AN ELECTROENCEPHALOGRAPHIC INVESTIGATION OF SHORT-TERM EFFECTS OF THREE HYPOTHALAMIC HORMONES (TRH, LH/FSH-RH, GH-RIH) IN NORMAL SUBJECTS

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1. Three hypothalamic regulatory hormones, thyrotrophin-releasing hormone (TRH), luteinizing hormone/follicle-stimulating hormone-releasing hormone (LH/FSH-RH) and growth hormone-release inhibiting hormone (GH-RIH) given intravenously had no effect on the electroencephalographic response known as the contingent negative variation (CNV) in normal subjects.
2. TRH was given as a 10 ml infusion of 600 μ g over 8 min to six subjects. This produced subjective sensations and a rise in heart rate but no significant alteration of CNV magnitude.
3. LH/FSH-RH was given in a dose of 200 μ g in 10 ml over 2 min to six subjects. This had no effect on CNV magnitude or heart rate and produced no subjective effects.
4. GH-RIH was given as a 10 ml infusion of 250 μ g over 10 min to six subjects. Again there was no alteration in the magnitude of the CNV; the heart rate was slowed.

Introduction

Small polypeptide and probably other non-peptide hormones released from the hypothalamus control anterior pituitary function and can be regarded as neuroendocrine transducers since they translate neurologically transmitted signals into hormonal output. Since the reports that one of these peptides, thyrotrophin-releasing hormone has a psychopharmacological action in man (Prange, Wilson, Lara, Alltop & Breese, 1972; Kastin, Ehrensing, Schalach & Anderson, 1972) it has been suggested that many hypothalamic hormones may have a physiological role in the control of mood and behaviour (Lancet, 1972). However, there is little information concerning any objective effects of these hormones on brain activity.

The electroencephalographic phenomenon known as the contingent negative variation (CNV) (Walter, Cooper, Aldridge, McCallum and Winter, 1964) has recently been shown to be a useful objective measure of certain effects of centrally acting drugs on the brain in man (Ashton, Millman, Telford & Thompson, 1974). The CNV is a special type of cortical evoked response consisting of a slow negative potential which develops between a warning signal and an

imperative signal requiring a response from the subject. It probably arises in part from the ascending reticular activating system which, through its neural connections, elicits potential changes in the cortex that can be measured with scalp electrodes (Rebert, 1972; Rebert & Knott, 1970). It appears to reflect the degree of alertness, attention and arousal of the subject (McCallum & Walter, 1968; Tecce, 1972). The magnitude of the CNV is consistently increased by central stimulant drugs such as caffeine (Ashton *et al.*, 1974), pemoline (Ashton, Millman, Telford & Thompson, unpublished observations) and dextro-amphetamine (Hollister, 1974) and decreased by central depressant drugs such as nitrazepam (Ashton *et al.*, 1974), alcohol (Kopell, Tinklenberg & Hollister, 1972) and barbiturates (Hollister, 1974). It is also significantly altered by cigarette smoking (Ashton *et al.*, 1974), intravenous nicotine (Ashton, Millman, Telford & Thompson, 1975), tetrahydrocannabinol (Kopell *et al.*, 1972) and LSD (Hollister, 1974). It therefore seemed possible that any central effects of hypothalamic peptide hormones might be reflected in changes in CNV magnitude.

This paper describes an investigation of the effects of three regulatory hormones: TRH, LH/FSH-RH and GH-RIH on the CNV in normal subjects.

Methods

Subjects

The subjects were healthy volunteers who were mainly members of the hospital staff. There were twelve males and four females aged 20-35 years. Some subjects took part in more than one experiment. Informed consent was obtained from the subjects and approval was obtained from the appropriate Ethical Committees.

Treatment and dosage

The regulatory hormones were given as 10 ml intravenous infusions in physiological saline. TRH was given in a dose of 600 μ g over 8 min, LH/FSH-RH in a dose of 200 μ g over 2 min, and GH-RIH in a dose of 250 μ g over 10 min. Control infusions of saline delivered at the same rate were given once or twice before each hormone infusion. The subjects did not know whether they were receiving saline or hormone.

