

Measurement of micro-albuminuria in patients with diabetes mellitus

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Summary Persistent micro-albuminuria (MA) in patients with diabetes mellitus identifies a high-risk group for the development of vascular complications. Since the methodology involved in measuring MA has not been standardised, MA in 40 diabetic patients was measured in order to compare nephelometry with radio-immunoassay (RIA). The effect of storage (7 days), the influence of freezing and thawing on MA levels, and possible differences between glass and plastic containers were also assessed. An excellent correlation ($r = 0,971$) was found between RIA and nephelometry. Urine could be safely stored at 4°C in either plastic or glass containers without any significant influence on MA concentrations. It is concluded that nephelometry is an excellent method for accurately measuring MA.

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In type I diabetes mellitus persistent micro-albuminuria (MA) signifies early diabetic nephropathy,¹ while in type II diabetes, it is associated with a markedly increased risk for the development of macrovascular disease.² Accurate measurement of urinary albumin levels is thus essential to identify these high-risk groups correctly. Unfortunately, controversy surrounds the methodology involved in measuring MA.³ In this study, some of these aspects were addressed. Specifically, we aimed to measure the urine albumin concentration in diabetics, comparing: (i) nephelometry with radio-immunoassay (RIA); (ii) freshly analysed urine with samples stored for 1 week; (iii) the effects of storage at 4°C with storage at -20°C; and (iv) possible differences between glass and plastic storage containers.

Patients and methods

Forty diabetic patients were studied (14 type I and 26 type II) and had to fulfil all the following criteria for inclusion: (i) no proteinuria, haematuria, leucocytes, nitrates or ketonuria on urine strip testing (Combur-9-Test; Boehringer Mannheim); (ii) no excessive physical exertion during the 24 hours before examination; and (iii) no other diseases except diabetes.

A spot midstream sample was collected from each subject during a routine outpatient visit. Each sample

was divided into 8 aliquots, 4 in plastic and 4 in glass containers. All the samples were centrifuged and subjected to: (i) immediate nephelometry and determination of urine creatinine value; (ii) nephelometry after 7 days of storage at 4°C and -20°C, respectively; and (iii) RIA within 2 weeks after collection (stored at -20°C).

Nephelometry was performed on an automated nephelometer (Beckman Array) and RIA (Pharmacia 100 AB; Sweden) according to the manufacturer's instructions. Creatinine levels were measured using the Beckman Astra-8. Statistical analysis was by Spearman rank correlations.

Results

There was an excellent correlation ($r = 0,971$) between urinary albumin levels measured with nephelometry and RIA (Fig. 1). No influence on albumin levels was apparent whether urine was stored in glass or plastic containers ($r = 0,996$). When compared with immediate nephelometry, neither storage at 4°C nor -20°C caused any significant change in mean albumin concentrations ($r = 0,991$ and $r = 0,958$, respectively). In two samples (5%), however, thawing after freezing at -20°C did cause a marked drop in albumin levels (from 26,8 mg/l to 7,9 mg/l and 23,8 mg/l to 5,9 mg/l, measured by nephelometry). Similar changes were detected in these samples employing RIA.

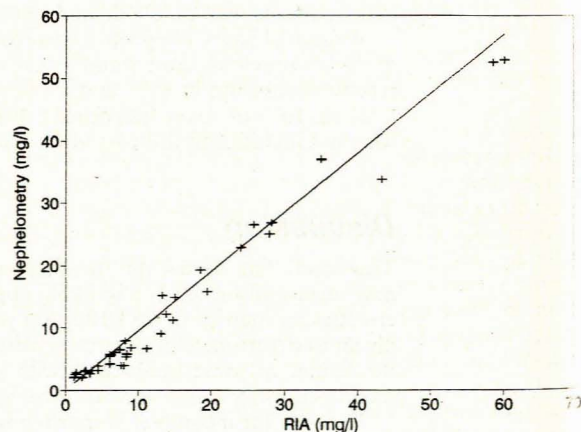


FIG. 1. Correlation between RIA and nephelometric measurement of urinary albumin ($r = 0,971$).

