The reaction of glucose with the amino group of many proteins occurs normally in all individuals. The reaction of glucose with haemoglobin produces glycohaemoglobin compounds. The most important form of the glycohaemoglobin compounds is haemoglobin A1c (HbA1c). HbA1c is formed when glucose is bound to the beta-chain of a haemoglobin molecule. Once glucose is bound this way, it is no longer available for metabolism and therefore accumulates in the red blood cells for the life-span of the red cell. The rate of HbA1c accumulation is directly related to the level of glycaemia and the life-span of red cells which averages 120 days. Therefore HbA1c concentration is an index of integrated value for glucose over the preceding two to three months. This index is not affected by wide diurnal glucose variation, exercise or food intake.

Two large, prospective, randomized clinical trials demonstrated a strong relationship between hyperglycaemia and the development of microvascular complications of diabetes. HbA1c was measured in both studies i.e. Diabetes Control and Complications Trial (DCCT) and the United Kingdom Prospective Diabetes Study (UKPDS), to assess glycaemic control in type 1 and type 2 diabetes, respectively. Both studies established a direct relationship between the HbA1c and the risk of complications (Fig.1).
The findings of these studies set the stage for American Diabetes Association to recommend that a primary treatment goal in adults with diabetes should be near-normal glycaemia with HbA1c < 7% only when measured by the method used for the DCCT and UKPDS studies.

HbA1c MEASUREMENT

There are currently more than 30 HbA1c assay methods in use. Potentially, reported HbA1c from the same blood sample could differ considerably among these methods unless they are standardised to a common reference e.g. without standardisation, the same blood sample could be reported as 7% in one laboratory and 9% in another.

Both the DCCT and UKPDS used the same method for HbA1c measurement. From a clinical point of view, it is essential that HbA1c results be traceable to the DCCT/UKPDS results where the relationships to risk for vascular complications have been established to ensure the same clinical benefits to patients.

In 1996, the National Glycohemoglobin Standardisation Programme (NGSP) was initiated to standardise HbA1c results among laboratories worldwide to DCCT-equivalent values. The NGSP website http://www.ngsp.org gives detailed information regarding those Laboratories worldwide which participate on their certification programme to ensure that the HbA1c methods in use continue to produce DCCT-equivalent results.

HbA1c AS A DIAGNOSTIC TOOL

HbA1c is well established as an integral part in the management of patients with diabetes mellitus. The recommendation that HbA1c must be measured at least every six months in patients with established type 1 and type 2 diabetes has had a significant impact in the use of this test.

In 2011, WHO recommended that HbA1c can be used as a diagnostic test provided that stringent quality assurance requirements are met and the assay is standardised to a criteria aligned to the international reference values such as DCCT equivalent values offered by NGSP certified laboratories or methods (as listed on NGSP website). The WHO went further to recommend the diagnostic decision limit of 6.5% for diagnosing diabetes. However, it is important to note that values below 6.5% may not be used to exclude the diagnosis of diabetes.

When HbA1c is used for diagnosis of diabetes, unlike the fasting blood glucose and Oral Glucose Tolerance (OGTT) tests, no prior fasting is necessary as food intake does not affect the test and patient preparation, appointments and special dietary requirements which normally apply to OGTT are not necessary.

HbA1c must never be used as the sole test to diagnose diabetes in the following situations:

- all symptomatic children and young people
- if symptoms are suggestive of Type 1 diabetes in any patient
- if the symptoms suggestive of diabetes are of a short duration
- patients who are at high risk of diabetes and are acutely ill
- patients on medication which may cause rapid glucose rise such as corticosteroids and antipsychotics
- acute pancreatic damage or surgery
- high prevalence of haemoglobin variants which may potentially interfere with the current method in use
- alternative additional diagnostic tools such as fasting and/or random glucose or OGTT are recommended in these situations.
LIMITATIONS OF HbA1c

All HbA1c measurements are only possible if HbA (Adult type haemoglobin) is present. In patients without HbA, no HbA1c will be measurable.

- Recent blood glucose changes will not be reflected on the HbA1c concentration.

- Asplenia and iron deficiency anaemia are associated with a prolonged life-span of red blood cells and therefore more than average accumulated HbA1c in red blood cells.

- Recent blood transfusion or increased erythropoiesis due to haemolysis or blood loss will reduce HbA1c concentration.

- The presence of Hereditary Persistent Foetal Haemoglobin (HPFH) interferes with the measurement of HbA1c because HbF is not glycated at the same rate as HbA.

- Certain methods suffer HbA1c measurement interference in the presence of heterozygous haemoglobin variants such as AS, AC, AE and other rarer types. Boronate affinity chromatography, which suffers interference from HbF, appears to be interference free in the presence of these haemoglobin variants.

REPORTING UNITS FOR HbA1c

Following the DCCT study in 1993, the issue of standardisation became an important objective for scientists and clinicians. The International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) Working Group on Standardisation of HbA1c established a true international reference measurement system for HbA1c and the successful preparation of pure HbA1c calibration material available for manufacturers to standardise their HbA1c test kits. This should reduce significant inter-method and inter-laboratory variability. It has been agreed to report HbA1c using the units of mmol/mol (IFCC) and percentage (NGSP) as it appears in the Lancet Laboratory HbA1c report.

HbA1c is best measured by a method that is traceable or comparable to the method used in the DCCT study. Results produced by this method will be in DCCT-equivalent values and therefore allow the best translation of the conclusions of DCCT/UKPDS studies to benefit the local patients. Lancet Laboratories uses a test method that is comparable to that used in the DCCT study.