Improving epinephrine responses in hypoglycemia unawareness with real-time continuous glucose monitoring in adolescents with type 1 diabetes

RUNNING TITLE: Improving hypoglycemia unawareness in type 1 diabetes

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Objective. To determine whether real-time continuous glucose monitoring (CGM), with preset alarms at specific glucose levels would prove a useful tool to achieve avoidance of hypoglycemia and improve the counterregulatory response to hypoglycemia in adolescents with type 1 diabetes with hypoglycemia unawareness.

Research design and methods. Adolescents with type 1 diabetes with hypoglycemia unawareness underwent hyperinsulinemic hypoglycemic clamp studies at baseline to determine their counterregulatory hormone responses to hypoglycemia. Subjects were then randomised to either standard therapy or real-time CGM for 4 weeks. The clamp study was then repeated.

Results. The epinephrine response during hypoglycemia after the intervention was greater in the CGM group compared to standard therapy.

Conclusions. A greater epinephrine response during hypoglycemia suggests that real-time CGM is a useful clinical tool to improve hypoglycemia unawareness in adolescents with type 1 diabetes.

Hypoglycemia unawareness is defined as the onset of neuroglycopenia before autonomic activation (1). Patients have defective symptomatic and counterregulatory responses, in particular impaired epinephrine response to hypoglycemia. Both defective counterregulatory responses and hypoglycemia unawareness constitute the hypoglycemia-associated autonomic failure (HAAF) associated with recurrent iatrogenic hypoglycemia (2-4).

In adults, it has been demonstrated that as little as 2-3 weeks of avoidance of hypoglycemia reverses hypoglycemia unawareness and improves the attenuated epinephrine component of defective counterregulation in affected patients (5-7). Although strict avoidance of hypoglycemia can restore autonomic symptoms of hypoglycemia and improve counterregulatory responses to hypoglycemia, this is difficult to achieve in practice. Real-time continuous glucose monitoring (CGM) allows patients to view their blood glucose levels almost instantaneously and offers potential to reduce hypoglycemia frequency. This study was designed to determine whether real-time CGM with preset alarms at specific glucose levels would prove a useful tool to achieve avoidance of hypoglycemia and therefore improve the counterregulatory response to hypoglycemia in adolescents with type 1 diabetes with hypoglycemia unawareness.

RESEARCH DESIGN AND METHODS
Adolescents with type 1 diabetes aged 12-18 years with hypoglycemia unawareness attending Princess Margaret Hospital diabetes clinics were invited to participate. Hypoglycemia unawareness score was determined by the use of modified Clarke’s questionnaire (8). This questionnaire has been shown to accurately identify patients with impaired awareness of hypoglycemia.
Improving hypoglycemia unawareness in type 1 diabetes

for both clinical and research purposes (9). A score of $\geq 8$ is suggestive of hypoglycemia unawareness. Consent was obtained for all participants. All subjects underwent a hyperinsulinemic hypoglycemic clamp study at baseline to assess hypoglycemic symptoms and hormonal responses. Subjects were then randomised to either standard therapy (standard group) or to use real-time (Medtronic Minimed Paradigm® REAL-Time System) CGM (CGM group) for 4 weeks. At the end of the 4-week period, all patients underwent a repeat hypoglycemic clamp study.

**Hyperinsulinemic hypoglycemic clamp.** During this procedure, the antecubital vein was cannulated for insulin and glucose infusion and blood was sampled from the contralateral hand vein placed in a box heated to 60°C. Regular insulin (Human Actrapid; Novo Nordisk, Crawley, UK) was infused at a constant rate of 80mU/m²/min. Target plasma glucose levels were achieved by adjusting the rate of infusion of 20% glucose in water. Plasma glucose concentrations were maintained initially at euglycemia (5-6mmol/L) over a period of 1 hour. Following this, blood glucose was lowered over 30 minutes to a nadir of 2.8mmol/L. The blood glucose concentration of 2.8mmol/L was maintained for 40 minutes for the hypoglycemia phase. Euglycemia was then restored.

For the duration of the clamp procedure, blood glucose was analysed at the bedside using a glucose oxidase technique (Yellow Springs Instruments 2300, Yellow Springs, OH, USA). Additional samples of arterialised venous blood was taken to measure plasma insulin, glucagon, epinephrine, norepinephrine, cortisol and growth hormone concentrations.

**Study intervention.** Following the first hypoglycemic clamp study, both groups were advised to strictly avoid hypoglycemia with fingerstick testing at least 4-6 times daily to maintain blood glucose levels between 6-10mmol/L for the 4 week period. In addition, the CGM group wore real-time CGM with subcutaneous sensor with preset low alarms at 6mmol/L and advised to institute standard hypoglycemia treatment for blood glucose levels below 6mmol/L with target blood glucose level of 8mmol/L.

The CGM Group received an additional 2 hours’ instructions regarding sensor insertion and usage. Sensors were changed every 3 days.

**Outcome measures.** The major outcome measure was the epinephrine response to hypoglycemia measured during the hypoglycemia clamp study. Plasma epinephrine levels were measured by ELISA (Diagnostika GMBH, Hamburg, Germany) and samples were analysed in duplicate. The interassay coefficient of variation at 10pmol/L and 5460pmol/L were 2% and 5.5%, respectively.

**RESULTS**

Eleven subjects were studied, including 5 subjects in the standard group (age 15.0±0.8yrs, A1C 7.9±0.3% since diagnosis, duration 6.5±1.2yrs) and 6 subjects in the CGM group (age 13.8±0.7yrs, A1C 7.7±0.2% since diagnosis, duration 5.2±1.4yrs).