CNV recording

The CNV was obtained by the method described by Ashton *et al.* (1974). Briefly, subjects were presented with series of paired signals. The warning signal was a momentary flash of light and the imperative signal was a tone delivered through a loud-speaker. The subject was required to press a button in response to the imperative signal, giving a measure of reaction time. The paired warning and imperative signals were separated by an interval of 1.25 s and were presented at random intervals (4-8 s) in a series of 10, each series lasting 1 min 10 seconds. The EEG was derived between the left mastoid and vertex positions from two silver/silver chloride stick-on electrodes and was amplified by a Beckman Type S Dynograph recorder; an earth electrode was placed on the right mastoid. The output was fed into a CAT (Computer for the Averaging of Transients) and the averaged response to each series of ten paired signals was traced out by an X-Y recorder. The magnitude of the CNV thus obtained for a series of ten paired signals was determined in terms of area and expressed in μ V seconds. A typical CNV is shown in Figure 1.

Figure 1 Typical CNV record from one subject derived from a series of ten signal pairs. S_1 = flash stimulus which causes visual evoked response = VER, S_2 = tone stimulus which causes auditory evoked response = AER. The CNV is situated between the VER and AER and bounded by the record above and the heavy horizontal and vertical lines. Calibration: 500 ms and 15 μ V negative upwards; CNV is measured as μ Vs.

Other measurements

Heart rate was recorded from an ECG in some experiments and monitored continuously from a Devices rate meter. All subjects also completed a personality inventory (Eysenck & Eysenck, 1964) and a motivation scale (Lynn, 1969). At the end of the experiment they were asked about any subjective effects experienced during the infusions and their observations noted.

Procedure

The subjects lay on a couch in the Subject Room. A needle was inserted into an antecubital vein and connected to a syringe which delivered a slow saline infusion. Scalp and chest electrodes were applied for EEG and ECG recordings. The leads from the subject led through cables to a separate Control Room where all recording equipment was housed. Subjects could be observed through a one-way window and via closed circuit television. One observer, who was administering the infusions, remained in the Subject Room but was in communication with the Control Room via a microphone and earphone.

Instructions for the CNV procedure were given from a tape recording and the subject performed a practice run responding to ten pairs of signals. The experiment was started after the subject had been resting for 30 min and his heart rate was steady. Timed infusions of either saline or hormone were then given. CNV recordings were taken three times at intervals of 2-3 min during the 8 and 10 min infusions of the TRH and GH-RIH and their saline controls. A further CNV recording was made 5 min after the end of the infusions. For the 2 min

infusions of LH/FSH–RH and the corresponding saline controls, three CNV recordings were made during the 6 min immediately after each infusion in most subjects, although in some subjects recording was continued for 15 min and in one subject the CNV was measured 4 h after LH/FSH–RH infusion.

Results

TRH experiments

Six subjects (five males and one female) received 8 min infusions of 600 µg TRH. Two further male subjects received 15 and 30 min infusions of 600 µg. The results on the CNV are shown in Table 1. Mean CNV during 8 min saline infusion was 9.26 µVs; that during 8 min TRH infusion was 8.73 µVs and after the infusions it was 9.00 µVs. Student's *t*-tests showed no significant difference between any of these values. The two subjects who received longer infusions also showed no significant change in CNV magnitude. In the subject who had a 30 min infusion CNV magnitude was measured at 5 min intervals for 75 min after the start of TRH infusion and still showed no change. It was concluded that under these conditions, TRH had no effect on the magnitude of the CNV. It is possible that changes in serum levels of T₃ (triiodothyronine) or T₄

(thyroxine) caused by TRH could affect the CNV. However, according to Lawton, Ellis & Sufi (1973), T₃ levels start to rise at about 30 min and do not reach a peak until 2–4 h after intravenous TRH, and T₄ is variable, not always rising after 3 h. The effects of such long term influences would not have been detected in the present experiments.

A significant increase in heart rate ($P < 0.001$) was, however, observed during TRH infusion, as shown on Table 2. In addition, five of the eight subjects reported subjective effects during infusion, including flushing, palpitation, nausea and urethral pain, effects which have been described before and which are due in part to a direct action on smooth muscle (Ormston, 1972). It was thought that these symptoms could account for the slight tendency of CNV magnitude to decrease during infusion.

