normal dermatan sulfate band. Microbiological examination showed the presence of Enterococcus species, 10⁶ organisms per milliliter, in the inoculated urine.

The anomalous result was therefore almost certainly attributable to bacterial degradation of the abnormal dermatan sulfate originally present in the urine. This case emphasizes the necessity of obtaining fresh urine for analysis or, if that is impossible, adding an appropriate preservative (e.g., thimerosal) that does not affect the electrophoretic patterns obtained. The finding of a false-negative result on screening clearly has important implications for further investigation of the patient.

References


Bronchial carcinoma is not an inevitable consequence of cigarette smoking, except for subjects exhibiting high metabolic oxidative activation of chemical carcinogens, as well as of the antihypertensive drug debrisoquine (1), which is also metabolized (2) via the cytochrome P450 enzyme P450db1.

A co-segregation of dextromethorphan O-demethylation with debrisoquine 4-hydroxylation has been demonstrated (3) and, in view of the innocuousness and ubiquitous availability of dextromethorphan, we propose dextromethorphan metabolism as a phenotypic marker for susceptibility to lung cancer.

We studied 146 randomly selected normal volunteers who had no recent illness, were receiving no drug, and had no evidence of hepatic or renal dysfunction, and 20 cancer patients who had a definite diagnosis of bronchogenic carcinoma proven by histology.

Each subject was given 64 μmol of dextromethorphan orally and all urine was collected for the subsequent 8 h. The concentrations of dextromethorphan and its metabolite dextrorphan were determined by gas chromatography (3).

The frequency distribution of the logarithmically transformed data was trimodal (Figure 1). The values for log metabolic ratio ranged from -2.81 to 0.30.

Most cancer patients were rapid metabolizers, with log metabolic ratios of less than -2; they probably have homozygous dominant genotypes, as suggested by Ayesh et al. (4), for debrisoquine hydroxylation. Heterozygotes and recessive poor metabolizers should be considered at relatively low risk.

Neither sex nor smoking habits affected the metabolic ratios in our population. Dextromethorphan oxidation evidently can be used as a safe and reliable test to determine whether an individual smoker is at high risk of developing cancer.

References
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intact PTH was therefore modified for monitoring PTH concentrations during parathyroid surgery within 15 min (<4, 5). We suggest a modification of another commercially available immunometric assay of PTH for use in hypercalcemic crisis.

An immunochemiluminometric assay of PTH (Ciba Corning, Fernwald, F.R.G.) was modified as follows: The first incubation step with labeled antibody was shortened to 1 h and was performed at 37 °C, to limit the overall assay time to 2 h. The calibration curve remained linear, its slope decreased to 31.1% for 1 h of incubation at 37 °C (40.1% for 2 h, 19.8% for 0.5 h), compared with 24-h incubation at room temperature. Shortening of the second incubation step—i.e., the separation by solid-phase-bound antibodies—performing both steps together was less useful. PTH concentrations determined with the rapid assay modification correlated with the results of the original assay (r = 0.98, slope = 1.01; Figure 1). Patients with primary hyperparathyroidism were distinguished from normal individuals, except for two patients with mild hypercalcemia (<2.8 mmol/L) and PTH values within the normal range (55 and 58 ng/L). One patient with hypercalcemic crisis and total serum calcium >4.0 mmol/L showed extremely high PTH concentrations (>1900 ng/L) in both assays. The limit of detection was about 4 ng/L (2 SD above B). Intra-assay CVs ranged from 6% (sample with 35 ng/L) to 12% (at 230 ng/L). Inter-assay CVs were between 9% and 18% for normal or above-normal PTH values (compared with 9% to 13% for the original method).

In conclusion, this rapid assay of intact PTH is a valuable diagnostic tool in hypercalcemic crisis. It can easily be performed within 2 h with minor modification of a commercially available kit. It allows the reliable discrimination of the high PTH values seen in severe primary hyperparathyroidism from the normal or suppressed values in hypercalcemia of malignancy.

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Urinary Total Hydroxyproline Measured by HPLC: Comparison of Spot and Timed Urine Collections, P. S. Wilson, M. Kleerekoper, H. Bone, and A. M. Parfitt (Bone and Mineral Research Lab., Henry Ford Hospital, 2799 W. Grand Blvd., Detroit, MI 48202)

Total hydroxyproline (THP) in 24-h timed urine collections has long been measured as a marker of bone resorption. Analysis of a spot (i.e., untimed) urine collection after an overnight fast provides a useful indicator of osteolytic activity in patients with multiple myeloma and with breast cancer (1, 2). In our study the second-voided urine after an overnight fast was obtained from 70 patients being evaluated for metabolic bone disease, and a timed 2-h urine collection was then begun immediately. Total hydroxyproline/creatinine (THP/Cr) ratios were compared in these paired samples.

Urinary THP was measured by the HPLC method reported by Dawson et al. (3), with the following modifications: After drying the 50-μL aliquot of urine, add 50 μL of 6 mol/L HCl to each reaction tube and 200 μL of HCl/phenol solution to the reaction vial. Hydrolyze the samples for 21 h at 110 °C. The redrying solution for physiological hydrolysates is methanol/sodium acetate (1 mol/L)/triethylamine (TEA) (2/2/1 by vol); derivatizing reagent is methanol/TEA/phenylisothiocyanate/water (7/1/1/1 by vol).

Chromatography—Using the 3.9 mm × 15 cm Waters "Pico-Tag" column for acid hydrolysates, elute isocratically at 100% solvent A (0.14 mol/L sodium acetate plus, per liter, 0.5 mL of TEA, titrated to pH 6.4 with glacial acetic acid) for the first 3 min after injection, then increase linearily to 100% B (acetonitrile/water, 60/40) over the next 2 min. Hold at 100% B for 2 min, then increase the flow rate to 1.5 mL/min during the next minute. Reverse the gradient to 100% A in 1 min; re-equilibrate the column with solvent A (1.5 mL/min) for the next 7.5 min; then return to a flow rate of 1 mL/min.

Total analysis time is 17 min. The hydroxyproline peak is eluted in about 6 min. Quantification is by the external standard method, with use of peak area vs concentration. The standard curve is linear over the range 1.9–30 mg/L. We routinely measure urinary THP concentrations in the range of 1–20 mg/L (8–153 μmol/L) with representative CVs of 4.5% (intra-assay, n = 14) and 6.3% (interassay, n = 15). Creatinine was measured with an Astra-4 Automated Stat/Routine Analyzer (Beckman Instruments, Carlsbad, CA).

The study population represents a cross section of metabolic bone diseases, including osteoporosis, hyperparathyroidism, renal osteodystrophy, and Paget's disease. Urinary THP/Cr ratios ranged from 8 to 108 mg/g (normal 7–21 mg/g), with excellent agreement between the timed (x) and spot (y) collections in all cases as demonstrated by