

INCREASES IN SERUM CONCENTRATIONS OF FOLLICLE-STIMULATING HORMONE (FSH) DURING THYROTROPHIN-RELEASING HORMONE (TRH) INFUSIONS IN NORMAL MEN

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SUMMARY

Increases in serum FSH values to approximately 50% above basal concentrations were found during TRH infusions of 2.0 $\mu\text{g}/\text{min}$ into normal men ($n = 10$). These increases could not be explained by cross-reactivity of thyrotrophin (TSH) in the FSH assay. No significant change was found in serum concentrations of FSH during saline infusions ($n = 4$) nor in LH concentrations during either TRH or saline infusions. Many previous studies using single injections of TRH have failed to demonstrate changes in serum FSH concentrations. It is possible that prolonged infusions of TRH, as used in this study, are more likely to lead to non-specificity of the effects of this tripeptide on pituitary hormone secretion.

In a large number of studies, TRH administered in single injections has been found to release only TSH and prolactin (Burger & Patel, 1977). Stimulation of the release of other pituitary hormones by this tripeptide has not usually been demonstrable in normal subjects. In some abnormalities, particularly pituitary tumours, there appears to be a breakdown in the specificity of the action of TRH. In acromegaly, depression and chronic renal failure, TRH may stimulate GH secretion (Irie & Tsushima, 1972; Schalch *et al.*, 1972; Gonzalez-Barcena *et al.*, 1973; Maeda *et al.*, 1975; Czernichow *et al.*, 1976), and in Cushing's disease and Nelson's syndrome, it may stimulate ACTH secretion (Krieger & Luria, 1977). In the rare type of pituitary adenoma that secretes excessive amounts of LH and FSH, production of these hormones may be increased by TRH (Snyder & Sterling, 1976).

One group has reported that TRH, given either as single injections or as prolonged infusions, stimulates FSH secretion in normal men (Mortimer *et al.*, 1973; Mortimer *et al.*, 1974). In a few women, an apparent stimulatory effect of TRH on LH secretion has been described (Franchimont, 1972). Anderson *et al.* (1971) found small increases in both luteinizing hormone (LH) and FSH following TRH injections, but only the LH increases were statistically significant. The present paper describes small, but statistically significant increases in serum concentrations of FSH, but not LH, during 4 h constant TRH infusions in normal men.

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MATERIALS AND METHODS

Twelve normal men aged 20-33 years were studied. None had a history of endocrine or serious medical disease. All had normal basal concentrations of LH, FSH, TSH, prolactin, testosterone, triiodothyronine (T3) and thyroxine (T4) by radioimmunoassay (Bremner *et al.*, 1977).

Synthetic TRH (supplied courtesy of Roche, Australia Ltd.) was administered to ten subjects in a dosage of 2.0 $\mu\text{g}/\text{min}$. TRH was dissolved in 0.9% saline containing 1% human serum albumin. The same solvent without added TRH was infused into four men, two of whom received both saline and TRH infusions, separated by several weeks. One additional man received a TRH infusion after having consumed diethylstilboestrol, 60 mg per day for more than 6 months in the course of an attempted sex reversal. Infusions were administered through an indwelling needle in an arm vein using an LKB infusion pump under continuous supervision. Studies began between 08.00 and 10.00 hours and all infusions lasted 4 h. Subjects were allowed to eat as they wished and were usually supine throughout the study.

Blood was sampled through an indwelling venous needle from the opposite arm. Two blood samples 30 min apart were obtained prior to the infusions; sampling was continued at 30 min intervals throughout the infusions. Blood was allowed to clot and the serum was separated by centrifugation and frozen at -20°C until assayed. LH, FSH (Alford *et al.*, 1973) and TSH (Patel *et al.*, 1971) were measured by radioimmunoassay. The first antibody used in the FSH assay was supplied by the National Institute of Arthritis, Metabolism and Digestive Diseases of the United States (NIAMDD). The specificity of the FSH assay was tested with preparations of TSH (Research Standard A of the Medical Research Council of Great Britain), prolactin (VLS#3, supplied by the NIAMDD) and the synthetic TRH used in the infusions.