Taking an albumin/creatinine ratio (mg/mmol) of 2,3 as indicative of MA, the urine of 30% of our patients exceeded this value. This compares favourably with the generally accepted prevalence of MA in diabetes.¹

Discussion

RIA is considered the gold standard for measuring MA.³ Although nephelometry has been used⁴ to measure MA, published reports comparing RIA with nephelometry

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are lacking. In a recent study,⁵ these two methods compared very well, although over a wide range of proteinuria nephelometry measured on average 20% lower than RIA. Our study confirms this impressive correlation, but found a smaller difference (7%) between the measurements in the normo- and micro-albuminuria range.

Since nephelometry is a far simpler, non-isotope method, widely available and hence less expensive, we suggest that it should become the method of choice to measure MA. We could not confirm previous results⁶ suggesting that albumin adsorbs to plastic surfaces, thus causing falsely low levels of albumin when urine is stored in such containers. We did confirm, however,⁷ that at least in a minority of samples (5%) freezing at -20°C and thawing decreased albumin levels markedly. It has been suggested that this phenomenon is associated with the formation of precipitates consisting mainly of urates⁷ and that it can be prevented by adjusting the pH of urine to neutral either before or after deep-freeze storage.⁸

We conclude that nephelometry can be used for the accurate measurement of MA in diabetic subjects. Urine can be safely stored for up to at least 1 week at 4°C in

either plastic or glass containers.

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Scombroid poisoning

Case series of 10 incidents involving 22 patients

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Summary

Scombroid poisoning is a form of ichthyosarcotoxism caused by the consumption of 'spoiled' fish of the dark meat varieties. It can be considered a mild-to-moderate form of 'food poisoning' and it occurs world-wide. Ten incidents, involving 22 patients, were reported to Tygerberg Hospital Pharmacology and Toxicology Consultation Centre in the first quarter of 1990. Cape yellowtail (*Seriola lalandii*) was involved in all the cases. The presenting symptoms and signs (in order of frequency) were: skin rash, diarrhoea, palpitations, headache, nausea and abdominal cramps, paraesthesia, an unusual taste sensation and breathing difficulties. The patients responded well to antihistamines and, in most, the condition resolved within 12 - 24 hours.

Although histamine plays an important role in the pathogenesis of scombroid poisoning, the exact mechanism is still unresolved. The condition should be recognised and not confused with a true seafood allergy. Health workers are urged to alert

the authorities when outbreaks of suspected cases of scombroid poisoning are encountered in order to establish the possible cause and to prevent further cases.

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Scombroid poisoning (also referred to as histamine food poisoning) is a form of ichthyosarcotoxism caused by the consumption of 'spoiled' fish, which has undergone autolytic changes as a result of improper storage conditions.¹⁻⁴ The term 'scombroid poisoning' originates from the fact that spoiled fish from the family Scombridae (e.g. tuna, mackerel and bonito) were originally implicated in incidents of this type of poisoning.¹ However, it seems that non-scombroid fish are also involved, and on rare occasions even certain cheeses.¹ (Table I summarises fish species which may be implicated in scombroid poisoning.)

Clinically, scombroid poisoning resembles a histamine-like or acute allergic reaction.¹ Most cases are mild and self-limiting, even without treatment. Serious complications are rarely encountered and no deaths have been reported in recent times.^{1,5}

Scombroid poisoning occurs world-wide.² Between 1968 and 1986 a total of 188 outbreaks of 'histamine poisoning' involving 1 107 cases were reported to the Centers for Disease Control in the USA.¹ Most of the outbreaks were rather small, involving 5 or fewer individuals per incident. However, in 1973 a large outbreak, which involved 232 individuals, occurred in the USA, after the victims had ingested commercially canned tuna.³ A reference to an outbreak of histamine food poisoning in South Africa, involving 70 people, was found.² Further details of the latter incident could unfortunately

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