All these effects are qualitatively as well as quantitatively similar for both substances. We therefore conclude that they can be accounted for by the steric effect of the ortho-substituents rather than by changes in the dissociation of the hydroxyl group of tyrosine induced by a substituent located ortho to it.

Discussion. Figure 2 documents quite clearly that the inhibition of oxytocin effect on the rat uterus by IOT and MOT is not purely competitive. Over a broad range of inhibitor concentrations the pattern of the dose-response curves is very similar to that found for irreversible blockade of receptors in the case of so-called receptor reserve[17]. From a chemical point of view, it seems very unlikely that the substituent groups can provide a strong link to the receptors such as to cause an irreversible decrease in their number. On the other hand, however, the irreversibility of inhibitor binding is only a relative phenomenon, indicating that the 'active' substance is more readily released from the binding site that the inhibitor itself. In the series OT–MOT–IOT, such a difference in the disappearance rate from the receptor can be accounted for by increasing lipophilicity. It is instructive to mention in passing that prostaglandins were also found to inhibit the hydroosmotic effect of an oxytocin analogue, [Tyr(Me)^2]-oxytocin, showing the 'receptor-reserve'-pattern[18]. This, in our opinion, is a reflection of the great difference in lipophilicity between the 2 agents which determines their tightness of binding to the receptor and/or their effective concentration in the vicinity of the receptor. Such factors may also be operative in cellular or subcellular (e.g., membrane) preparations.

We are aware that any attempt to generalize would be pure guesswork. At any rate, however, the empirical evidence itself suggests that IOT is not very suitable for displacement experiments in uterus receptor studies and the interpretation of any such investigations in which iodotyrosine analogues are employed as markers deserves great caution.

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Potentiation by crude kallikrein of the myotropic effect of angiotensin I in the isolated rabbit aortic strip

Z. S. Ercan and R. K. Türker[1]

Department of Pharmacology, Faculty of Medicine, University of Ankara, Ankara (Turkey), 6 September 1976

Summary. Crude kallikrein (Padutin®), but not pure kallikrein, when preincubated with angiotensin I caused a potentiation of the myotropic effect of decapeptide on the isolated continuously superfused rabbit aortic strip. Addition of converting enzyme inhibitor, SQ 20881, to the medium inhibited this potentiation. The potentiation by crude kallikrein of the myotropic effect of angiotensin I is probably due to the conversion of decapeptide to octapeptide angiotensin II. This study indicates that Padutin is not a pure kallikrein preparation and probably contains a kininase fraction which causes the conversion of angiotensin I.

The conversion of angiotensin I (A I) to angiotensin II (A II) has been shown to be mediated by an enzyme present in many tissues especially in the lung and mesenteric circulation [2–4]. On the other hand, it has been shown that converting enzyme also causes the degradation of bradykinin and the synthetic inhibitor of this enzyme (nonapeptide, SQ 20881)[5] produces an inhibition in the conversion of A I but potentiates the effects of bradykinin[6]. The data presented in this paper indicate that crude kallikrein (Padutin®), but not pure kallikrein, when preincubated with A I caused a potentiation of the myotropic effect of A I on the continuously superfused rabbit aortic strip.

Material and method. The myotropic activity of A I was determined on the continuously superfused spirally cut rabbit aortic strips[7] as described previously[8]. This

Potentiation by crude kallikrein (Padutin®) of the myotropic effect of A I on the isolated continuously superfused rabbit aortic strips

Concentrations of angiotensins (ng/ml)		Control responses to angiotensins	Responses to A I incubated with Padutin (0.1 U/ml) Without SQ 20881	With SQ 20881 (50 ng/ml)
A I	5	0	23.0 ± 2.4	8.6 ± 0.9
	10	0	37.3 ± 2.8	16.0 ± 1.6
	20	0	66.1 ± 3.1	31.7 ± 1.1
	40	3.2 ± 1.0	85.4 ± 2.5	43.2 ± 2.1
	80	8.0 ± 1.1	–	65.5 ± 2.0
A II	5	28.7 ± 2.5	–	–
	10	40.0 ± 2.8	–	–
	20	76.6 ± 3.3	–	–
	40	96.6 ± 5.5	–	–

Per cent of maximum responses measured on the recorder (mean ± SEM of 10 experiments).

