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The peptide encoded by angiotensin II complementary RNA does not interfere with angiotensin II action

Angiotensin II [Asp-Arg-Val-Tyr-Ile-His-Pro-Phe (AII)] is a major regulator of blood pressure. Its effects on blood vessels (vasoconstriction) and adrenal cortex (steroidogenesis) are mediated by specific receptors at the surface of its target cells. Recently it was shown that the octapeptide (IIA) specified by the RNA sequence complementary to the mRNA sequence for AII could inhibit the binding of ^{125}I -AII to its receptor present on rat adrenal particles [1]. That report together with a few others [2–8] (using different hormones) suggested that peptides encoded by complementary strands of nucleic acid (designated 'complementary peptides') have the ability to bind one another. The proposed hypothesis was that ligands and receptor binding sites can be derived from complementary sequences of nucleic acid. Since it may have important applications in the study of the mechanism of hormonal action, we decided to verify this interesting hypothesis.

The octapeptide IIA, Lys-Gly-Val-Asp-Val-Tyr-Ala-Val (specified by the nucleotide sequence of the rat angiotensinogen cDNA; see [1]) was synthesized by a standard solid phase method [9] and purified by h.p.l.c. The identity of the peptide was verified by FAB mass spectrometry and had a $(M + 1) = 851$ mass unit.

The steroidogenic action of AII is well known. In our system (for methods, see [10]), AII brought the level of aldosterone secretion by bovine adrenal glomerulosa cells (400 000 cells incubated for 2 h) from a basal value around 40 ng/ml to a maximal value around 200 ng/ml [EC_{50} (concentration producing 50 % of the maximal effect) 1.2 ± 1.1 nM, $n = 5$]. The complementary peptide IIA had no effect (at concentrations as high as 10 μM) on the basal secretion of aldosterone. In five different experiments, the steroidogenic effect of AII (1 nM) was not inhibited by increasing concentrations of IIA (up to 10 μM).

The ability of AII to contract rabbit aorta strips is well documented (for methods, see [11]). In our system, IIA (up to 10 μM) had no effect on the contraction of rabbit aorta strips. Most importantly, the responses to half-maximal doses of AII (2 ng/ml) were unchanged in the presence of increasing concentrations of IIA (up to 10 μM). In some experiments, in order to facilitate a putative interaction, AII and IIA were preincubated together for 15 min before addition to the baths. Despite this precaution, IIA did not modify the response of the aortae to AII.

Finally, the binding of ^{125}I -AII (120 000 c.p.m., ~ 1 nM) to bovine adrenal cortex membranes (for methods,

see [12, 13]) was not affected by concentrations of IIA as high as 10 μM . All dose-displacement curves performed in the presence or in the absence of 100 μM -IIA were superimposable (results repeated three times).

These results obtained with three different systems indicate that IIA does not interfere with AII action. The systems used by our group are different from the systems used by Elton *et al.* [1]. However, according to the proposed hypothesis, IIA should be a receptor for AII and should act by buffering the amount of available AII. In that respect, the system used to measure the amount of available AII is only an accessory which does not directly interact with IIA; the results should be identical whatever the system used.

Our results question the validity of the hypothesis that there is a molecular recognition code in which peptide ligands and their receptor binding sites can be encoded by complementary nucleotide sequences.

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