LH/FSH–RH experiments

Six subjects (two males and four females) received 2 min infusions of 200 µg of LH/FSH–RH; one male subject received 400 µg in 2 min. The results on the CNV are shown in Table 3. Mean CNV after saline infusion was 7.69 µVs and after LH/FSH–RH 6.84 µVs. There was no significant difference between the values. Two of the female subjects (1 and 4) were taking an oral contraceptive pill, the other two were both measured during

Table 1 Effect of TRH on CNV magnitude

Subject	Sex	Mean CNV magnitude (µVs)			Subjective effects during infusion
		Saline	TRH (8 min infusion)	Post TRH	
1	M	9.77	8.92	13.75	+
2	F	11.90	11.61	—	0
3	M	3.30	4.89	6.04	++
4	M	8.86	6.83	8.43	0
5	M	10.51	7.98	5.62	+
6	M	11.24	12.12	11.15	+
	Mean	9.26	8.73	9.00	
	s.d.	3.11	2.78	3.45	
			(15 min infusion)		
7	M	8.61	6.5	8.45	0
			(30 min infusion)		
8	M	9.49	7.86	7.88	++
	Mean	9.21	8.43	8.76	
(Subjects 1–8)	s.d.	2.64	2.49	2.85	

TRH given as an i.v. infusion of 600 µg in 10 ml.

t-tests: saline *v* TRH infusion (subjects 1.6) $t = 0.8212$ (NS).
(subjects 1.8) $t = 1.6475$ (NS).

the middle of the menstrual cycle. CNV readings taken 15 min after infusion in two subjects and 4 h later in one female subject showed no change. Thus in these subjects LH/FSH–RH had no effect on CNV magnitude. There were no subjective effects following infusion and no effect on the heart rate.

GH–RIH experiments

Six male subjects received 10 min infusions of 250 µg GH–RIH; one further subject received 375 µg. In this experiment two 10 min control infusions of saline were given before infusion of GH–RIH. The results on the CNV are shown in

Table 2 Effect of TRH on heart rate.

Subject*	Sex	Mean heart rate (beats/min)		
		Saline	TRH (8 min infusion)	Post TRH
1	M	67.50	76.36	72.25
2	F	74.38	82.08	—
3	M	74.17	82.05	71.67
4	M	68.89	76.18	68.75
	Mean	71.24	79.17	70.97
	s.d.	3.56	3.35	1.97
(30 min infusion)				
8	M	66.54	69.41	65.00
	Mean	73.62	77.22	69.48
(Subjects 1-6)	s.d.	7.58	5.24	3.39

*Same subjects as in Table 1.

t-tests: saline *v* TRH infusion (subjects 1-4) $t = 23.8356$; $P < 0.001$.
(subjects 1-4, 8) $t = 6.6239$; $P < 0.01$.

Table 3 Effect of LH/FSH–RH on CNV magnitude.

Subject	Sex	(Mean CNV Magnitude (µVs))	
		Saline	Post LH/FSH–RH (200 µg)
1	F(p)	7.41	8.44
2	F	5.57	4.98
3	F	11.40	7.51
4	F(p)	6.54	5.11
5	M	9.10	7.80
6	M	6.09	7.17
	Mean	7.69	6.84
	s.d.	2.20	1.45
(400 µg)			
7	M	6.81	6.82
	Mean	7.56	6.83
	(1-7)		
	s.d.	2.04	1.32

LH/FSH–RH given as a 2 min i.v. infusion of 200 µg in 10 ml.

(p) = taking oral contraceptive

t-tests: saline *v* LH/FSH–RH (subjects 1-6) $t = 1.1256$ (NS)
(subjects 1-7) $t = 1.1187$ (NS).

