Technical method

A reduced incubation period for the destruction of somatic adenosine triphosphate in urine specimens

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The use of luciferin-luciferase adenosine triphosphate (ATP) assay as a rapid alternative to cultural methods for bacteriuria screening has been recently reported. 1,2 Measurement of bacterial ATP in urine is dependent on effective removal of host-derived somatic ATP. Various methods have been described 3–4 which destroy or remove unwanted somatic ATP, however, many of the earlier techniques 3–7 involved complex or time-consuming methodology. The advent of a commercially available somatic nucleotide releasing agent (NRS; Lumac BV, Schaesberg, The Netherlands) has greatly simplified these earlier procedures. The addition of NRS in equal volume to a sample of urine allows the quantitative release of ATP from somatic cells within a period of 10–15 s. 8 Subsequent destruction of released somatic ATP using a high quality ATPase enzyme usually involves incubation at room temperature for 45 min. 1,8 This study was carried out to determine whether higher temperature incubation (37°C) for a reduced period (20 min) would significantly alter the sensitivity of the screening technique.

Material and methods

One thousand routine urine specimens were screened for bacteriuria using two preincubation methods for the destruction of somatic ATP, 45 min at room temperature and 20 min at 37°C. The method, equipment and reagents used to carry out the 45 min room temperature preincubation technique have been described. 1 The same method was used for the rapid technique with preincubation being carried out in a 37°C incubator for 20 min. Specimens giving a relative light unit (RLU) value of greater than 711 (104 organisms/ml) 1 by either or both techniques were cultured on salt-free McCon-...
### Technical method

#### Results of 1000 urine specimens screened by bioluminescence using two preincubation techniques

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<th>37°C for 20 min</th>
<th>Room temp for 45 min</th>
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</thead>
<tbody>
<tr>
<td>Bioluminescence-negative</td>
<td>608 (60.8%)</td>
<td>627 (62.7%)</td>
</tr>
<tr>
<td>Bioluminescence-positive</td>
<td>392 (39.2%)</td>
<td>373 (37.3%)</td>
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We would like to thank Dr DM Jones and Mr P Davis for advice and encouragement, Mrs B Jinks who typed the manuscript and our colleagues who ran the screening system on a routine basis.

### References


Requests for reprints to: Mr PW McWalter, Chief Medical Laboratory Scientific Officer, Withington Hospital, West Didsbury, Manchester M20 8LR, England.

### Letters to the Editor

#### Hypothermia and pancreatitis

Dr Foulis’ recent study relating hypothermia with the morphology of the associated acute pancreatitis is of great interest. This work, together with Dr Foulis’ previous comprehensive survey relating the histological pattern of pancreatitis with various clinical diagnoses may shed some light on the pathogenesis of pancreatitis.

We would like to report certain findings of our own and consider their relevance to Dr Foulis’ studies.

We have shown that at temperatures below 37°C the inhibitory action of prostacyclin (PGI₂) on in vitro platelet aggregation is diminished. Furthermore, the in vitro synthesis of PGI₂ by vascular endothelium is decreased at low temperatures. We have also suggested that at low temperatures the vasodilatory action of PGI₂ is diminished. The raised serum non-esterified fatty acid (NEFA) concentrations reported in hypothermic patients may be of relevance since we have shown that high NEFA concentrations inhibit vascular PGI₂ synthesis and accelerate PGI₂ decay in albumin solutions.

Hypothermia may thus impair synthesis, increase the rate of decay and diminish the potency of PGI₂. These changes would result in platelet activation and predisposition to thrombosis and ischaemia. Abnormal platelet behaviour has been demonstrated in an animal study where thrombocytopenia occurred during induced hypothermia. An increased incidence of thrombotic phenomena in patients with hypothermia is evident from Dr Foulis’ own observations and from the literature he cites.

Ischaemic damage to the pancreas may occur in hypothermic patients as a result of platelet activation (as discussed above) or of “microcirculatory shock” as suggested by Dr Foulis. Such damage to the pancreas may cause the release of various enzymes. Amongst these, elastase, is thought to play a role in the destruction of vessel walls in patients with pancreatitis. Damage to vascular endothelium would further impair PGI₂ synthesis and expose deeper layers of vessels which may initiate platelet aggregation due to their collagen content. The release of trypsin is thought to activate the coagulation cascade, enhancing the ten-
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