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Total glycosylated haemoglobin (HbA_1) levels in diabetic children

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SUMMARY Total glycosylated haemoglobin (HbA_1) levels from capillary blood were studied retrospectively during a 1-year period in 148 diabetic children aged between 1·9 and 16·8 years. The clinic range for HbA_1 was 6·7 to 22·2% and the results were normally distributed (mean ± SD 13·2 ± 2·8%). The normal range in non-diabetics using this method is 4·9 to 8·0%. Results from children who had had diabetes for more than 5 years were higher than those from children with diabetes of 2 to 5 years' duration. Girls had higher average values during the 1-year period than boys. HbA_1 measurement serves to identify the deficiencies of current diabetic treatment regimens. It also has more immediate practical benefits in focusing attention on children whose control is deteriorating.

Interest in diabetic management has intensified lately as evidence has increased that good glucose control can reduce microvascular complications.[1–3] In the past accurate assessment of control has not been possible. It is not sufficient to rely solely on clinical well-being and normal growth; random tests of blood sugar in the diabetic clinic are of little help because the normal day-to-day routine will have been interupted, and the time at which such tests are taken will vary. Urine tests remain the cornerstone of control, but there are obvious pitfalls promoting inappropriate insulin adjustments.

Glycosylation of HbA appears to be a slow, non-enzymatic process taking place during the life of the red cell. Glycosylated haemoglobin (HbA_1) levels reflect the integrated blood glucose level during the preceding 2 months.[4 5] They are an objective measurement being independent of patient co-operation, time of day, insulin administration, or meals, and their role in the management of the diabetic patient has recently been reviewed.[6] Ion exchange chromatography is one method used to separate HbA_1 from HbA, so that the percentage of HbA_1 can be calculated.[7] At the children's diabetic clinic in Nottingham the patients are seen at 3- to 4-monthly intervals, the capillary HbA_1 and blood glucose being measured at each outpatient visit. We have examined HbA_1 results during one year, in groups of children according to duration of diabetes, gender, and presence or absence of remission period. The remission period is regarded as the period during which the child requires no more than 0·3 units of insulin per kg body weight a day to remain free of glycosuria. The way in which HbA_1 results can alter individual patient management is discussed.

Methods

All the results from 148 diabetic children collected between December 1979 and November 1980 were analysed retrospectively. The children were juvenile onset (type 1) diabetics of more than 3 months' duration, attending the children's diabetic clinic at the University Hospital, Nottingham. Capillary blood for HbA_1 and blood glucose measurement was taken from fingerpricks (0·1 ml) into sequestrene and fluoride bottles at each outpatient visit.

All children were receiving insulin, most of them being on a twice-daily regimen of short and intermediate acting pork insulin—Velosulin and Insulatard*, or Actrapid and Monotard†. Parents and children were instructed to give insulin 20–30 minutes before meals, and 50–70% of the total daily dose of insulin was given in the morning. The proportions of short and intermediate acting insulins were adjusted in each child in an attempt to provide optimal control. All children were reviewed by the dietician, and advice based on a controlled carbohydrate intake was given. In most children control is based on a minimum of twice-daily Clinitest (2-drop method) measurement of glycosuria. An increasing proportion of children have been introduced to home blood sugar measurement as an alternative. HbA_1 determination was introduced as a routine clinic procedure on capillary blood in

* Nordisk, † Novo.

1979. Attempts are made to achieve the best diabetic control possible using a structured education programme and extensive home visiting by a diabetic health visitor. Particular care is given to avoid excessive insulin dosage of more than 1 unit/kg a day because of concern with induction of Somogyi effect[8] or nocturnal hypoglycaemia.[9]

Sequestrene samples were stored at 4°C before the weekly measurement of HbA_1, using ion exchange chromatography (Bio-Rad column test). A small amount of whole blood was mixed with a haemolysis reagent to lyse the red cells and free the haemoglobin. An aliquot of the haemoglobin was then applied to a weakly acidic cation exchange resin in a disposable column. An elution/developing reagent was added and separation of the 'fast' moving HbA_1 from the remainder occurred. A spectrophotometer compared the concentrations of glycosylate with total haemoglobins to enable the percentage of HbA_1 to be calculated. Maximal within run and run-run coefficients of variations for these were 3·8% and 7% respectively. These values are those obtained under optimal conditions in a temperature controlled room with one person carrying out the assays. The mean of HbA_1 levels in 129 non-diabetic women and men for this method is 6·5% (95% confidence limits 4·9–8·0%).

Results

One hundred and forty-eight patients (59 girls and 89 boys) had 467 HbA_1 measurements during the year. All results are expressed as mean ± standard deviation unless stated otherwise. The age range of the children ranged from 1·9 to 16·8 (mean 11·2 ± 3·5) years, and duration of diabetes ranged from 3 months to 13·4 years (mean 4·9 ± 3·3 years). The mean insulin dose was 0·82 ± 0·23 unit/kg a day.