Table 4 Effect of GH-RIH on CNV magnitude

		Mean CNV magnitude (μ Vs)			
Subject	Sex	Saline 1	Saline 2	GH-RIH (250 μ g)	Post GH-RIH
1	M	4.75	3.47	2.70	3.74
2	M	4.94	5.55	4.02	3.61
3	M	4.81	5.25	5.95	5.45
4	M	9.11	6.68	7.95	6.13
5	M	7.08	4.78	4.58	5.57
6	M	10.92	10.99	7.91	3.34
	Mean	6.94	6.12	5.52	4.64
	s.d.	2.60	2.60	2.14	1.21
				(375 μ g)	
7	M	9.60	7.74	7.71	10.09
	Mean	7.32	6.35	5.83	5.42
	(1-7)				
	s.d.	2.58	2.46	2.12	2.34

GH-RIH given as 10 min i.v. infusion of 250 μ g in 10 ml.

t-tests: Saline 1 v Saline 2: *t* = 1.4548 (NS).

(Subjects 1-6)

Saline 1 v GH-RIH: *t* = 2.3438 (NS).

Saline 2 v GH-RIH: *t* = 0.9361 (NS).

Saline 2 v post GH-RIH: *t* = 1.4430 (NS).

Saline 1 v post GH-RIH: *t* = 1.9823 (NS).

Table 5 Effects of synacthen and hydrocortisone on CNV magnitude

Subject	Sex	Mean CNV magnitude (μ Vs)				
		Saline	Time after synacthen infusion (min)			
			0	3	6	Mean 0-6
1	M	10.27	15.63	8.14	9.95	11.24
2	M	8.50	9.89	5.30*	5.15	6.78
3	M	9.94	11.64	13.91	9.68	11.75
4	M	7.94	4.42*	4.92	4.22	4.52
5	F	11.40	11.80	7.40	12.27	10.49
	Mean	9.61	10.68	7.93	8.25	8.96
	s.d.	1.39	4.08	3.61	3.43	3.15

(*Subjects complained of nausea at the time shown).

		Time after hydrocortisone infusion (min)				
1	M	8.50	9.58	9.37	10.20	9.72
2	M	9.94	9.48	14.99	5.77	10.08
3	M	7.94	7.49	8.94	8.64	8.36
	Mean	8.79	8.85	11.10	8.20	9.39
	s.d.	1.03	1.18	3.38	2.25	0.91

Synacthen given as i.v. infusion of 0.25 mg in 5 ml over 2 min.

t-tests: saline v after infusion (mean 0-6 min): 0.6979 (NS).

saline v 0, 3, 6 min : all NS.

Hydrocortisone hemisuccinate given as i.v. infusion of 100 mg in 5 ml over 2 min.

t-tests: saline v after infusion (mean 0-6 min): 1.8340 (NS).

saline v 0, 3, 6 min : all NS.

Table 4. Mean CNVs during the two saline infusions were 6.94 and 6.12 μ Vs; during and after GH-RIH infusion the mean CNV was 5.52 and 4.64 μ Vs respectively. There was no significant difference between any of these values. The tendency for the CNV magnitude to decline over time is well known and the slightly lower values during and after GH-RIH were probably due to this time factor. However, it was not thought advisable to randomize the sequence of experiments because of possible carryover effects of the hormone.

No subjective effects were experienced but a decrease in heart rate of about 10 beats/min was noted in all subjects during GH-RIH infusions.

Tetracosactrin and hydrocortisone experiments

Preliminary observations of the effects of tetracosactrin (Synacthen) and hydrocortisone on the CNV were also made in a few subjects. Five subjects received a 2 min infusion of tetracosactrin (0.25 mg) in buffered saline (5 ml) and three subjects received a 2 min infusion of hydrocortisone hemisuccinate (100 mg) in 5 ml. The CNV was measured during saline infusion before hormone administration and at 0, 3 and 6 min after the end of hormone infusion. The results are shown in Table 5. After tetracosactrin, there was no significant overall change in CNV magnitude. However, two subjects (2 and 4, Table 5) showed a short-lived increase in CNV magnitude. Further experiments would be required to determine whether or not these changes were important. The three subjects who received hydrocortisone also showed no significant overall change in CNV magnitude, but no conclusion could be drawn from such a small number of observations.

Personality data and reaction times

The personality data showed that subjects were within the normal range of extraversion, neuroticism and motivation. Reaction times usually improved during the course of the experiments, an effect probably due to learning. There was no correlation between CNV magnitude and reaction time or personality data.