At baseline the epinephrine response to hypoglycemia was blunted and there was no difference between subjects randomised to standard or CGM groups (percentage change 288±151% vs. 214±72%, standard vs. CGM group respectively, p=0.688). Following the intervention, there was a greater epinephrine response in the CGM group.
Improving hypoglycemia unawareness in type 1 diabetes

(percentage change 114±83 vs. 604±234, standard vs. CGM group, respectively p=0.048). This represents a greater percentage rise in epinephrine concentrations during hypoglycemia following therapy in the CGM group (p=0.375 vs. 0.031, standard vs. CGM group, respectively) as shown in Figure 1. Peak adrenaline response during hypoglycemia after the intervention was also greater in the CGM group compared to the standard group (1093±221pmol/L vs. 572±162pmol/L, p=0.048). Subjects in the CGM group reported higher adrenergic symptoms scores after the intervention compared to standard group (5.4±0.4 vs. 3.4±0.2, p<0.001).

The mean A1C at baseline was 7.9±0.3% for both groups. Following the intervention, there was no deterioration in glycaemic control in the standard or CGM group (A1C 7.9±0.4% vs. 8.3±0.3%, p=0.587).

The glucagon response was absent at baseline and after intervention in both groups. There was no change in cortisol and growth hormone responses to hypoglycemia for both groups.

DISCUSSION

The epinephrine response to hypoglycemia in patients with type 1 diabetes with hypoglycemia unawareness was greater following the use of real-time CGM with low glucose alarms compared to standard medical therapy alone. The use of CGM was not associated with deterioration in A1C. This greater epinephrine response during hypoglycemia suggests that real-time CGM is a useful clinical tool to improve hypoglycemia unawareness in adolescents with type 1 diabetes. The high risk of associated severe hypoglycemia requires that hypoglycemia unawareness is recognised and treated. This study demonstrates that blunted counterregulatory responses to hypoglycemia do occur in adolescents with relatively short duration of diabetes. In addition to the blunted epinephrine response, most of these subjects reported no adrenergic symptoms during their baseline hypoglycemic clamp study. A limitation of this study is the sample size. Although small, evaluating counterregulatory response with hypoglycemia clamp studies is a robust method and this technique limits inclusion of a large number of subjects.

Author contributions. T.T.L. wrote manuscript, collected and researched data. J.H. reviewed/edited manuscript, collected data and contributed to study design. R.J.D reviewed/edited manuscript and contributed to study design. E.M.L reviewed/edited manuscript and researched data. E.A.D. contributed to discussion, researched data and reviewed/edited manuscript. T.W.J contributed to study design, researched data and wrote manuscript.

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REFERENCES

1. Cryer PE. Hypoglycaemia: the limiting factor in the glycaemic management of Type I and Type II diabetes. Diabetologia. 2002 Jul;45(7):937-48.
Improving hypoglycemia unawareness in type 1 diabetes

2. Cryer PE. Mechanisms of hypoglycemia-associated autonomic failure and its component syndromes in diabetes. Diabetes. 2005 Dec;54(12):3592-601.
3. Heller SR, Cryer PE. Reduced neuroendocrine and symptomatic responses to subsequent hypoglycemia after 1 episode of hypoglycemia in nondiabetic humans. Diabetes. 1991 Feb;40(2):223-6.
4. Dagogo-Jack SE, Craft S, Cryer PE. Hypoglycemia-associated autonomic failure in insulin-dependent diabetes mellitus. Recent antecedent hypoglycemia reduces autonomic responses to, symptoms of, and defense against subsequent hypoglycemia. J Clin Invest. 1993 Mar;91(3):819-28.
5. Fanelli C, Pampanelli S, Epifano L, Rambotti AM, Di Vincenzo A, Modarelli F, Ciofetta M, Lepore M, Annibale B, Torlone E, Perriello G, De Feo P, Santeusanio F, Brunetti P, Bolli GB. Long-term recovery from unawareness, deficient counterregulation and lack of cognitive dysfunction during hypoglycaemia, following institution of rational, intensive insulin therapy in IDDM. Diabetologia. 1994 Dec;37(12):1265-76.
6. Cranston I, Lomas J, Maran A, Macdonald I, Amiel SA. Restoration of hypoglycaemia awareness in patients with long-duration insulin-dependent diabetes. Lancet. 1994 Jul 30;344(8918):283-7.
7. Dagogo-Jack S, Rattarasarn C, Cryer PE. Reversal of hypoglycemia unawareness, but not defective glucose counterregulation, in IDDM. Diabetes. 1994 Dec;43(12):1426-34.
8. Clarke W, Jones T, Rewers A, Dunger D, Klingensmith GJ. Assessment and management of hypoglycemia in children and adolescents with diabetes. Pediatr Diabetes. 2008 Apr;9(2):165-74.
9. Geddes J, Wright RJ, Zammitt NN, Deary IJ, Frier BM. An evaluation of methods of assessing impaired awareness of hypoglycemia in type 1 diabetes. Diabetes Care. 2007 Jul;30(7):1868-70.

FIGURES

Figure 1 - Change in epinephrine response during hypoglycemia. Data are means±SE.
Figure

Change in Epinephrine Response During Hypoglycemia

- Standard Group
- CGM Group

* p=0.031
p=0.375

Baseline | After 4 weeks

Percentage %