The mean HbA_1 of all results for the year was 13·2 ± 2·8% (range 6·2–21·8) and the data were distributed normally with only 7 (1·5%) values falling within the normal range for non-diabetic patients (Fig. 1). The children's mean HbA_1 was 13·1 ± 2·1% (using each child's average HbA_1 throughout the year). Statistical analysis was carried out using Student's unpaired *t* test. The HbA_1 results were analysed according to duration of diabetes (Table 1). There was no significant difference between HbA_1 results of the 0–2 and 2–5 year duration groups. HbA_1 results from patients with diabetes of more than 5 years' duration were higher than both the 2–5 ($P<0·05$) and the 0–2 year group ($P<0·01$).

Seven (4·7%) patients were in remission (honeymoon) phase of the diabetes. In each the average of all values of HbA_1 during remission was obtained (10·8 ± 1·4%) and compared with the average

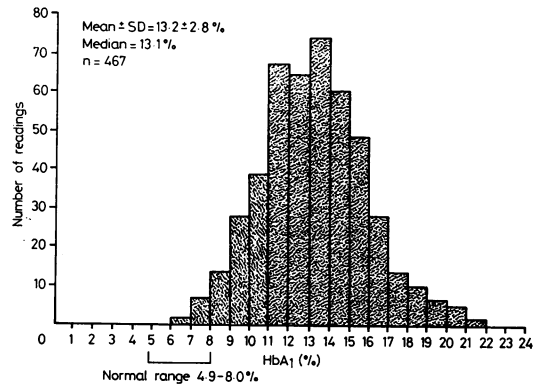

Fig. 1 *HbA_1 values during one year from the children's diabetic clinic.*

Table 1 *HbA_1 values from 467 children of different duration of diabetes*

Duration (years)	*Percentage of children*	*HbA_1 (%) (mean ± SD)*
<2	27·4	12·8 ± 2·7
2–5	34·9	13·0 ± 2·8
>5	37·7	13·7 ± 2·6

HbA_1 over the year of all patients out of remission (13·2 ± 2·1%). The HbA_1 levels in remission were significantly lower than the rest ($P<0·01$) (Table 2). A sex difference was present between HbA_1 levels of boys (12·8 ± 2·3%) and girls (13·6 ± 1·9%) compared with the patients' average HbA_1 throughout the year ($P<0·05$). Table 3 shows that this sex difference is present only in children older than 12 years.

There was a weak correlation between HbA_1 levels and clinic blood glucose (correlation coefficient 0·23). High (20–25 mmol/l) clinic blood glucose levels were more common in children with high

Table 2 *Mean HbA_1 results for each patient comparing remission and non-remission phase of diabetes*

	Insulin dosage (U/kg per day)	*HbA_1 (%) (mean ± SD)*
Remission (n = 7)	0·25 ± 0·07	10·8 ± 1·4
Post remission (n = 141)	0·83 ± 0·26	13·2 ± 2·1
		$P<0·01$

Table 3 *Average HbA_1 values for each child over a 1-year period for boys and girls (n = 148) above and below 12 years old*

Age (years)	*Boys*		*Girls*	
	HbA_1(%) mean	*%*	*HbA_1(%) mean*	*%*
< 12	12·4 ± 2·0	33	12·4 ± 1·8	16
> 12	13·4 ± 2·3	28	14·3 ± 2·0	23

HbA_1 levels but could occur in those with more acceptable HbA_1 values.

Discussion

In diabetic children HbA_1 levels have been well correlated with other methods of assessing metabolic control—such as home blood glucose measurements, 24-hour urine glucose excretion, and height velocity.[10 11] The prevailing blood glucose concentration of the preceding 4–12 weeks appears to be the main factor to affect the level of HbA_1,[12 13] although rapid increases in HbA_1 level may occur in 2–4 weeks during a period of poor control—that is HbA_1 level is more sensitive to 'sin than repentance'. Some conditions which affect red cell life span are known to lead to inaccurate results—haemolytic anaemias, haemoglobinopathies, and uraemia.[14 15] Capillary samples can be used for HbA_1 measurement and provide a more legitimate reason for routine fingerpricks than clinic blood glucose analysis. The overall HbA_1 results from our clinic with a mean of 13·2%, which is just over twice the non-diabetic mean (6·5%), provide a sobering view of our achievements in a clinic where the importance of good control is stressed. It is tempting to accept this disappointing figure as merely further confirmation of the deficiencies of currently available insulin therapy. However, it is important to bear in mind that many factors influence the success of total diabetic management, and at least some may be amenable to more intensive supervision. Each diabetic child and family must be tackled as a separate problem often needing the resources of overtaxed home visiting nurses. In an individual child patterns of HbA_1 values can be useful in deciding on priority use of this limited staff. Sometimes a satisfactory HbA_1 value contradicts the impression of poor control based on a summary review of a misleading catalogue of erratic urine tests. By contrast a high HbA_1 can bring reality to a child or his parents if they or the doctor are being deceived by fictitious results.[16 17] Such revelations must be handled carefully in case the child is forced into an even more defensive posture. Fig. 2 summarises our approach to a child with high HbA_1 values.

