for the same groups from siliconized tubes showed significant differences in values <11 pmol/L (mean 11.6 pmol/L, SD 2.5 pmol/L, $p < 0.05$) and in values between 11 and 28 pmol/L (mean 23.6 pmol/L, SD 5.3 pmol/L, $p < 0.001$), whereas values >28 pmol/L did not show significant differences. Thus, as expected from these results and from the regression study, a misclassification of subjects was produced. According to our reference interval, five (55.5%) of the nine subjects with low FT4 values had values for samples from siliconized tubes >11 pmol/L; similarly, 16 of the 89 subjects (18.0%) with normal values of FT4 had values >28 pmol/L; whereas for the subjects with FT4 >28 pmol/L no misclassification of any type was observed. Thus some low values were misclassified as normal and some normal values as high.

This increase in FT4 appears to be related to FT4 concentration, because it was noticeable only for samples with normal or low concentrations of FT4. Thus, when assessing FT4 concentrations with the GammaCoat "Two-Step" kit, one should take into account, among other things, the type of serum sample used for the assay, particularly if the reference intervals are based on different sample-collection conditions.

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Labetalol: False-Positive Indices by EMIT-d.a.u. Assay and Toxi-Lab A Urine Screen

To the Editor:

We report for the first time the false-positive appearance of amphetamines by both the Syva EMIT-d.a.u. assay as well as on the Marion Laboratory Toxi-Lab A urine screen in a 69-year-old woman being treated for hypertension with the drug labetalol ("Normodyne," Schering Corp.). In addition, the Toxi-Lab A screen gave the false impression of the presence of trimethoprim and its metabolites.

On the seventh day after admission for treatment of malignant hypertension, a routine basic drug urine screen was requested. The toxicology laboratory reported a positive result for amphetamine by the EMIT-d.a.u. assay (*1*). The Toxi-Lab A screen, involving thin-layer chromatography (*2*), showed migration patterns (R_f values) and color characteristics extraordinarily similar to amphetamine and methamphetamine. In stage I, however, the color characteristic of the unknown drug was slightly more orange than amphetamine. In stage III, the migration patterns and color characteristics of two other unknown spots were indicative of trimethoprim and its metabolite.

Upon reporting our findings, we were immediately contacted by the clinician attending the patient. He informed us that the patient was neither receiving nor had access to either drug, amphetamine and trimethoprim, reported by the laboratory. Repeating the urine screen on the same urine above as well as a newly collected specimen (27 h later) showed results similar to those described above. We also observed a positive result for amphetamines with the EMIT-d.a.u. confirmatory procedure (*1*).

After reviewing the patient's chart, we noted that the only medications the patient was receiving were cimetidine, ranitidine, nitroprusside, and labetalol (Normodyne, 300 mg, orally, twice a day). The labetalol · HCl formulation was analyzed in comparison with the patient's urine specimens and pure standards of amphetamine and trimethoprim. By Toxi-Lab A thin-layer chromatography, the labetalol drug migrated slightly below the amphetamine standards (R_f, 0.42 and 0.35, respectively) but was similar in color characteristics and fluorescence. However, the patient's urine specimen was indistinguishable from the amphetamine and trimethoprim standards. The Toxi-Lab A screen acetone/sodium hydroxide confirmatory thin-layer chromatographic procedure (*2*) was also unable to distinguish the patient's specimen from amphetamine · standards. Both the EMIT-d.a.u. and the confirmatory d.a.u. were positive when we analyzed the labetalol formulation given the patient, the EMIT-d.a.u. being positive at a labetalol concentration of 1.0 μg/mL. Previously, neither Syva Co. nor Marion Laboratories have reported false-positive responses for either amphetamines or trimethoprim in patients receiving labetalol.

The metabolism of labetalol, an adrenergic receptor blocking agent used in the treatment of hypertension, is mainly through conjugation to glucuronide metabolites (*3*), present in plasma and excreted in the urine. About 55 to 75% of a dose appears in the urine as conjugates or unchanged labetalol within 24 h of dosing. Apparently, when labetalol is given therapeutically for the treatment of hypertension, the parent drug (secondary amine) and its metabolites both cross react with antibodies in Syva's urine screen for drug abuse. This cross reactivity may be ascribable to the 1-methyl-3-phenyl-propylamino side chain on labetalol, making it structurally similar to amphetamine-like drugs. At present, we also cannot account for the similarity of a false-positive pattern for the pyrimidine-derivative trimethoprim, by the Toxi-Lab A system. Thus, clinical toxicology laboratories should be cognizant of these potential sources of error (false-positive results) when these methods are used to screen for drugs of abuse.