Statistical significance was tested by Student's *t* test for paired observations and by Duncan's new multiple range test (Duncan, 1955).

RESULTS

Serum concentrations of FSH increased by approximately 50% ($P < 0.05$) above basal values during TRH infusions in the normal men (Fig. 1). FSH concentrations were significantly increased above basal values by the first measurement at 30 min after beginning the TRH infusions and remained above basal values until the infusions were completed at 4 h. Similar increases in FSH were not seen in the saline infusions and no significant changes in serum concentrations of LH were found in either the TRH or saline infusions (Fig. 1).

Since serum concentrations of TRH, TSH and prolactin increased markedly during the TRH infusions (Bremner *et al.*, 1977) the possible cross-reactivity of these hormones in the FSH assay was tested (Fig. 2). TRH and prolactin exhibited no cross-reactivity in concentrations much higher than those found in the present infusions. The TSH preparation in high concentrations, cross-reacted in the FSH assay. The maximal concentrations of TSH achieved in the present infusions (24.0 ± 3.2 , mean \pm SEM) however, would have measured less than 0.2 mu/ml in the FSH assay and therefore TSH cross-reactivity could not have accounted for the FSH increases found. In addition, a portion of the cross-reactivity of standard pituitary TSH in the FSH assay may have been due to FSH contained in the TSH preparation (stated to be less than 0.9 mu FSH (Steelman-Pohley bioassay) per 50 μu TSH).

The results obtained in one subject who received a TRH infusion while consuming high

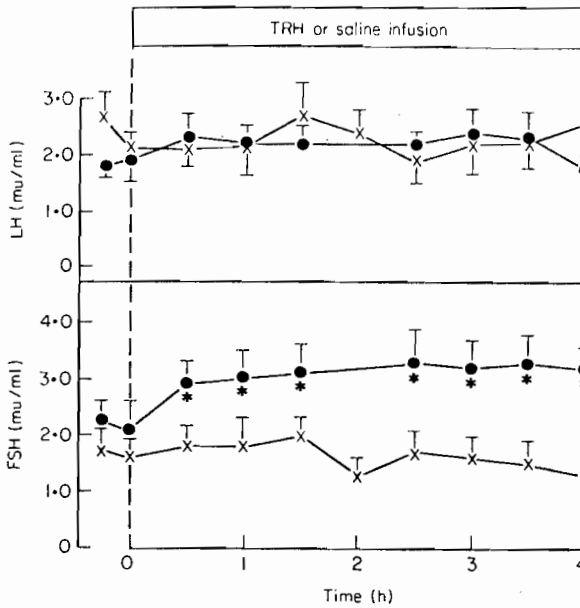


Fig. 1. Serum concentrations of LH and FSH (mean \pm SEM) before and during infusion of TRH (2.0 μ g/min) (●) or saline (X) into normal men. Asterisk denotes significantly greater than basal values ($P < 0.05$).

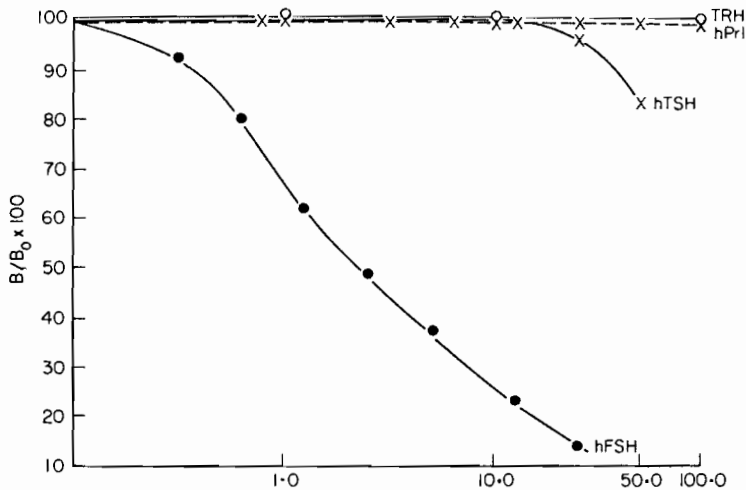


Fig. 2. Cross-reactivity of various hormonal preparations in the FSH assay. Units on the horizontal axis as follows: hFSH (μ u/ml), hTSH (μ u/ml), hPrI (ng/ml), TRH (μ g/ml).