Our data have confirmed that patients have low HbA_1 in the remission phase of their diabetes. We have not routinely measured HbA_1 levels in newly diagnosed diabetics as they tend not to produce helpful information other than correlating with the degree of hyperglycaemia at the time.[18] Daneman *et al.*[19] found that HbA_1 levels fell to a nadir between 3 weeks and 6 months after diagnosis and this corresponded to the remission phase of diabetes when insulin requirements are low due to some recovery of β cell function. We have also shown that HbA_1 levels are higher in patients of more than 5 years' duration and this corresponds to the progressive failure of endogenous insulin production. Others have confirmed this loss of β cell function by demonstrating failure of C-peptide release.[20 21] The

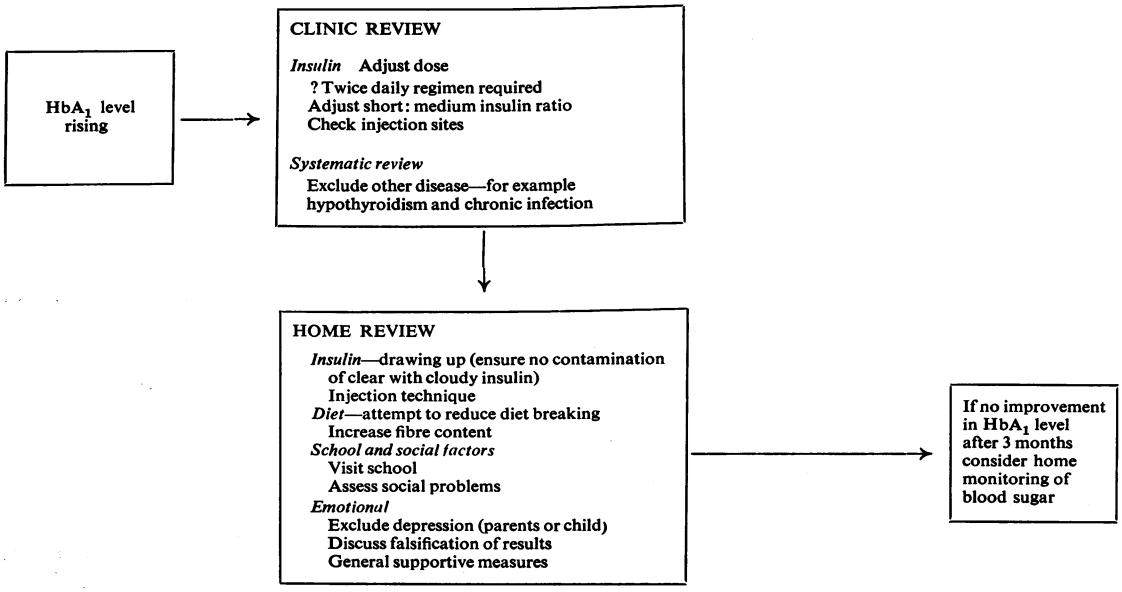

Fig. 2 *Our approach to a child with rising HbA_1 values.*

fact that levels of HbA_1 are higher in girls than boys is due to the higher results of girls over 12 years old. Factors during adolescence—such as teenage rebellion, deterioration of control around menstruation, increasing over-insulinisation, or emotional problems—are the likely events. Tattersall and Lowe[22] have recently reviewed the management problems of the adolescent diabetic and stressed the need for help with emotional problems as well as with diabetic control.

The reporting of HbA_1 values will enable clinicians to judge their own success and that of others in developing methods of diabetic management. This objective comparison is especially important while we assess the benefits of new techniques which are often expensive, time consuming, and demanding of children and their families. Regular home blood glucose measurement is an innovation which needs such analysis. In making these comparisons it is important to take into account the differing methodology for the measurement of glycosylated haemoglobin. It may indeed be more valid to report HbA_1 results as a ratio of the non-diabetic mean.

