Decreased or Absent ACTH-Like Activity of Blocked Synthetic Tricosapeptide.* (26995)

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A synthetic tricosapeptide synthesized by Hofmann *et al.*(1) which duplicates the first 23 amino acids, beginning with the N terminus, of naturally occurring ACTH(2-5) has been shown to increase urinary excretion of 11-oxysteroids and of 17-ketosteroids in adult male prisoners and in a patient with scleroderma(6). Trials of a variant of this molecule with certain groups blocked by a variety of means have been undertaken in a search for steroidogenic, electrolyte, carbohydrate or other effects. In contrast to the ACTH-like activities of the unblocked parent tricosapeptide this molecule appears to be less active or inert.

Materials and Methods. Fig. 1 depicts the molecular structure of the active tricosapeptide and its blocked congener. In the deblocked molecule R, R′, and R″ refer to H^+, OH^-, and H^-, respectively; in the blocked molecule R, R′, and R″ represent acetyl, amino, and formyl groups, respectively(7,8).

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The blocked tricosapeptide amide, dissolved in the gelatin-saline base employed by Armour and Co. in preparation of commercial Acthar gel, was administered in aliquots which contained 6.7 mg of material. This dosage was therefore comparable to that of the unblocked tricosapeptide (6.4 mg of deblocked peptide with ACTH-like activity) per injection. Four healthy male adult prisoners served as subjects. Blood and sera obtained in the fasting state prior to and during the series of injections were analyzed for levels of sugar, NPN, CO_2, chloride, sodium, potassium, total protein, albumin, globulin, calcium, inorganic phosphorus, protein-bound iodine, total and alpha- and beta lipoprotein, cholesterol, phospholipids, triglycerides and non-esterified fatty acids (NEFA) by methods in regular use in these laboratories(6). Urinary excretion in refrigerated 24-hour specimens of 11-oxysteroids (Porter-Silber chromogens), 17-ketosteroids, and pressor amines was normal before and during therapy. Possible effects upon carbohydrate metabolism were sought by means of oral glucose tolerance tests (1.75 g of glucose per kilo of body weight) and intravenous

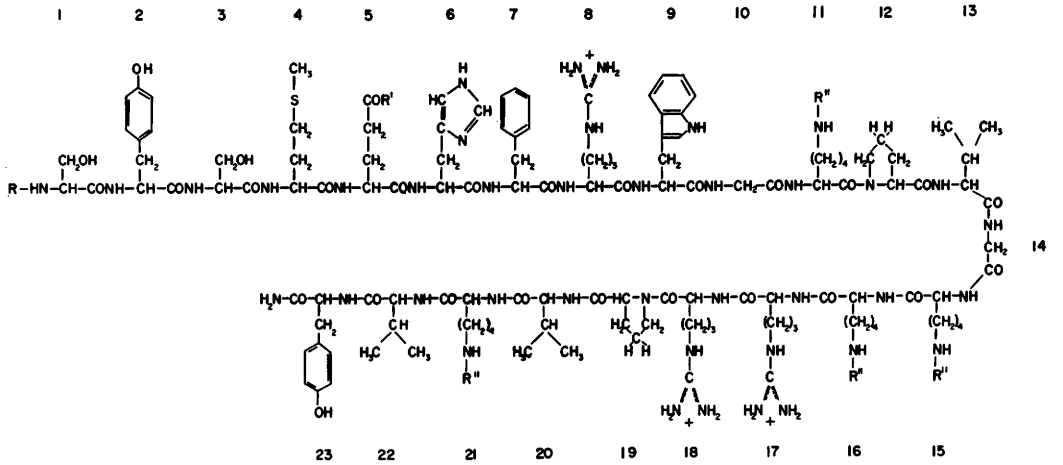
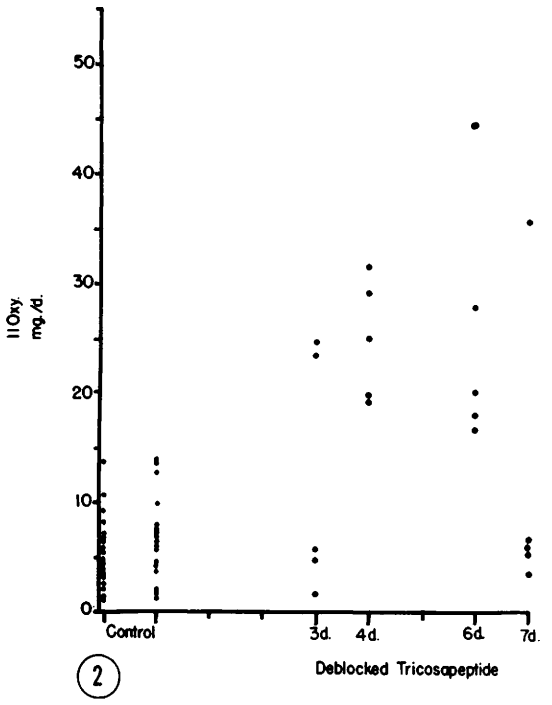


FIG. 1. Structure of Deblocked and Blocked Synthetic Tricosapeptide. In the deblocked molecule R, R′, and R″ refer to H^+, OH^-, and H^-, respectively; in the blocked molecule R, R′, and R″ represent acetyl, amino, and formyl groups, respectively.



