

### Matters arising

#### Glycated haemoglobin and glycated protein and glucose concentrations in necropsy blood samples

We read the paper by John *et al*<sup>1</sup> with considerable interest as it confirms the results of similar work conducted and reported by ourselves<sup>2</sup> and others<sup>3</sup> some time ago.

In our experiments we measured glycated haemoglobin (HbA<sub>1c</sub>) by mobile affinity electrophoresis in blood collected from 73 subjects at necropsy. HbA<sub>1c</sub> was measured in preserved and unpreserved specimens and its stability *in vitro* was studied. There was no clinically important change in HbA<sub>1c</sub> concentrations in samples stored for more than 40 days at 4°C. There was poor correlation between glucose and HbA<sub>1c</sub>, and no significant differences between the mean values of HbA<sub>1c</sub> in specimens collected up to 72 or more hours after death from subjects with no previous history of diabetes. The concentrations were within the laboratory reference range; raised concentrations were shown in eight specimens collected from diabetic subjects.

HbA<sub>1c</sub> and glucose were also measured in specimens of blood collected from the subclavian and femoral veins and right atrium (13 subjects) and from the left ventricle (five subjects). There was no clinically important difference in HbA<sub>1c</sub> concentrations collected from different sites in the body even though the glucose concentrations showed some variation, particularly the right atrium (range, 0.4–68.0 mmol/l).

We concluded that HbA<sub>1c</sub> was a more reliable measurement than glucose in necropsy specimens for assessing glycaemic status in known or previously undiagnosed diabetes. While we are pleased that John *et al* have not reported data in their paper which contradicts our observations, we believe a

more thorough search of the published findings may have obviated the need for repeating some of our experiments, leaving more time to pursue new courses of study.

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#### References

- 1 John G, Scott KLM, Hawcroft DM. Glycated haemoglobin and glycated protein and glucose concentrations in necropsy blood specimens. *J Clin Pathol* 1988;41:415–8.
- 2 Hindle EJ, Rostron GM, Gatt JA. The diagnostic value of glycated haemoglobin levels in post-mortem blood. *Ann Clin Biochem* 1985;22:144–7.
- 3 Chen C, Glagov S, Mako M, *et al*. Post-mortem glycosylated hemoglobin (HbA<sub>1c</sub>): Evidence for a history of diabetes mellitus. *Ann Clin Lab Sci* 1983;13:407–10.

Drs Scott and John comment:

We are grateful to Dr Hindle for highlighting his paper. We did not attempt to address the question of *in vitro* specimen stability in either preserved or unpreserved specimens, nor did we attempt to correlate glucose concentration with HbA<sub>1c</sub>, as both topics have been thoroughly investigated by many other workers.

The use of agar electroendosmosis<sup>1,2</sup> and affinity chromatography<sup>3,4</sup> have been widely investigated for the diagnosis of diabetes (assessed by OGTT), and different levels of sensitivity and specificity have been reported. The electroendosmosis and affinity chromatography methods together represent the most widely used methods for estimating glycated haemoglobin in the United Kingdom; the affinity electrophoresis method is

relatively little used (data from the United Kingdom NEQAS for glycated haemoglobin). The precision reported by Dr Hindle for this method is also poorer than that found for the former methods.

Overall, the affinity chromatography method has been found to display the best discrimination between diabetics and normal controls which probably reflects its superior precision intrabatch CV = 1%). We believe that newer, more precise methods should be investigated.

We are interested to read that Dr Hindle *et al* also found that glucose concentration in the right atrium was increased. Unlike ourselves, they did not further investigate the cause for this increase and therefore could draw no conclusions as to the reason for this high glucose concentration.

#### References

- 1 Albutt EC, Nattrass M, Northam BE. Glucose tolerance and glycosylated haemoglobin measurement for the diagnosis of diabetes mellitus, an assessment of the criteria of the WHO Expert Committee on Diabetes Mellitus 1980. *Ann Clin Biochem* 1985;22:67–73.
- 2 Lester E, Frazer AD, Shepherd CA, Woodroffe FJ. Glycosylated haemoglobins as an alternative to the glucose tolerance test for the diagnosis of diabetes mellitus. *Ann Clin Biochem* 1985;22:74–8.
- 3 Hall PM, Cook JGH, Sheldon J, Rutherford SM, Gould BJ. Glycosylated hemoglobin and glycosylated plasma proteins in the diagnosis of diabetes mellitus and impaired glucose tolerance. *Diabetes Care* 1984;7:147–50.
- 4 John WG, Richardson RW. Glycosylated haemoglobin levels in patients referred for oral glucose tolerance tests. *Diabetic Med* 1985;3:46–8.

## Book reviews

**Antibodies.** Vol 1. **A Practical Approach.** Ed D Catty. (Pp 217; £19.) IRL Press. 1988. ISBN 0 947946 85 3.

Many laboratories require a book on modern, general and sometimes specialised immunological techniques. The first volume of this book covers general techniques in a wealth of detail. It is clear that the authors from the Universities of Birmingham and Oxford are extremely experienced in the techniques which they describe. A general introduction is followed by three chapters

antibodies: immunoaffinity chromatography, gel immunodiffusion, immunoelectrophoresis and immunostaining, and haemagglutination and haemolysis. A sound knowledge of immunology is assumed but sufficient detail is provided to enable the inexperienced to master the techniques quickly and an adequate note of caution is given to those considering the production of human monoclonal antibodies. Occasionally it would have been helpful if the rationale of describing techniques for the production of polyclonal antibodies and both murine and human monoclonal antibodies. The last three chapters cover general applications of some techniques were explained, such as

antigen dosage for immunisation. Each chapter is relatively self contained which has occasionally resulted in the description of slightly different techniques to achieve the same objective but this also adds to the versatility of this book. An appendix contains an extensive list of suppliers of specialist items. Indeed, it is a practical approach and, although oriented towards clinical laboratories, it will also be invaluable to anyone using immunologically based techniques and I strongly recommend it. The second volume will contain further general techniques and some specialised ones and will be available early next year.

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