphorylated but not further metabolized and can be used in positron emission tomography scanning. This can be used as an alternative approach to central nervous system metabolic imaging.

Musen et al. (abstract 627) and Simonson et al. (abstract 634) performed functional magnetic resonance imaging during insulin infusion, showing signal changes in the hypothalamic and pituitary regions at 73 mg/dl glucose in seven persons with type 1 diabetes and at 65 mg/dl in nine nondiabetic persons, suggesting higher and more variable onset of hypoglycemia in type 1 diabetes. In persons without diabetes during gradual induction of hypoglycemia, there were 5- to 12-min delays from the initial increase in hypothalamic and pituitary signal to increases in circulating glucagon, epinephrine, cortisol, and growth hormone There also was increased anterior cingulate cortex activity, suggesting that this region may play a role in the response to hypoglycemia.

A number of additional studies were presented at the ADA meeting on aspects of hypoglycemia. Sanders et al. (abstract 640) induced hypoglycemia in rats with insulin injection twice in 1 day and then once on a 2nd day, showing that despite attenuated glucagon and epinephrine response, there was preservation of increased feeding, which appeared to represent a conditioned response, as in a separate experiment, food intake also increased following a saline injection 1 day subsequent to hypoglycemia caused by insulin injection. Porcellati et al. (abstract 108) and Fanelli et al. (abstract 629) compared cognitive function during insulin-induced hypoglycemia in nine persons with type 1 diabetes with and without ingestion of a mixture of amino acids, showing this to limit the degree of memory impairment and to sustain attention and process information, although hypoglycemic symptoms were unaffected. Epinephrine and norepinephrine responses were similar, but the glucagon response increased after amino acid administration, although it remained lower in persons with type 1 diabetes than in nondiabetic persons. Fisher et al. (abstract 285) studied mice not expressing the insulin receptor in the brain. They showed a reduction in the norepinephrine and epinephrine response to insulin-induced hypoglycemia, a shift in glucocorticoid response to require lower glucose levels, and no change in glucagon response, with a consequent 57% reduction in hepatic glucose production, suggesting that insulin regulates central glucose sensing, affecting the sympathetic and adrenal cortical and medullary response to hypoglycemia. Paranjape and Briski (abstract 635) reported that activation of lateral hypothalamic area neurons containing the appetite-stimulating peptide orexin-A was decreased following four versus one dose of NPH insulin during a 24-h period, suggesting habituation of this neuronal response to insulin-induced hypoglycemia.

Nguyen et al. (abstract 619) studied eight persons with type 1 diabetes during 2-h insulin-induced hypoglycemia and control infusions. Glucose estimated from a feed-forward neural network, using skin impedance, heart rate, and rate-corrected QT interval as inputs, accounted for approximately half of the variance in the actual blood glucose with sensitivity of 0.7818 and specificity of 0.9304 for prediction of hypoglycemia, a potential approach for avoidance of hypoglycemic episodes. Hershey et al. (abstract 620) reported that children with type 1 diabetes and more than three severe hypoglycemic episodes had reduced gray matter in the right posterior hippocampal/parahippocampal region, potentially causing loss of spatial memory.

Høi-Hansen et al. (abstracts 621 and 622) performed 6-day continuous glucose monitoring in 119 patients with type 1 diabetes, showing 0.8 symptomatic and 3.0 silent episodes per week of glucose <40 mg/dl, with frequency twice as great among those not treated with an ACE inhibitor or angiotensin receptor blockers, for whom impaired hypoglycemia awareness increased the likelihood of silent hypoglycemia 1.6-fold. In a study of nine persons with high and nine with low renin-angiotensin system activity, only the former showed decreased memory during insulin-induced hypoglycemia, with this group showing a lesser autonomic response to hypoglycemia and both abnormalities potentially increasing the likelihood of severe hypoglycemia. Raju et al. (abstract 636) performed continuous glucose monitoring (CGMS; Medtronic MiniMed) in 17 persons with type 1 diabetes following no intervention or bedtime administration of a 26-g carbohydrate snack without or with acarbose, a 39-g carbohydrate cornstarch bar, or the β_2-adrenergic agonist terbutaline (5.0 mg), with 6, 3, 4, 4, and no patients having glucose <50 mg/dl and 10, 7, 9, 7, and 4 having glucose <70 mg/dl. The benefit of terbutaline occurred at the expense of a significant increase in morning glucose, with levels of 96, 106, 118, 135,

and 188 mg/dl, respectively. Interestingly, the continuous glucose monitoring method failed to identify 16% of 560 glucose levels <70 mg/dl (plasma glucose measured every 15 min), while of 972 CGMS values <70 mg/dl, 49% were not confirmed by plasma glucose.