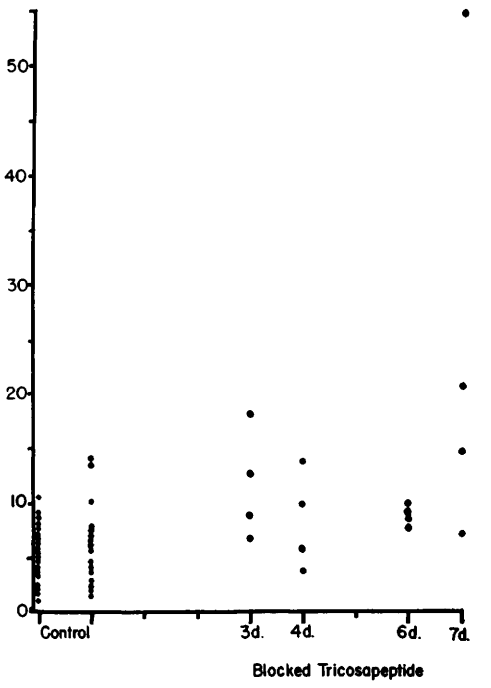
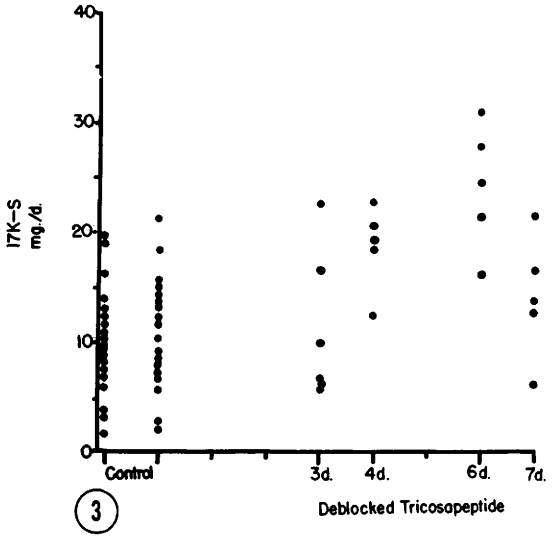


FIG. 2. Failure of the blocked tricosapeptide to induce increases in urinary 11-oxysteroids. In contrast to the rise in urinary 11-oxysteroids observe the following administration of the deblocked tricosapeptide. The blocked tricosapeptide in general had little if any effect.



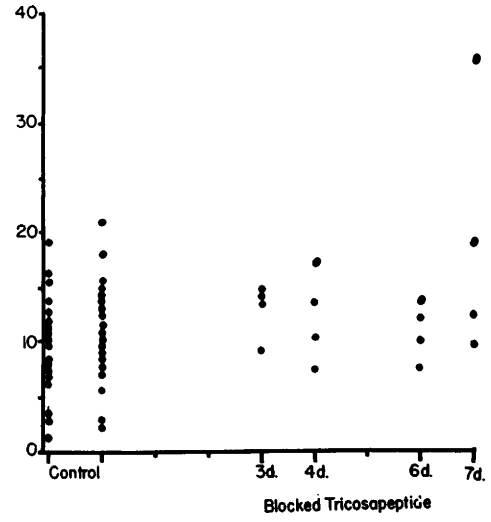


FIG. 3. Failure of blocked tricosapeptide to induce increases in urinary 17-ketosteroids. In contrast to the rise in urinary 17-ketosteroids observe the following administration of the deblocked tricosapeptide. The blocked tricosapeptide in general had little if any effect.

TABLE I. Mean Change in Serum Electrolytes

		CO_2 (meq/L)	Cl (meq/L)	K (meq/L)
Tricosapeptide (deblocked)				
5 subjects	6.7 mg* peptide/d/6d	+1.1	+1.10	−.44
	6.7 mg peptide/d/7d	+ .9	−1.54	−.64
Tricosapeptide (blocked)				
4 subjects	6.4 mg peptide amide/d/6d	−1.2	+2.82	−.60
	6.4 mg peptide amide/d/7d	−1.3	+1.88	−.28

*equals 160 subcutaneous adrenal ascorbic acid depleting units.

insulin tolerance tests (0.1 unit of commercial regular insulin per kilo of body weight).

Results. There were no increases in blood pressure or body weight nor did any of the subjects develop Cushingoid manifestations. The deblocked tricosapeptide increased the urinary 11-oxysteroids and to a lesser extent the 17-ketosteroids while administration of the blocked congener, with the exception of one specimen in one patient, did not produce these changes. The sole exception occurred after the seventh injection of the blocked tricosapeptide (Figs. 2, 3).

The serum electrolytes showed a slight rise in CO_2 and a decrease in chloride and potassium when the deblocked tricosapeptide was administered, in keeping with changes induced by commercial ACTH. With the blocked tricosapeptide only the serum potassium values decreased (Table I).

The glucose and insulin tolerance tests were not altered by the blocked tricosapeptide whereas the deblocked polypeptide may have manifested a slight hyperglycemic and perhaps anti-insulin effect(6). Neither was there any pattern of change in serum lipids elicited by the blocked molecule.

Summary. Substitution of acetyl for hydrogen in the NH_2 group of the N-terminal serine, NH_2 for OH^- in the carboxyl of glutamine in position 5, and formyl for an amino hydrogen of lysine in positions 11, 15, 16, and 21 decreases or eliminates the ACTH-like activity of a synthetic tricosapeptide which duplicates the first 23 amino acids of naturally occurring ACTH.

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Repopulation of Thymus by Immunologically Competent Cells Derived from Donor Marrow. (26996)

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The cortical region of the thymus becomes atrophic following X-irradiation(1); however, the thymus recovers weight rapidly in irradiated mice that receive an injection of isologous(2,3) or parental bone marrow(2). This recovery may result from repopulation of the thymus by implanted cells, although Kaplan

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