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preparation has been shown to be extremely sensitive to A II but not to A I[9]. A I and A II solutions were made from their stock solutions in 0.1 N acetic acid (100 µg/ml). 4 different concentrations of A I were prepared (1, 2, 4, 8 µg/ml) then crude kallikrein (Padutin®, Bayer), which was freshly diluted in saline, incubated with A I solution having a final concentration of 0.1 U/ml. These solutions were kept at 37 °C for a period of 30 min at pH 7.4. The same procedure was repeated in another series of experiments by adding SQ 20881 at the concentrations of 50 to 100 ng/ml to A I solutions before incubating with pure kallikrein. The same experimental design was followed with pure kallikrein (Kallikrein® KZC 1/75, Bayer AG). The dose-response curves of A I and A II alone, A I incubated with crude and pure kallikrein and A I incubated with crude and pure kallikrein containing SQ 20881 were determined in the aortic strips.

Results. The low concentrations of A II and relatively higher concentrations of A I (above 40 ng/ml) induced a dose-dependent contraction when superfused over the rabbit aortic strips. The contractile effect of A I was found to be potentiated after preincubation with crude kallikrein. No potentiation was observed after incubation of A I with pure kallikrein. Crude kallikrein itself neither potentiated nor relaxed when relatively higher concentrations were superfused over the rabbit aortic strips. Nonapeptide SQ 20881, which is a potent inhibitor of converting enzyme causes a significant inhibition in the response of aortic strips to A I preincubated with crude kallikrein. This inhibition was obtained when SQ 20881 was added to the medium before incubation of crude kallikrein. The degree of the inhibition by SQ 20881 was found to be almost the same when nonapeptide was used at the dose range of 10 to 100 ng/ml. Under the same experimental conditions, preincubation of SQ 20881 with A I alone did not influence the contractile response to decapeptide. The calculated results are shown in the table.

Discussion. The results of the present study clearly indicate that crude kallikrein, but not pure kallikrein, when incubated with A I causes a definite potentiation in the response of rabbit aorta. This potentiation is probably due to the conversion of A I to A II by crude kallikrein. This assumption has been based upon the following findings: 1. A I preincubated with crude kallikrein induces a myotropic activity which is equal to that obtained with A II on the aortic strip. No other fragments of angiotensin-peptides described having higher or equal myotropic activity when compared with the effect of parent peptide in the aortic strips[3]. 2. Converting enzyme inhibitor, nonapeptide SQ 20881, inhibits the potentiation of A I preincubated with crude kallikrein. In order to measure the quantity of the conversion of A I to A II by crude kallikrein, the dose-response curve of A II in each aortic strips was determined, and it indicated that almost 100% conversion occurred after preincubation of decapeptide with crude kallikrein. Since pure kallikrein did not cause a potentiation on the myotropic effect of A I, it is therefore obvious that this potentiation is not due to kallikrein itself in Padutin. It is highly possible that crude kallikrein preparations contains a kininase fraction which causes the conversion of A I to A II. The presence of a carboxypeptidase B in crude kallikrein preparations has been described previously[10].

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Neuronal accumulation and metabolism of ^{3}H-l-norepinephrine in rat portal vein: Evidence in relation to possible uneven alpha receptor distribution[1]

G. S. Takimoto, A. K. Cho and J. A. Bevan

Department of Pharmacology and Brain Research Institute, University of California, The Center for the Health Sciences, Los Angeles (California 90024, USA), 9 August 1976

Summary. The rate of accumulation and metabolism of ^{3}H-1-norepinephrine in the neuronal plexus of rat portal vein produces a small transmitter concentration gradient across the longitudinal smooth muscle layer which cannot account for the prejunctional supersensitivity observed and suggests localization of the alpha-adrenergic receptors adjacent to the nerve plexus.

The rat portal vein consists of 2 smooth muscle layers; an inner circular layer (10–20 µm thick) and an outer longitudinal layer (50–70 µm thick)[2]. A two dimensional adrenergic network is located between the smooth muscle layers[2,3]. Prejunctional hypersensitivity of the longitudinal muscle to norepinephrine (NE) applied exogenously has been demonstrated in vitro in chronically denervated and cocaine-treated tissues[2] amounting to a 13–30fold parallel shift of the dose-response curves. Because of the dimensions of the hypersensitivity in this vessel and the unlikelihood that the adrenergic nerve plexus could effectively reduce the concentration of NE throughout the longitudinal smooth muscle layer sufficiently to account for the shift in the dose-response curve, it has been postulated that the alpha-adrenergic receptors are located on the longitudinal smooth muscle cell layer directly adjacent to the nerve plexus[2,3,4,9].

In the present experiments, the rate of transport of ^{3}H-1-NE into the neuronal plexus was obtained from the neuronal accumulation and metabolism of ^{3}H-1-NE. It is self evident that NE must enter the tissue from its tissue bath at a rate equal to its rate of entry into the neuronal plexus at the steady state. From this data it was possible to calculate the necessary transmitter concentration gradient from the tissue bath to the perisynaptic region[5] to effect such a rate of entry. Since this calculation showed

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