content. Comparison of myocardial enzyme activities in biopsies with those in autopsies revealed significant differences for CK and aspartate aminotransf erase (EC 2.6.1.1), but not for HBDH and LDH content [5]; therefore, we did not measure the first two enzymes in the present study. Van der Laarse et al. [5] observed a HBDH content (mean ± CV) of 122 U/g (±12.3%) determined at 25°C. Applying the temperature correction factor of 1.3 results in a content of 159 U/g (±12.3%), which is comparable with the value of 161 U/g (±15.2%) observed in the present study at 37°C. For LDH van der Laarse et al. [5] found 166 U/g (±13.3%), which, after applying a temperature correction factor of 2.1, results in a LDH content of 349 U/g (±13.3%). This is slightly lower but still comparable with the LDH content of 397 U/g (±14.5%) observed in the present study. In another study, van der Laarse et al. [7] compared LDH content in myocardial autopsies from nonhypertrophic (n = 33) and hypertrophic human hearts (n = 36) and found contents of 182 U/g (±12.6%) at 25°C (382 U/g at 37°C) in nonhypertrophic and 155 U/g (±20%) at 25°C (326 U/g at 37°C) in hypertrophic myocardium. The mean LDH content of 397 (±14.5%) in our study corresponds very well with the value of 382 U/g (±12.6%) found by van der Laarse et al. in nonhypertrophic hearts.

Our present study also shows that, not only for LDH (P < 0.0001) and HBDH (P < 0.0001), but also for myoglobin (P < 0.0003) and FABP (P < 0.0115) the tissue content is negatively correlated to heart mass, and therefore to cardiac hypertrophy. In three patients with hypertrophic hearts (mean ± SD, 696 ± 179 g), mean contents for FABP, myoglobin, LDH, and HBDH were respectively 0.50 ± 0.05 mg/g, 2.63 ± 0.30 mg/g, 134 ± 28 U/g, and 330 ± 62 U/g. This indicates that the mean enzyme or protein content is ~10–15% lower than in nonhypertrophic hearts.

In hypertrophic hearts, we found that variation in cardiac tissue FABP content is comparable with the variation in cardiac tissue LDH or HBDH content, enzymes known to be stable on autopsy. Part of the variation in the present study is introduced by variation in heart weight, i.e., cardiac hypertrophy. Variations in myoglobin content were greater, making FABP, for reasons of both specificity and variation, more suitable for early assessment and early quantification of myocardial tissue damage.

### Table 1. Patients' data and mean enzyme and protein content of left ventricular tissue from 14 individuals.

<table>
<thead>
<tr>
<th></th>
<th>Mean</th>
<th>CV, %</th>
<th>Minimum</th>
<th>Maximum</th>
<th>Median</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart mass, g</td>
<td>328</td>
<td>18</td>
<td>215</td>
<td>430</td>
<td>328</td>
</tr>
<tr>
<td>Autopsy delay, h</td>
<td>14</td>
<td>61</td>
<td>3</td>
<td>24</td>
<td>12</td>
</tr>
<tr>
<td>Age of patient, years</td>
<td>68</td>
<td>28</td>
<td>20</td>
<td>90</td>
<td>68</td>
</tr>
<tr>
<td>LDH, U/g</td>
<td>397</td>
<td>14.5</td>
<td>277</td>
<td>542</td>
<td>401</td>
</tr>
<tr>
<td>HBDH, U/g</td>
<td>161</td>
<td>15.2</td>
<td>114</td>
<td>208</td>
<td>161</td>
</tr>
<tr>
<td>Myoglobin, mg/g</td>
<td>2.79</td>
<td>25.3</td>
<td>1.52</td>
<td>4.33</td>
<td>2.75</td>
</tr>
<tr>
<td>FABP, mg/g</td>
<td>0.57</td>
<td>15.4</td>
<td>0.42</td>
<td>0.80</td>
<td>0.56</td>
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<tr>
<td>Myoglobin/FABP</td>
<td>4.98</td>
<td>26.2</td>
<td>3.06</td>
<td>9.67</td>
<td>4.77</td>
</tr>
</tbody>
</table>

*Mean ± CV: 300 g (±17%) for women and 367 g (±12%) for men.

References


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Interference of Alum on Analysis of Methamphetamine in Urine Specimens

To the Editor:

It has been reported [1–4] that the presence of many commonly available commercial products, such as vinegar,
tea, salt, eye drops, liquid soap, detergents, bleach, \( \text{H}_2\text{O}_2 \), \( \text{NaHCO}_3 \), and \( \text{NaClO}_3 \) might interfere with the analysis of drugs of abuse in producing false-positive, false-negative, or invalid test results. It is widely believed by drug abusers in Taiwan that alum can interfere with the analysis of urinary methamphetamine. We have, therefore, conducted a study to evaluate the alleged interferences of alum on the analysis of methamphetamine in urine specimens by non-instrumental on-site immunoassay kits (listed below), Emit-st, and the thin-layer chromatography-based Toxi-Lab, which are commonly used in Taiwan.