Zammitt et al. (abstract 626) measured recovery following a reduction in glucose to 45 mg/dl for 1 h for 20 persons with type 1 diabetes, showing that cognitive function was impaired for 40 min following recovery of euglycemia (two consecutive glucose levels >72 mg/dl). The authors comment that this finding "has practical implications for tasks such as driving." Akram et al. (abstract 632) reported 0.44 episodes · person^{-1} · year^{-1} of severe hypoglycemia among 401 insulin-treated outpatients with type 2 diabetes (one-third type 1 diabetes) at the Steno Diabetes Center, with all episodes occurring in 66 of the patients. ACE inhibitor/angiotensin receptor blocker treatment reduced the risk by half, while the risk doubled for every 10 years of insulin therapy, increased 2.4-fold for persons living with a partner (presumably because of more reporting), and tripled with self-reported reduced hypoglycemia awareness. For those who had had one severe hypoglycemia episode, the risk of another episode doubled with the presence of macrovascular disease, tripled with the presence of neuropathy, and was reduced by two-thirds for each 10-year duration of diabetes before initiation of insulin treatment. Israelian et al. (abstract 637) reported reduced increments in glucagon and growth hormone, although not in epinephrine and cortisol, during a 2-h period of insulin-induced hypoglycemia, as well as slower rates of decrease in insulin secretion, in 14 persons with type 2 diabetes (mean duration 5.3 years) compared with 21 nondiabetic subjects.

Type 1 diabetes

Rewers et al. (abstract 1,055) noted that 1 in 465 children in the U.S. develops type 1 diabetes by 10 years of age and reported that follow-up of 31,760 unselected neonates and 1,336 first-degree relatives showed that the DR3/4, -4/4, -4/1, -4/8, or -4/9 with DQB1*0302, or the DR3/3 HLA genotypes, found in 10% of the population, were present in 70% of those developing diabetes, with antibody testing at ages 18 and 42 months allowing a 62% sensitivity for detecting those in this group who ultimately developed diabetes, suggesting a potential screening strategy. Casu et al. (abstract 121) further characterized the risk associated with autoantibody to islet tyrosine phosphatases, which may play a role in insulin secretion. Among family members with versus without this autoantibody, 86 vs. 63% developed type 1 diabetes over a 10-year period. Stene et al. (abstract 122) reported that an increase in A1C within the normal range was associated with an almost fivefold increased likelihood of progression to overt diabetes among high-risk children who had developed a positive autoantibody. Brady et al. (abstract 119) studied children in the high-risk HLA group or with a positive family history of type 1 diabetes. Those who had autoantibody to insulin, GAD, or islet tyrosine phosphatases were twice as likely to have taken a multivitamin supplement, with somewhat greater risk for those with the A/A or G/A genotype of the vitamin D receptor intron 8 polymorphism. Goodwin et al. (abstract 123) compared 29 sibling pairs concordant for type 1 diabetes with diagnosis before age 10 years with 27 pairs diagnosed after age 10. Evidence of thyroid autoimmunity was present in 17 vs. 6%, and 24 vs. 4% had a parent with type 1 diabetes, suggesting different genetic determinants in those with younger age at onset.