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Manganese Concentration in the Hair of Greying ("Salt and Pepper") Men Reconsidered

To the Editor:

Pigmented structures from various sources are known to be rich in trace metals, including Zn, Cu, and Mn. The last is in relatively high concentration in melanin-containing tissues (*1, 2*). In determining the concentration of Mn in black hairs as compared with white, only Cotzias et al. (*1*) have demonstrated, by neutron activation, a lower amount of Mn in white hair than in black from the same greying "salt and pepper" subjects. But their results were open to criticism. We applied the Student's *t*-test for paired data to the results obtained by these authors for five subjects. The difference in Mn contents for white hair [2.83 (SD 3.04) μg/g dry weight] and dark hair [18 (SD 19) μg/g dry weight] was insignificant ($p = 13$).

Since 1964, analytical methods have improved greatly. Consequently, we repeated this sort of study, using new improved techniques. Besides Mn, we also measured and compared the Cu and Zn content in six men with both black and white hair. The conditions and precautions in taking the hair samples were the same as those previously described (*3*). The subjects' ages ranged from 35 to 45 years. Hair from each individual was divided into black and white. The entire fibers were included in the samples, with no segmentation, because we were not concerned here with variations along the hair fiber (*4*). A 0.5-g hair sample was washed with a 10 mL/L solution of nonionic detergent, rinsed, and dried, all as described by Harrison et al. (*5*). An aliquot of the washed hair was digested with HNO_3 and H_2SO_4, without loss, in a Perkin-Elmer miniautoclave. Then we determined the Mn content by flameless atomic absorption spectrometry, with Zeeman background correction (*3*). Cu and Zn were determined by flame atomic absorption spectrometry (*6, 7*).

Table 1. Concentrations of Cu, Mn, and Zn in the Hair of Six "Salt and Pepper" Men

	Cu		Mn		Zn	
	μg/g dry wt					
Hair color	Black	White	Black	White	Black	White
	12.96	11.88	0.21	0.16	160	156
	9.14	8.00	0.24	0.13	91	76.6
	10.60	9.13	0.20	0.10	172	168
	11.00	10.71	0.24	0.05	177	182
	12.12	6.8	0.25	0.11	206	187.5
	13.95	15.00	0.29	0.21	260	240
Mean	11.6	10.25	0.24	0.13	177.7	168.3
SD	1.7	2.95	0.03	0.05	55.5	53.4
	NS		$p < 0.01$[a]		NS	

[a] By Student's *t*-test for paired data. NS, not significant.

Table 1 summarizes our results. The Cu and Zn content of black and white hair from the same individual compared well with values for different subjects and with most published values (*8–11*). The most interesting results were those for Mn. Indeed, we agree with Cotzias et al. (*1*) that white hair has a lower concentration than black hair. However, the standard deviation of the values in their publication (see calculations above) left this conclusion equivocal. Measurement of Mn requires a rigorous methodology, and research carried out since 1964 on measurement of Mn in unexposed subjects has always revealed a relatively wide range of values. In contrast, our values were closely grouped, and lower. White and black hairs, compared by Student's *t*-test of paired data, had a significantly different Mn content ($p < 0.01$).

Does Mn play some important role in hair pigmentation?

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Estrogen and Progesterone Receptor Content in Breast Tumor Cytosols Calculated with a Pocket Calculator

To the Editor:

Those who don't have a calculator with a linear regression chip can reduce receptor data for any number of XY pairs of specific receptor-bound ligand and paired free ligand at equilibrium by using the linear regression equation (*1*): $\bar{y} = m\bar{x} + b$, where $\bar{y}$ equals the mean of the dependent variables and $\bar{x}$ equals the mean of the independent variables. The slope m can be calculated as: $(y_2 - y_1)/(x_2 - x_1)$, where x_2 and y_2 are obtained with the highest con-