Discussion

These results show that intravenous TRH, LH/FSH-RH and GH-RIH have no significant effect on CNV magnitude in normal subjects under these experimental conditions. Short-term effects were specifically investigated in this study since these peptides appear to be quickly metabolized;

for example the half-life of TRH (Redding, 1969) and GH-RIH (Lancet, 1974) is known to be less than 10 min. It is possible that longer term effects on brain activity may occur, although the few subjects tested at 1-4 h after peptide administration showed no change in CNV magnitude. The investigation was confined to normal subjects; the possibility that effects may be seen in certain hypothalamic deficiency states or in other abnormal conditions is not ruled out. The lack of the effect on normal subjects is unlikely to be due to insensitivity of the method used since the CNV has been shown to be a measure of brain activity sufficiently sensitive to show significant changes in magnitude with several psychoactive drugs given in doses too small to cause subjective effects (Ashton *et al.*, 1974). It is possible that the CNV, while showing changes in general arousal via the reticular activating system (Rebert, 1972; Rebert & Knott, 1970; McCallum & Walter, 1968; Tecce, 1972), may not detect subtle changes in mood and affect caused by alterations in limbic system activity. However, limbic system activity is thought to be involved in selectively motivated, goal-directed behaviour requiring attention (Routtenberg, 1968) and is thus likely also to influence CNV magnitude. In addition it seems unlikely that changes in mood and behaviour, as reported with some of the hypothalamic hormones (Lancet, 1972), would occur without concomitant changes in cortical arousal.

TRH is a potent stimulator of the release of thyroid stimulating hormone (Fleischer, Burgus, Vale, Dunn & Guillemin, 1970; Anderson, Bowers, Kastin, Schalach, Schally, Snyder, Utiger, Wilber & Wise, 1971) and prolactin (Bowers, Friesen, Hwang, Guyda & Folkers, 1971; Jacobs, Snyder, Wilber, Utiger & Daughaday, 1971) and has been widely used in man. Transient improvement in mental depression was first recorded in 1972 (Prange *et al.*, 1972; Kastin *et al.*, 1972) after an intravenous preparation of TRH and also after oral administration (Vis-Melsen & Wiener, 1972). However, the antidepressant effect of TRH has not been supported by further studies (Coppen, Montgomery, Peet, Bailey, Marks & Woods, 1974; Ehrensing, Kastin, Schalach, Friesen, Vargas & Schally, 1974; Mountjoy, Price, Weller, Hunter, Hall & Dewar, 1974; Hall, Hunter, Price & Mountjoy, 1975). The present results lend some support to these negative findings although it is possible that TRH has effects in depressed patients which do not occur in normal subjects. Suggestions of improvement in behaviour problems (Tiwary, Frias & Rosenbloom, 1972) and psychoses (Wilson, Lara & Prange, 1972) following TRH and of a stimulating effect upon human cerebral biopotentials recorded from the scalp and

analysed by computer (Itil, 1974) require confirmation.

LH/FSH-RH (Schally, Baba, Arimura, Redding & White, 1971) stimulates the release of both luteinizing hormone and follicle stimulating hormone in men, women and children (Besser, McNeilly, Anderson, Marshall, Harsoulis, Hall, Ormston, Alexander & Collins, 1972). This hormone has been held responsible for aggressive behaviour in birds (Matthewson, 1961) and has been shown to induce mating behaviour in female rats (Wilson *et al.*, 1972; Moss & McCann, 1973; Pfaff, 1973). LH/FSH-RH may also cause improvement in abnormal human behaviour (Tiwary, 1974). If this hormone were shown to have a significant central action in man, the implications would be far-reaching. The present experiments, however, showed no effect on the CNV in normal men, nor in normal women whether or not they were taking oral contraceptives.