Lamb et al. (abstract 1050) reported correlation of BMI with a higher birth weight, greater weight gain from birth to 15 months, and lower annual income in 1,034 children followed for up to 10 years. The Search for Diabetes in Youth Study Group (abstract 1012) identified 6,382 youth with diabetes from a population of 3.4 million in 2001, for an overall diabetes prevalence of 1.8 per 1,000 children in 2001, increasing from 0.3 at age 0–4 years to 1.3 at age 5–9, 2.3 at age 10–14, and 3.4 at age 15–19. Gilliam et al. (abstract 1905) reported that 57 and 84% of those with type 1 and type 2 diabetes had a positive family history of diabetes. The Group (abstract 124) ascertained diabetes type among 1,311 newly diagnosed children with diabetes in 2002 in a population of 5 million. Among those aged 0–9 years, the majority had type 1 diabetes with evidence of autoimmunity, with annual incidence 26/100,000 among non-Hispanic whites, 15/100,000 among African Americans, 15/100,000 among Hispanics, 7/100,000 among Asian/Pacific Islanders, and 6/100,000 among American Indians, with the great majority type 1, but at age 10–19 years, respective incidences were 30, 41, 34, 29, and 49/100,000, with three-quarters, half, half, one-third, and one-fifth, respectively, having evidence of type 1 diabetes. Rodriguez et al. (abstract 262) further analyzed 2,436 participants in the study, finding, among children age 2–10 years with diabetes, prevalence rates for metabolic syndrome of 9–11% in non-Hispanic whites, African Americans, and Hispanics, 18% among Asian/Pacific Islanders, and 0% among American Indians. At age 10–19 years, however, the respective prevalences were 15, 28, 34, 35, and 70%. Furthermore, Liu et al. (abstract 1875) reported from the study that 95% of non-Hispanic white, 100% of black, and 91% of Hispanic type 2 diabetic children were overweight or obese, while this was seen in 33, 56, and 47% of the respective type 1 diabetic children.

Rewers et al. (abstract 258) presented data from the SEARCH for Diabetes in Youth Study on 831 children with new-onset diabetes, with 57% hospitalized at diagnosis and ketoacidosis present in 23%, 36% of those <5 years of age vs. 16.0% of those >14 years of age, with lower parental income and education associated with greater risk of ketoacidosis. Edge et al. (abstract 259) compared 43 cases of cerebral edema occurring during treatment of ketoacidosis with 169 control subjects and found an association with lower pH and plasma sodium and higher plasma potassium at presentation, with greater fluid volumes given in the first 4 h, with insulin given in the 1st h of fluid treatment, and with a more rapid fall in plasma potassium levels; rates of change of glucose and sodium levels were not greater in affected patients.

Williams (abstract 1026) reported that of persons with apparent type 2 diabetes who had been diagnosed within 2 years in Helsinki, Belfast, and Barcelona, 7, 8, and 12%, respectively, were GAD positive, with mean age 3–7 years younger and BMI 2–5 kg/m^2 lower than GAD-negative patients, and with 39–75 vs. 10–25% requiring insulin.

Islet transplantation

Scharp et al. (abstract 194) reported a method of 25- to 50-μm-thick polyethylene glycol encapsulation of each transplanted islet, with subcutaneous implantation in streptozotocin-induced diabetic baboons showing maintenance of near euglycemia at 3 months in five of eight animals treated with cyclosporine

for 30 days and subsequently with metformin, and continued insulin independence for 14–20 months in three of the animals. Crutchlow et al. (abstract 100) administered exenatide to streptozotocin-induced diabetic mice, showing prolongation of islet function to 17 vs. 12 days in vehicle-treated animals. β-Cell proliferation and islet vascularization could not be shown to be different, suggesting an effect on apoptosis. The treated mice unexpectedly displayed severe hypoglycemia, a caveat for adaptation of this approach to human islet cell transplantation. Zhang et al. (abstract 2025) studied diabetic mice with or without administration of C-statin, a potent angiogenic inhibitor, showing that impaired islet vascularization reduced functional islet mass following transplantation. Contreras et al. (abstract 196) described a protocol transplanting islets into a subcutaneous omental pouch with the angiogenic factors vascular endothelial growth factor and fibroblast growth factor to promote islet revascularization and allow insulin to enter the portal vein, thereby allowing a lower islet dose and producing greater arginine-stimulated insulin release than seen with intrahepatic transplantation.

Fiorina et al. (abstract 324) examined 15 type 1 diabetic persons 2 years after renal transplantation using cyclosporine, mycophenolate mofetil, and steroids, which were tapered after 6 months. Fasting C-peptide increased from 0.14 at baseline to 0.27 ng/ml at 2 years, with a decrease in the insulin requirement from 57 to 47 units daily, suggesting that over the long-term, improvement can be seen in islet function, presumably reflecting amelioration of the autoimmune disease directed against islets and, perhaps, resumption of islet proliferation.

