in patients who are taking T₄ is not as great as in those patients who are not on T₂ replacement.

If we disregard the 14 samples that gave erroneously high results with the Amerlite assay, the use of TSH as a front-line screen in our laboratory has changed the numbers of samples that require additional assays to be done after the initial screening assay. Using FT₄ as a screening assay and the cutoff values noted, we would do a TSH assay on 44% of samples and an FT₃ assay on a further 5% of patients' samples. If we used TSH as the screening assay, 24% of all patients would require an FT₄ assay and an additional 9% would require an FT₃ assay if the strategy proposed by Caldwell et al. were adopted. In the latter case, therefore, we would perform 16% fewer additional tests after the screening assay.

We conclude that it is premature to propose that TSH assays replace estimates of free thyroid hormones in the diagnosis of thyroid dysfunction, but with further careful evaluation TSH assays may be reliable as screening tests with free thyroid hormones as backup tests.

We are grateful to Amersham International for the loan of the Amerlite system to carry out this evaluation.

References

Effect of Glucose Loading on Concentrations of Chromium in Plasma and Urine of Healthy Adults
Brian W. Morris, Huw Griffiths, and Graham J. Kemp

We report here a small study designed to identify the effect of a 75-g oral glucose load on concentrations of chromium in plasma and urine of apparently healthy volunteers. We detected a consistent and significant (P <0.01) decline in plasma chromium after glucose administration, the nadir of the chromium response coinciding with the zenith of the glucose concentration.

Additional Keyphrases: trace elements · nutrition · atomic absorption spectroscopy

Since the early work in the 1950s-1970s (1-4) there has been much interest in chromium and the nutritional state of chromium, especially in diabetes and in other conditions associated with glucose intolerance.

The minimum daily human requirement for chromium necessary to maintain health is not known. Mertz (5) suggested an allowance of 50 to 200 μg/day. Schroeder et al. (6) found a mean chromium content of 72 μg in the daily diet of institutionalized subjects, and Levine et al. (7) reported a daily intake of 5 to 125 μg in the diet of elderly subjects. The requirement for chromium in the diet therefore seems to be variable, and may depend on the chemical form in which it is available.

Impaired glucose tolerance in humans was improved by the simple addition of 150 μg of inorganic chromium salt to the daily diet (7-9), and various workers (10, 11) have reported the apparently beneficial effects of an organic form of chromium, "glucose tolerance factor" (GTF).

The improvements in glucose tolerance observed on supplementing the diet with chromium salt may have resulted from the conversion of inorganic chromium to the biologically active form (GTF), which potentiates the effects of available insulin (12).

Now that more-reliable techniques for determination of chromium concentrations in plasma and urine have become available, findings reported for diabetic patients (13-15)
and for healthy and diabetic subjects after an oral glucose load (16, 17) have been contradictory.

We have previously reported decreases in chromium in plasma from fasting diabetic patients, as compared with normal controls, measured with a sensitive and precise electrothermal atomic absorption technique (18). We report here the effects of an oral glucose load in normal subjects.

Experimental Procedure and Methods

This small study involved five apparently healthy volunteers (two men, age 33, 31 y, weight 66, 63 kg; and three women, age 31, 26, 25 y, weight 60, 54, 57 kg) with no family history of diabetes mellitus or history of occupational exposure to chromium. The oral glucose tolerance test consisted of taking 75 g of glucose in the morning after an overnight fast of at least 11 h.

We used the following study protocol: The participants ate no principal meal later than 2000 hours, light snacks only from 2000 to 2200 hours, and water only after 2200 hours. They supplied a midstream urine sample on rising the next day, and urine and fasting blood samples at 0900 hours, for determinations of chromium and glucose, after which they drank 75 g of glucose dissolved in 250 mL of de-ionized/distilled water. We sampled blood from them at 15, 30, 45, 60, 90, 120, 150, and 180 min after this glucose load, and urine samples at 30, 60, 120, and 180 min after the load. The participants also drank 150 mL of de-ionized/distilled water 0, 15, 30, 45, and 60 min after the load.

The glucose and water administered were chromium-free. Blood samples were taken, when possible, with a 21-gauge indwelling winged needle. Blood samples for chromium determinations were centrifuged immediately and the corresponding plasmas were separated into chromium-free containers. Samples for glucose determinations were collected into tubes containing fluoride/oxalate, and plasma glucose was measured within 20 min of sampling, in a Clandon YSI 23AM glucose analyzer (YSI Scientific Division, Yellow Springs Instrument Co. Inc., Yellow Springs, OH 45387).