GH-RIH or somatostatin is the latest of the hypothalamic regulatory hormones to have become available (Brazeau, Vale, Burgus, Ling, Butcher, Rivier & Guillemin, 1973; Coy, Coy, Arimura & Schally, 1973). It has been shown to have a wide range of actions in pharmacological doses, including lowering of plasma growth

hormone levels and reduced secretion of growth hormone in response to various stimuli (Hall, Besser, Schally, Coy, Evered, Goldie, Kastin, McNeilly, Mortimer, Phenekos, Tunbridge & Weightman, 1973; Hansen, Ørskov, Seyer-Hansen & Lundbaed, 1973; Parker, Rossman, Siler, Rivier, Yen & Guillemin, 1974; Mortimer, Tunbridge, Carr, Yeomans, Lind, Coy, Bloom, Kastin, Mallinson, Besser, Schally & Hall, 1974; Siler, Vandenberg, Yen, Brazeau, Vale & Guillemin, 1973) as well as reduced secretion of insulin, glucagon and gastrin (Siler *et al.*, 1973); Alberti, Christensen, Christensen, Engkjaer, Hansen, Iversen, Jundbaek, Seyer-Hansen & Ørskov, 1973; Bloom, Mortimer, Thorner, Besser, Hall, Gomez-Pan, Roy, Russell, Coy, Kastin & Schally, 1974). Its role in central neuro-endocrinology has not been defined, but the present results suggest that it has no effect on cortical activity as measured by the CNV. The previously noted bradycardia (Lancet, 1974) is confirmed.

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References

- ALBERTI, K.G.M.M., CHRISTENSEN, N., CHRISTENSEN, S., ENKJAER, HANSEN, Aa., IVERSEN, J., LUNDBAEK, K., SEYER-HANSEN, K. & ØRSKOV, H. (1973). Inhibition of insulin secretion by somatostatin. *Lancet*, ii, 1299-1301.
- ANDERSON, M.S., BOWERS, C.Y., KASTIN, A.J., SCHALACH, D.S., SCHALLY, A.V., SNYDER, P.J., UTIGER, R.D., WILBER, J.F. & WISE, A.J. (1971). Synthetic thyrotropin-releasing hormone. *New Eng. J. Med.*, 285, 1279-1283.
- ASHTON, H., MILLMAN, J.E., TELFORD, R. & THOMPSON, J.W. (1974). The effect of caffeine, nitrazepam and cigarette smoking on the contingent negative variation in man. *Electroenceph. clin. Neurophysiol.*, 37, 59-71.
- ASHTON, H., MILLMAN, J.E., TELFORD, R. & THOMPSON, J.W. (1975). The contingent negative variation: an objective electroencephalographic method for measuring the effects of centrally acting drugs on brain activity in man. *Br. J. Pharmac.*, 54, 177-178P.
- BESSER, G.M., MCNEILLY, A.S., ANDERSON, D.C., MARSHALL, J.C., HARSOULIS, P., HALL, R., ORMSTON, B.J., ALEXANDER, L. & COLLINS, W.P. (1972). Hormonal responses to synthetic luteinizing hormone and follicle stimulating hormone-releasing hormone in man. *Br. med. J.*, 3, 267-271.
- BLOOM, S.R., MORTIMER, C.H., THORNER, M.O., BESSER, G.M., HALL, R., GOMEZ-PAN, A., ROY, V.M., RUSSELL, R.C.G., COY, D.H., KASTIN, A.J. & SCHALLY, A.V. (1974). Inhibition of gastrin and gastric-acid secretion by growth-hormone release-inhibiting hormone. *Lancet*, ii, 1106-1109.
- BOWERS, C.Y., FRIESEN, H.G., HWANG, P., GUYDA, H.J. & FOLKERS, K. (1971). Prolactin and thyrotropin release in man by synthetic pyroglutamyl-histidyl-prolinamide. *Biochem. Biophys. res. Comm.*, 45, 1033-1041.
- BRAZEAU, P., VALE, W., BURGUS, R., LING, N., BUTCHER, M., RIVIER, J. & GUILLEMIN, R. (1973). Hypothalamic polypeptide that inhibits the secretion of immunoreactive pituitary growth hormone. *Science*, 179, 77-79.
- COPPEN, A., MONTGOMERY, S., PEET, M. & BAILEY, J. (1974). Thyrotropin-releasing hormone in the treatment of depression. *Lancet*, ii, 433-435.
- COY, D.H., COY, E.J., ARIMURA, A. & SCHALLY, A.V. (1973). Solid phase synthesis of growth hormone-release inhibiting factor. *Biochem. Biophys. res. Comm.*, 54, 1267-1273.
- EHRENSING, R.H., KASTIN, A.J., SCHALACH, D.S., FRIESEN, H.G., VARGAS, J.R. & SCHALLY, A.V. (1974). Affective state and thyrotropin and prolactin responses after repeated injections of thyrotropin-