There can be little doubt that HbA_1 determination, especially on capillary blood samples, if available, has earned a place in regular monitoring of diabetic control. It provides for objective assessment in both the clinic and the home. Those responsible for diabetic clinics must search for ways to improve control but there are inevitable obstacles which cannot be overcome by mere manipulations of the insulin regimen. There is, indeed, a danger that clinicians will be even more tempted to use inappropriately high insulin dosages preventing all the problems of the so-called Somogyi phenomenon.[8 23] Parents of diabetic children are rapidly learning the significance of HbA_1, and it is a wise precaution to ensure that they do not see a normal value as a necessary goal. Such an unrealistic target can rapidly promote frustration, despair, and disenchantment. They should look on it as one of a succession of developments which will perhaps revolutionise diabetic management in their child's lifetime.

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References

[1] Drash A. The control of diabetes mellitus. Is it achievable? Is it desirable? *J Pediatr* 1976; **88:** 1074–6.

[2] Cahill G F, Jr, Etzwiler D D, Freinkel N. 'Control' and diabetes. *N Engl J Med* 1976; **294:** 1004–5.

[3] Siperstein M D, Foster D W, Knowles H C, Jr, Levine R, Madison L L, Roth J. Control of blood glucose and diabetic vascular disease. *N Engl J Med* 1975; **296:** 1060–3.

[4] Gabbay K H, Hasty K, Breslow J L, Ellison R C, Bunn H F, Gallop P M. Glycosylated hemoglobins and long-term blood glucose control in diabetes mellitus. *J Clin Endocrinol Metab* 1977; **44:** 859–64.

[5] Gonen B, Rubenstein A H. Haemoglobin A_1 and diabetes mellitus. *Diabetologia* 1978; **15:** 1–8.

[6] Anonymous. Haemoglobin A_1 and diabetes: a reappraisal. *Br Med J* 1980; **281:** 1304–5.

[7] Welch S G, Boucher B J. A rapid micro-scale method for the measurement of haemoglobin A_1 (a+b+c). *Diabetologia* 1978; **14:** 209–11.

[8] Somogyi M. Exacerbation of diabetes by excess insulin action. *Am J Med* 1959; **26:** 169–91.

[9] Gale E A M, Tattersall R B. Unrecognised nocturnal hypoglycaemia in insulin-treated diabetics. *Lancet* 1979; **i:** 1049–52.

[10] Williams M L, Savage D C L. Glycosylated haemoglobin levels in children with diabetes mellitus. *Arch Dis Child* 1979; **54:** 295–8.

[11] Paisey R B, Macfarlane D G, Sheriff R J, Hartog M, Slade R R, White D A J. The relationship between blood glycosylated haemoglobin and home capillary blood glucose levels in diabetics. *Diabetologia* 1980; **19:** 31–4.

[12] Dunn P J, Cole R A, Soeldner J S, *et al.* Temporal relationship of glycosylated haemoglobin concentrations to glucose control in diabetes. *Diabetologia* 1979; **17:** 213–20.

[13] Leslie R D G, Pyke D A, John P N, White J M. How quickly can haemoglobin A_1 increase? *Br Med J* 1979; **ii:** 19.

[14] Brooks A P, Metcalfe J, Day J L, Edwards M S. Letter: Iron deficiency and glycosylated haemoglobin A_1. *Lancet* 1980; **ii:** 141.

[15] Bunn H F, Shapiro R, McManus M, *et al.* Structural heterogeneity of human hemoglobin A due to non-enzymatic glycosylation. *J Biol Chem* 1979; **254:** 3892–8.

[16] Goldstein D E, Walker B, Sharon S, *et al.* Hemoglobin A_{1c} levels in children and adolescents with diabetes mellitus. *Diabetes Care* 1980; **3:** 503–7.

[17] Belmonte M M, Gunn T, Gonthier M. The problem of 'cheating' in the diabetic child and adolescent. *Diabetes Care* 1981; **4:** 116–20.

[18] Heinze E, Kohne E, Meissner C, Beischer W, Teller W M, Kleihauer E. Hemoglobin A_{1c} (HbA_{1c}) in children with long standing and newly diagnosed diabetes mellitus. *Acta Paediatr Scand* 1979; **68:** 609–12.

[19] Daneman D, Tsalikian E, Hengstenberg F, Becker D J, Drash A L. Glycosylated haemoglobin in children with insulin-dependent diabetes mellitus. *Diabetologia* 1980; **19:** 423–6.

[20] Ludwigsson J, Heding L G. Beta cell function in children with diabetes. *Diabetes* 1978; **27:** 230–4.

[21] Grajwer L A, Pildes R S, Horwitz D L, Rubenstein A H. Control of juvenile diabetes mellitus and its relationship to endogenous insulin secretion as measured by C-peptide immunoreactivity. *J Pediatr* 1977; **90:** 42–8.

[22] Tattersall R B, Lowe J. Diabetes in adolescence. *Diabetologia* 1981; **20:** 517–23.

[23] Gale E A M, Kurtz A B, Tattersall R B. In search of the Somogyi effect. *Lancet* 1980; **ii:** 279–82.

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