All analyses for chromium in plasma and urine were done within 48 h of collection, by electrothermal atomic absorption spectroscopy as described previously (18).

We measured urinary creatinine by a Jaffe-based reaction, in an RA 1000 analyzer (Technicon method no. SM4-0141K82). Urinary chromium was expressed relative to creatinine to account for variations in urine volumes.

The significance of maximum changes in concentration was assessed by use of Student's unpaired t-test.

Results and Discussion

The mean (± SEM) concentrations of the chromium and glucose in plasma at each sampling time are shown in Figure 1(a and b).

In plasma the concentration of glucose was significantly ($P < 0.01$) increased above basal concentration 15 min after the load; chromium was decreased below its basal value within 15 min, reaching its lowest value ($P < 0.01$) by 30 min after the load. The glucose concentration was greatest slightly before the nadir of the chromium values, but this difference was not significant.

Several workers (17, 19) have reported a decrease in the concentration of chromium in plasma after a glucose load. Davidson and Burt (17) reported a decrease in plasma chromium concentrations in normal nonpregnant women, in response to both oral and intravenous glucose. Liu and Morris (19) found that serum chromium decreased in normal subjects after 1 h during a 3-h glucose-tolerance test. Glinsmann et al. (16) reported an increase in serum chromium in response to a glucose challenge, and Levine et al. (7) reported similar increases in elderly subjects after glucose loading. Behne and Diehl (20) showed an increase in chromium in blood, which was greatest 60 to 120 min after a glucose load.

In our small study we have shown a significant decrease in chromium concentration in plasma, which coincides with the peak glucose concentration in plasma after a glucose load. Urinary excretion of chromium, however, was rather variable (Figure 1c); the small increase was consistent with findings of other workers (21, 22), but it was not statistically significant. Increased urinary excretion is inadequate as an explanation of the decrease in plasma chromium. Little is known about chromium concentration in extravascular water, but even if extracellular chromium were confined to the intravascular space, whose volume is approximately 3 L (23), the fall in plasma chromium by about 5 nmol/L at $t = 30$ min would require the loss of 15 nmol of chromium from the extracellular space. Normal creatinine excretion (24) in 30 min is 0.2–0.4 mmol; if this chromium were lost entirely in urine we would expect urinary chromium excretion to be
40 to 80 μmol per mole of creatinine, which is 100–200 times greater than the values we observed. Alternatively, if the concentration of chromium in total extracellular water, whose volume is approximately 20 L (23), reflects that in plasma, then we would expect the urinary excretion of chromium to be at least 1000 times greater than the values we observed. We conclude that increased urinary loss is not responsible for the observed decrease in the concentration of plasma chromium.

Further studies will be needed to identify the principal mechanism responsible for the decrease in chromium concentration in plasma and to identify any abnormalities in dynamic responses in diabetic subjects, in whom the basal concentration of chromium in plasma is known to be significantly decreased (15).

References
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An Enzyme Immunoassay Compared with a Ligand-Binding Assay for Measuring Progesterone Receptors in Cytosols from Breast Cancers

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To assay progesterone receptor (PR), we compared Abbott's enzyme immunoassay (PR-EIA) with a ligand-binding assay involving dextran-coated charcoal (PR-DCC), using cytosols prepared from 109 breast-cancer biopsies. Results by the two PR methods agreed well. Least-squares analysis produced a line of best fit having a slope of 0.88, an intercept on the PR-EIA axis of 16 fmol per milligram of protein, and a correlation coefficient \( r^2 \) of 0.87. To evaluate whether accurate PR-EIA measurements could be obtained on stored cytosols, we compared PR-EIA values for fresh cytosols with values for cytosols stored for various lengths of time up to 13 weeks. Agreement was excellent, especially when the samples showing very high binding (>600 fmol per milligram of protein) were excluded. The lines of best fit after least-squares analyses of the remaining values had slopes between 1.0 and 1.1, intercepts <3 fmol/mg, and \( r^2 \) all >0.91.

Additional Keyphrases: sample handling • monitoring therapy

Steroid receptors have become important as prognostic indices. Progesterone receptor (PR) status combined with estrogen receptor (ER) status defines at least three biological subgroups (ER+/PR+, ER+/PR−, ER−/PR−), which display different responses to endocrine therapy. In patients with recurrent breast cancer who were treated with endocrine therapy, PR presence improved the predictive accuracy of ER+ by a further 15–20%, to approximately 70% (1). Receptor-negative tumors rarely respond to hormone therapy.

In adjuvant chemotherapy, determination of PR added significantly to the predictive value of ER (2). Others have