- releasing hormone in depressed patients. *Am. J. Psychiat.*, 131, 714-718.
- EYSENCK, H.J. & EYSENCK, S.B.G. (1964). *Manual of the Eysenck Personality Inventory*. London: University of London Press.
- FLEISCHER, N., BURGUS, R., VALE, W., DUNN, T. & GUILLEMIN, R. (1970). Preliminary observations on the effect of synthetic thyrotropin releasing factor on plasma thyrotropin levels in man. *J. clin. Endocr. Metab.*, 31, 109-112.
- HALL, R., BESSER, G.M., SCHALLY, A.V., COY, D.H., EVERED, D., GOLDIE, D.J., KASTIN, A.J., McNEILLY, A.S., MORTIMER, C.H., PHENEKOS, C., TUNBRIDGE, W.M.G. & WEIGHTMAN, D. (1973). Action of growth-hormone-release inhibitory hormone in healthy men and in acromegaly. *Lancet*, ii, 581-584.
- HALL, R., HUNTER, P.R., PRICE, J.S. & MOUNTJOY, C.Q. (1975). Thyrotrophin-releasing hormone in depression. *Lancet*, i, 162.
- HANSEN, P.A., ØRSKOV, H., SEYER-HANSEN, K. & LUNDBAEK, K. (1973). Some actions of growth hormone release inhibiting factor. *Br. med. J.*, 3, 523-524.
- HOLLISTER, L.E. (1974). In *Research Advances in Alcohol and Drug Problems*, Vol. 1, ed. Gibbins, R.J., Israel, Y., Kalant, H., Popham, R.E., Schmidt, W. & Smart, R.G. p 253. New York: John Wiley & Sons.
- ITIL, T.M. (1974). Effects of steroid hormones and peptides on human brain function. Commun: IX Congr. Coll. Int. Neuropsychopharmac., Paris.
- JACOBS, L.S., SNYDER, P.J., WILBER, J.F., UTIGER, R.D. & DAUGHADAY, W.H. (1971). Increased serum prolactin after administration of synthetic thyrotropin releasing hormone (TRH) in man. *J. clin. Endocr. Metab.*, 33, 996-998.
- KASTIN, A.J., EHRENSING, R.H., SCHALACH, D.S. & ANDERSON, M.S. (1972). Improvement in mental depression with decreased thyrotropin response after administration of thyrotropin-releasing hormone. *Lancet*, ii, 740-742.
- KOPPEL, B.S., TINKLENBERG, J.R. & HOLLISTER, L.E. (1972). Contingent negative variation amplitudes: Marijuana and alcohol. *Archs. gen. Psychiat.*, 27, 809-811.
- LANCET, LEADER. (1972). Thyrotrophin-releasing hormone and depression. *Lancet*, ii, 1015-1016.
- LANCET, LEADER. (1974). Growth hormone release-inhibiting hormone. *Lancet*, i, 1148-1150.
- LAWTON, N.F., ELLIS, S.M. & SUFI, S. (1973). The triiodothyronine and thyroxine response to thyrotrophin-releasing hormone in the assessment of the pituitary-thyroid axis. *Clin. Endocr.*, 2, 57-63.
- LYNN, R. (1969). An achievement motivation questionnaire. *Br. J. Psychol.*, 60, 529-534.
- MATTHEWSON, S.F. (1961). Gonadotrophic hormones affect aggressive behaviour in starlings. *Science*, 134, 1522-1523.
- MCCALLUM, W.C. & WALTER, W.G. (1968). The effects of attention and distraction on the contingent negative variation in normal and neurotic subjects. *Electroenceph. clin. Neurophysiol.*, 25, 319-329.
- MORTIMER, C.H., TUNBRIDGE, W.M.G., CARR, D., YEOMANS, L., LIND, T., COY, D.H., BLOOM, W.R., KASTIN, A., MALLINSON, C.N., BESSER, G.M., SCHALLY, A.V. & HALL, R. (1974). Effects of growth-hormone release-inhibiting hormone on circulating glucagon, insulin and growth hormone in normal, diabetic, acromegalic and hypo-pituitary patients. *Lancet*, i, 697-701.