Fung et al. (abstract 347) reported outcome among 46 type 1 diabetic patients followed for a mean of 19 months and undergoing intensive insulin therapy while awaiting islet transplantation, with 28 islet transfusions performed in 15 patients. Five patients became completely insulin independent, complicated by two episodes of partial portal vein thrombosis and one febrile neutropenia, with similar quality of life and degrees of retinopathy, nephropathy, neuropathy, and carotid intimal thickness in either group. Maffi et al. (abstract 350) reported post–islet transplant renal function in 18 persons with type 1 diabetes receiving daclizumab and FK506, with sirolimus in 14 and micofenolate mofetil in 4. Two developed progressive deterioration of renal function at 2 months, failing to respond to withdrawal of FK506 and sirolimus; in the remaining patients, the creatinine increased from 1.0 to 1.3 mg/dl from 3 to 12 months (not statistically significant). Fiorina et al. (abstract 351) described neurological function in 36 type 1 diabetic persons following islet transplantation, 12 insulin-independent long-term and an additional 12 with sustained C-peptide secretion, whose exogenous insulin requirement decreased, with reduction in A1C and improvement in sural nerve conduction. Haller et al. (abstract 2019) treated a 5-year-old child, who had type 1 diabetes for 6 months, with 4×10^6 total nucleated cells/kg saved umbilical cord blood (with only 11% viability), reporting prolongation of the "honeymoon phase" of low (0.15–2 units/kg) insulin requirement and preservation of C-peptide secretion 12 months after the infusion, suggesting an important area for research in the treatment of recent-onset type 1 diabetes. Hong-McAtee et al. (abstract 2029) studied >1 year of 16 successful islet-transplanted type 1 diabetic patients, with hOKT3γone (Ala-Ala) or anti–thymocyte globulin + etanercept immunosuppression induction, and with sirolimus or everolimus and low-dose calcineurin inhibitors and/or mycophenolate mofetil maintenance. Eleven were insulin independent at 1 year, 4 of whom subsequently required insulin at 16–30 months, with no clear predictive patient or transplant characteristics determining the likelihood of long-term insulin independence.

Not all investigators report positive experience with islet transplantation. Gillard et al. (abstract 352) performed islet cell transplantation in 23 type 1 diabetic persons using anti–thymocyte globulin induction and maintenance treatment with tacrolimus and micofenolate mofetil. The adverse effects included one case of cytomegalovirus hepatitis that required parenteral ganciclovir, one case of cerebellar ataxia that responded to reduction in tacrolimus, and two cases of gastroenteritis that required parenteral hydration. Ten subjects developed gastroesophageal reflux, 13 developed muscle cramps, 22 experienced weight loss, 21 had elevated transaminases, with doubling of the median alanine transaminase, and 5 had anemia requiring transfusion during the 1st week following transplantation.

David Harlan (Bethesda, MD) reviewed the arguments for caution in islet transplantation. He noted that despite great progress, with 90% of transplanted persons insulin independent at 1 year, there are important limitations due to imperfections of current immunosuppressive approaches, including difficulties monitoring anti-islet immune responses and islet mass/function, and difficulties with islet supply and quality assessment, leading him to suggest that appropriate risk-benefit analysis be carried out to identify suitable candidates. "The number one problem," he commented, "is that the immunological process that kills the β-cell is not completely understood." He compared whole-organ pancreas replacement with islets, noting that from a number of viewpoints one can conclude that the whole-organ transplant is superior to transplantation of isolated islets. The whole organ, although a technically demanding operation, offers islets that immediately fully regain vascular supply, with one donor organ per recipient reliably restoring euglycemia, leading to excellent and longstanding glycemic control. Although isolated islet transplantation is easier for patients, there is difficulty with islet isolation; two or more donors are typically required per recipient, and glycemic control may not be as good. Considering the issue of autoimmunity, Harlan reviewed Sutherland's studies of four distal pancreas donations for transplantation into from identical twins discordant for diabetes (19). Since the donor was immunologically identical, immunosuppression was reasoned to be unnecessary. The first patient had had a successful kidney transplant from the same twin, but the transplanted distal pancreas developed islitis, leading to complete islet destruction within 5 weeks. The next patient developed positive islet antibodies and insulitis, and the graft was rejected in 6 weeks. The third patient also had recurrence of type 1 diabetes with insulitis and graft function lost within 12 weeks. Finally, the fourth patient received immunosuppression and remained insulin independent for 38 months. The fact that the autoimmune disease recurs rapidly upon reexposure of the patient to islets poses a dilemma: Is it appropriate to give permanent immunosuppression for a procedure that is in essence being recommended to many persons with type 1 diabetes to improve quality of life? In addition to recurrent autoimmunity and graft rejection, poor vascularization in the liver and poor islet

quality are important potential causes of islet transplant failure.