The general formula of alums is \( \text{M}^{3+} \cdot \text{M}^{3+} \cdot (\text{SO}_4)_{(1.5+0.5)} \cdot 2\text{H}_2\text{O} \), where \( \text{M}^{3+} \) and \( \text{M}^{3+} \) represent metal ions with valences of 3 and 1, respectively. In the present work, we selected aluminum potassium salt, \( \text{AlK(SO}_4)_2 \cdot 12\text{H}_2\text{O} \), for evaluation.

Methamphetamine hydrochloride was obtained from the Narcotic Department of the Department of Health, Taiwan. Toxi-Lab Extraction Tube A (lot no. 30103) and TOXI GRAMS A (lot no. 20061) were purchased from Analytical Systems (Laguna Hills, CA). Emit-st Urine Amphetamine Assay reagents (lot no. 3C319UL-D1B), calibrators (lot no. 3A319UL-CID), and photometer were obtained from Syva Co. (Palo Alto, CA). Noninstrumental on-site immunoassay kits studied were accuPINCH (Hyco Biomedical, Garden Grove, CA; lot no. 34417); Mach IV (Drug Screening System, Blackwood, NJ; lot no. 92064); AbuSign (Princeton Biomeditech, Princeton, NJ; lot no. 61300); and I.D. Block (International Diagnostic System, St. Joseph, MI; lot no. 51693). The cutoff concentrations for Toxi-Lab, Emit-st, accuPINCH, Mach IV, AbuSign, and I.D. Block are 0.5, 1.0, 0.5, 1.0, 0.5, and 1.0 mg/L, respectively.

Working urine solution containing 50 mg/L methamphetamine hydrochloride was prepared by diluting 4 mL of a 2.5 g/L stock solution (in ethanol) to 200 mL with drug-free urine. Test urine specimens containing 100, 75, 50, 25, and 0 g/L alum and 10 mg/L methamphetamine hydrochloride were prepared by pipetting 10 mL of working urine solution into five 50-mL volumetric flasks containing 5.0, 3.75, 2.5, 1.25, and 0 g of alum; each flask was then brought to the 50 mL mark with drug-free urine. Similar procedures were used to prepare other series of test specimens containing 5.0 and 1.2 mg/L methamphetamine hydrochloride. The pH of each test specimen was measured with a pH meter before testing.

pH values and test results are summarized in Table 1; further comments are as follows:

1) Toxi-Lab. Bubble formation was observed when high concentrations (7.5 and 100 g/L) of alum-containing specimens were added to extraction tubes, perhaps related to a reaction between alum and the salt that was originally in the tube. This led to poor extraction efficiency, which gave a false-negative result for the specimen containing 1.2 mg/L methamphetamine hydrochloride.

2) Emit-st. pH values of the alum-containing specimens are less than the low-end of the testable range specified by the enzyme immunoassay manufacturer. The low pH may have caused lower enzyme activity and generated false-negative results for specimens containing methamphetamine concentrations only slightly above the cutoff (1.2 mg/L). This is consistent with an earlier report, in which Emit® d.a.u.™ was used for studying the interference effects of NaCl and detergent (containing NaOH and NaOCl) [4].

3) accuPINCH. Interferences of alum were observed. It was difficult to distinguish color between positive and negative for specimens with low methamphetamine concentration (1.2 mg/L).

4) Mach IV and AbuSign. Formation of distinctively colored control bands on the membrane adopted for the immunochromatography reaction mechanisms (see product package inserts) cannot be readily observed. Interferences of alum on the formation of color bands for both tests appear to be similar, but to slightly different extents: It takes 75 g/L alum to interfere with the color-band formation mechanism for Mach IV, whereas 25 g/L alum causes the same effect for AbuSign. The presence of lower alum concentration (25 and 50 g/L) causes false-negative or uncertain results for Mach IV, presumably for the same reasons experienced by Emit-st and accuPINCH.

5) I.D. Block. No false-negative results were observed for any specimens containing alum. However, false-positive results were observed for urine specimens that contained alum but no methamphetamine.

6) pH values <4.0 were observed for all specimens containing alum at 25 g/L or more. These pH values are obviously not within the 5.0–8.0 range of normal urine [5]. Therefore, we recommend measuring pH before performing any analysis, to detect any possible interference of alum.

In summary, (a) the presence of alum at a low concentration (25 or 50 g/L) causes false-negative or uncertain results.