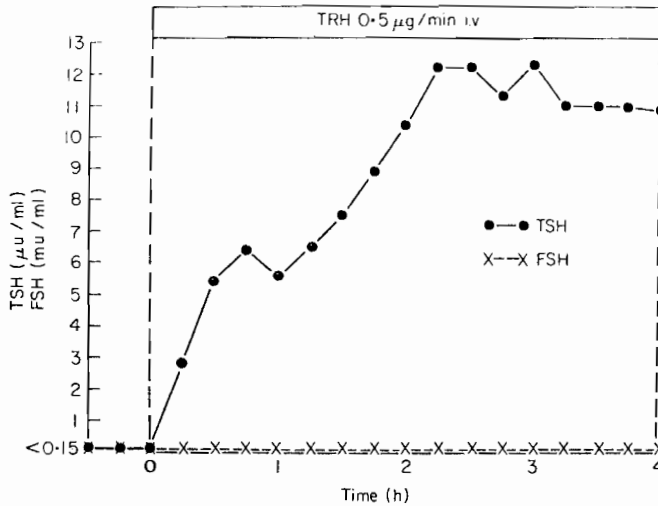


Fig. 3. Serum concentrations of TSH (●) and FSH (X) during a TRH infusion in a man receiving high dosages of oestrogens.

dosage oestrogens were further evidence against TSH cross-reactivity as an explanation for the increases observed in serum FSH levels (Fig. 3). A large increase occurred in serum TSH concentrations with an early peak at 45 min and a later peak at 2–3 h (Bremner *et al.*, 1977). In spite of the increase in serum TSH, FSH levels were consistently undetectable.

DISCUSSION

The present studies confirm that small, but statistically significant increases in serum FSH concentrations occur during prolonged infusions of TRH into normal men (Mortimer *et al.*, 1974). These increases in immunoassayable FSH could not be explained on the basis of cross-reactivity with TSH, prolactin and TRH which also increase during TRH infusions. No similar changes occurred in serum FSH concentrations during saline infusions nor in serum LH concentrations during either TRH or saline infusions.

The reason that increases in serum FSH were found in the present study and those of Mortimer *et al.* (1973, 1974) but not by most other investigators (Burger & Patel, 1977) is not obvious. Clearly, the small degree of the FSH increment could have contributed to its not being recognized. Administration of TRH as a prolonged infusion rather than as a single injection may be of importance since most of the studies showing a lack of TRH effect on FSH secretion, including one from this laboratory (Patel & Burger, 1972), have used single injections. However, Mortimer *et al.* (1973) also reported FSH increases following single injections of TRH.

The physiological importance of the small increases in FSH noted during TRH infusions is questionable. It is of interest, however, that prolonged primary hypothyroidism, which may cause increases in endogenous TRH production, is occasionally associated with precocious puberty (Hayles & Cloutier, 1972).

Other examples of the non-specificity of hypothalamic hormones for their respective pituitary hormones may be of more physiological importance. The development of the capacity of cell membrane receptors on somatotrophs to respond to TRH and LHRH

(Matsukura *et al.*, 1977) could be important in the aetiology of some cases of acromegaly. Similar alterations in specificity could be important in the development of Cushing's disease, Nelson's syndrome and gonadotrophin-producing pituitary tumours. A potentially important inhibitory effect of TRH on arginine and insulin-induced growth hormone secretion in normal subjects has been reported recently by Maeda *et al.* (1976). Increasing awareness of the diverse interactions of hypothalamic hormones on pituitary hormone secretion necessitates that some caution be exercised in interpreting the results of 'combined' pituitary function tests which use simultaneous administration of TRH, LHRH and insulin or arginine. It may be that administration of TRH as a prolonged infusion, as done in this and other studies (Mortimer *et al.*, 1974; Maeda *et al.*, 1976) is more likely to lead to non-specificity of its effects than its use as a single injection.

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