Letters to the Editor

Limitations of capillary plasma immunoreactive insulin assay as an epidemiological tool

Dear Sir,

Capillary blood sampling for biochemical investigation is being used increasingly as the sensitivity of assay techniques improve. A large number of oral glucose tolerance tests are performed in the Coventry Diabetes Study using capillary whole blood [1]. We are therefore able to compare capillary and venous plasma immunoreactive insulin values.

Subjects attended in the morning after an overnight fast and, on arrival, washed their hands with warm water for 2 min. The skin was dried and then punctured using an Autoclix (Boehringer, Mannheim, FRG) device and Lancet (Sherwood Medical, Crawley, UK). Blood ran freely into a heparinised Microtainer (Becton-Dickinson, Oxford, UK) via a scoop. The pulp was not squeezed and blood was not scraped from the finger. When 200 µl of blood had been collected the patient was venesected. Samples were spun down at 2000 g for 8 min and the plasma pipetted off and frozen at -20°C . Samples were then transported to the laboratory in dry ice. Samples were later thawed and plasma insulin assayed by double antibody radioimmunoassay [2].

Of 42 capillary samples, 3 were insufficient in quantity and 11 were visibly haemolysed. The mean intra-assay coefficient of variation (CV) was 8.6% for capillary samples and 6.0% for venous samples. The inter-assay CV was 16.9% for capillary and 17.1% for venous insulin measurements at fasting concentrations.

Non-haemolysed capillary samples from 28 (16 Asian, 12 Caucasian) subjects were compared with undiluted venous blood. Samples correlated well with a Spearman's correlation coefficient of 0.91 (Fig. 1). However, a significant ($p < 0.01$) median reduction of 33.2% (range 1.4–64%) was found in non-haemolysed capillary samples. No difference in this reduction was found between Caucasian and Asian subjects. The insulin concentrations were higher in Asians than Caucasians as previously communicated [3] and this will be more fully described later.

Fig. 1. Log capillary vs log venous insulin concentrations open squares (□) are Asians and closed squares (■) are Caucasians. Units are log (mU/l)

The reduction by 34% in capillary plasma samples was surprising. Incorporation of interstitial fluid may play a part in lowering insulin concentration but this is, perhaps, unlikely to be the only cause of so large a reduction, particularly as the terminal phalanx was not squeezed.

Although micro-haemolysis may have occurred in these 'no visible' haemolysis samples, this has previously been shown not to lead to significant reductions in immunoassayable insulin in venous blood [4]. Indeed, the median reduction in the haemolysed samples (7 Caucasian, 4 Asian) was no more than 40.2% (range 9.8–53%), many haemolysed capillary samples being only slightly lower than their venous equivalent.

Another possible mechanism for the reduction in immunoassayable insulin is an insulin degrading enzyme present in varying amounts in interstitial fluid and perhaps also present in erythrocytes: such an enzyme has been found in human lymphocytes [5]. It is unlikely that the difference is due to a real increase in plasma insulin during venesection as deep capillary blood sampling is more painful than venesection. However, in venesection the needle is seen and observed in situ, an event not occurring with the Autoclix.

These results suggest that capillary plasma immunoreactive insulin values can be used for studying the relative hyperinsulinaemia of Asians when compared with Caucasians but the data will not be comparable with surveys using venous plasma and the assay will not be sufficiently sensitive for use in clinical practice.

Yours sincerely,

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References

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