Immunosuppressive drugs are far from benign. The Edmonton experience shows creatinine clearance decreasing significantly from 98 to 92, 85, and 61 ml/min at 1, 2, 3, and 4 years, respectively. Harlan noted that renal insufficiency is a strong mortality predictor (20). He recalled the experience almost 2 decades ago that cyclosporin treatment early in the course of type 1 diabetes, while promoting improved islet function, was associated with the development of renal insufficiency, with a decrease in renal function by ~25% at 1 year (21). We need, then, to develop safe and effective ways to prevent islet rejection before recommending this for persons with type 1 diabetes who do not otherwise require immunosuppression. Potential approaches include the development of T-cell costimulatory pathway–modifying reagents, of agents interfering with T-cell trafficking, of Jak/Stat pathway modifiers to modify T-cell signaling/function, of strategies promoting immune regulation, of T-cell depletion, and of local immunosuppressive/barrier methods. Harlan warned of the consequences of immune system manipulation. Anti-CD154, which has been proposed for diabetes treatment (22), has been associated with thromboembolic complications, and anti–VLA-4 antibody treatment, which appeared highly promising as a treatment for multiple sclerosis, was found to cause progressive multifocal leukoencephalopathy. After we solve the immune problem, an important issue is limited supply, with perhaps 6,000 organ donors per year, of which 2,000 are used for pancreas transplants, allowing possibly 1,000 islet transplants per year, for ~1 million persons with type 1 diabetes, with Harlan remarking, "Why should we limit it to patients with type 1 diabetes?"

Risk-benefit analysis must include the great recent improvements in outcome for persons with type 1 diabetes based on results of the Diabetes Control and Complications Trial and Epidemiology of Diabetes Intervention and Complications studies, on new insulin preparations, and on improved blood pressure and lipid treatment approaches. Analysis from the University of Pittsburgh database shows a 20-year mortality of ~20% for persons developing type 1 diabetes in 1965–1969 and of ~10% for those diagnosed in 1975–1979 (23), with two Scandinavian articles suggesting similar improvement in outcome. It is not certain whether whole-organ pancreas transplantation improves mortality. Analysis of the United Network for Organ Sharing database, following persons who were on the waiting list and did or did not have transplant, showed worse survival for the first 90 days, with subsequent improvement in survival for persons having simultaneous pancreas and kidney transplantation but no improvement in survival for pancreas transplantation alone or following kidney transplantation (24), leading Harlan to conclude that kidney function is essential but that it is uncertain as to whether pancreas transplantation increases longevity. As far as quality of life, he noted that the Diabetes Control and Complications Trial results are not relevant, as transplant patients usually have had diabetes for >20 years and have existing retinopathy and neuropathy, which would not be likely to improve. Furthermore, in the Edmonton protocol, there was only 13% insulin independence at 4 years, so it is not clear that long-term benefit can be assumed, with the evidence that renal function worsens certainly arguing against benefit in improving nephropathy.

Who is a suitable candidate? Harlan suggested that persons with diabetes and renal failure are probably good candidates for islet transplants around the time of renal transplantation. Another criterion that has been proposed is of persons experiencing frequent hyper- or hypoglycemia, but Harlan contended that such patients are rarely seen, and, when encountered, typically are found to have complex underlying psychosocial issues, with hypoglycemia unawareness often greatly improved simply by meticulous avoidance of episodes.

Harlan concluded with Osler's 1907 caveat regarding "the limits of justifiable experimentation upon our fellow creatures," suggesting that "absolute safety and full consent are the conditions which make such tests allowable. We have no right to use patients entrusted to our care for the purpose of experimentation unless direct benefit to the individual is likely to follow." He suggested that "we're just not smart enough yet" to know which persons will be harmed by the cytotoxic treatment required for a transplant and that given the potential long life expectancy of many persons with type 1 diabetes, he did not believe that the risk of such an approach is justified.

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