### Table 1. Effects of alum on the analysis of methamphetamine in urine.

<table>
<thead>
<tr>
<th>Concentration of methamphetamine, mg/L</th>
<th>1.2</th>
<th>5.0</th>
<th>10</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>25</td>
<td>50</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>25</td>
<td>50</td>
</tr>
<tr>
<td></td>
<td>0</td>
<td>25</td>
<td>50</td>
</tr>
<tr>
<td>Toxi-Lab</td>
<td>+</td>
<td>+</td>
<td>±</td>
</tr>
<tr>
<td>Emit-st</td>
<td>+</td>
<td>+</td>
<td>−</td>
</tr>
<tr>
<td>accuPINCH</td>
<td>+</td>
<td>±</td>
<td>±</td>
</tr>
<tr>
<td>Mach IV</td>
<td>+</td>
<td>−</td>
<td>−</td>
</tr>
<tr>
<td>AbuSign</td>
<td>+</td>
<td>Δ</td>
<td>Δ</td>
</tr>
<tr>
<td>I.D. Block</td>
<td>+</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>pH value</td>
<td>7.6</td>
<td>3.9</td>
<td>3.7</td>
</tr>
</tbody>
</table>

* Alum concentration, g/L.
* False positive (drug-free urine specimens containing 25, 50, 75, and 100 g/L alum).
for Toxi-Lab, Emit-r, and accuPINCH on urine specimens containing 1.2 mg/L methamphetamine hydrochloride; (b) results generated by Mach IV and AbuSign tests are generally falsely negative or invalid in the presence of alum as low as 25 g/L; and (c) the presence of alum in the concentration range tested (25–100 g/L) produces false-positive results for I.D. Block on methamphetamine-free specimens.

Because the presence of alum may cause false-negative or uncertain results for Toxi-Lab, Emit-r, and accuPINCH and render tests invalid by Mach IV, AbuSign, and I.D. Block, we conclude that alum is an effective adulterant for the portable test kits tested. However, because alum at a concentration as low as 25 g/L will lower the specimen pH to below 4.0, pH measurement may be an effective way to detect the presence of this interferent.

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References

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Serum Troponin T: Diagnostic Marker for Acute Myocarditis

To the Editor:
Cardiac troponin T (cTnT), a contractile protein unique to cardiac muscle, can be differentiated from its skeletal muscle isofrom by immunoassay [1]. cTnT is highly concentrated in the cardiomyocyte, both as myofibrillar bound (94%) and as soluble cytosolic (6%) protein [2]. After myocardial cell necrosis an increased concentration of cTnT is observable in blood for more than a week [3]. With the now-available assay, one can detect myocardial cell necrosis in patients with angina at rest, which has so far been undetectable by conventional diagnostic methods [3,4]. Similarly, in all patients with Wolff-Parkinson-White syndrome undergoing effective radiofrequency ablation, the observed release of cTnT is not consistently paralleled by increases in blood of cardiac enzymes or myoglobin [5]. Thus cTnT measurements are particularly useful in clinical circumstances in which traditional enzyme determinations fail to diagnose myocardial cell damage efficiently, even in cardiac allograft rejection and perioperative myocardial infarction [6–8]. Given the high sensitivity of cTnT measurements, we were interested in whether determinations of cTnT might be useful for the noninvasive detection of myocardial cell damage in patients with acute myocarditis, classified as active or borderline according to detectable myocardial necrosis [9].

Seven patients (ages 19–27 years) admitted with chest pain and transient ST-segment or T-wave changes on the electrocardiogram constituted our study group. Except for moderate smoking by two individuals, no risk factors for coronary artery disease were identified. Cardiac catheterization revealed normal coronary arteries in all patients and a reduced left ventricular ejection fraction (<55%) in four of them. Because of the suspected myocarditis, at least five biopsies were taken from left ventricular myocardium. Informed consent was obtained from all patients. All agreed to the scientific analysis of their personal and medical data. The muscle specimens were all analyzed by histology and some (2 of 6 positives) by immunohistochemistry with anti-T-lymphocyte antibodies. According to the Dallas criteria [9], histological analyses supported the diagnosis of active myocarditis (n = 2) with myocyte necrosis or borderline myocarditis (n = 4). There were no clinical, angiographic, or histological features indicative of ischemic damage. One patient had no signs of myocarditis on histological analysis but revealed regional contractile dysfunction on left ventricular angiogram and an intermitting left bundle branch block. Serial blood samples were obtained from all patients until day 9 after admission to determine cTnT, total creatine kinase (CK) activity, and the CK-MB isoenzyme/total CK ratio (respectively determined with Troponin-T ELISA, CK-NAC, and CKMB-NAC assays, all from Boehringer Mann-