- MOSS, R.L. & McCANN, S.M. (1973). Induction of mating behaviour in rats by luteinizing hormone-releasing factor. *Science*, 181, 177-179.
- MOUNTJOY, C.Q., PRICE, J.S., WELLER, M., HUNTER, P., HALL, R. & DEWAR, J.H. (1974). A double-blind crossover sequential trial of oral thyrotrophin-releasing hormone in depression. *Lancet*, ii, 958-960.
- ORMSTON, B.J. (1972). In *Frontiers in Hormone Research*, Vol. 1, ed. Karger, S. pp. 42-43. Basel.
- PARKER, D.C., ROSSMAN, L.G., SILER, T.M., RIVIER, J., YEN, S.S.C. & GUILLEMIN, R. (1974). Inhibition of the sleep-related peak in physiologic human growth hormone release by somatostatin. *J. clin. Endocr. Metab.*, 38, 496-499.
- PFAFF, D.W. (1973). Luteinizing hormone-releasing factor potentiates lordosis behaviour in hypophysectomized ovariectomized female rats. *Science*, 182, 1148-1149.
- PRANGE, A.J., WILSON, I.C., LARA, P.P., ALLTOP, L.B. & BREESE, G.R. (1972). Effects of thyrotrophin-releasing hormone in depression. *Lancet*, ii, 999-1002.
- REBERT, C.S. (1972). Cortical and subcortical slow potentials in the monkey's brain during a preparatory interval. *Electroenceph. clin. Neurophysiol.*, 33, 389-402.
- REBERT, C.S. & KNOTT, J.R. (1970). The vertex non-specific evoked potential and latency of contingent negative variation. *Electroenceph. clin. Neurophysiol.*, 28, 561-565.
- REDDING, T.W. (1969). Preparation of tritiated thyrotropin-releasing hormone (TRH) and its distribution and excretion in mice and rats. *Proc. 51st Meet. Endocr. Soc.*, Abstr. no. 70, 65.
- ROUTTENBERG, A. (1968). The two-arousal hypothesis: reticular formation and limbic system. *Psychol. Rev.*, 75, 51-80.
- SCHALLY, A.V., BABA, Y., ARIMURA, A., REDDING, T.W. & WHITE, W.F. (1971). Evidence for peptide nature of LH and FSH-releasing hormones. *Biochem. Biophys. res. Commun.*, 42, 50-56.
- SILER, T.M., VANDENBERG, S., YEN, S.S.C., BRAZEAU, P., VALE, W. & GUILLEMIN, R. (1973). Inhibition of growth hormone release in humans by somatostatin. *J. clin. Endocr. Metab.*, 37, 632-634.
- TECCE, J.J. (1972). Contingent negative variation (CNV) and psychological processes in man. *Psychol. Bull.*, 77, 73-108.
- TIWARY, C.M. (1974). Testosterone, L.H.R.H., and behaviour. *Lancet*, i, 993.
- TIWARY, C.M., FRIAS, J.L. & ROSENBLOOM, A.L. (1972). Response to thyrotrophin in depressed patients. *Lancet*, ii, 1086.
- VAN DER VIS-MELSEN, M.J.E. & WIENER, J.D. (1972). Improvement in mental depression with decreased thyrotropin response after administration of thyrotropin-releasing hormone. *Lancet*, ii, 1415.
- WALTER, W.G., COOPER, R., ALDRIDGE, J.V.,

McCALLUM, W.C. & WINTER, A.L. (1964). Contingent negative variation: An electric sign of sensorimotor association and expectancy in the human brain. *Nature (Lond.)*, **203**, (4943), 380-384.

WILSON, I.C., LARA, P.P. & PRANGE, A.J. (1973). Thyrotrophin-releasing hormone in schizophrenia. *Lancet*, **ii**, 43-44.

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