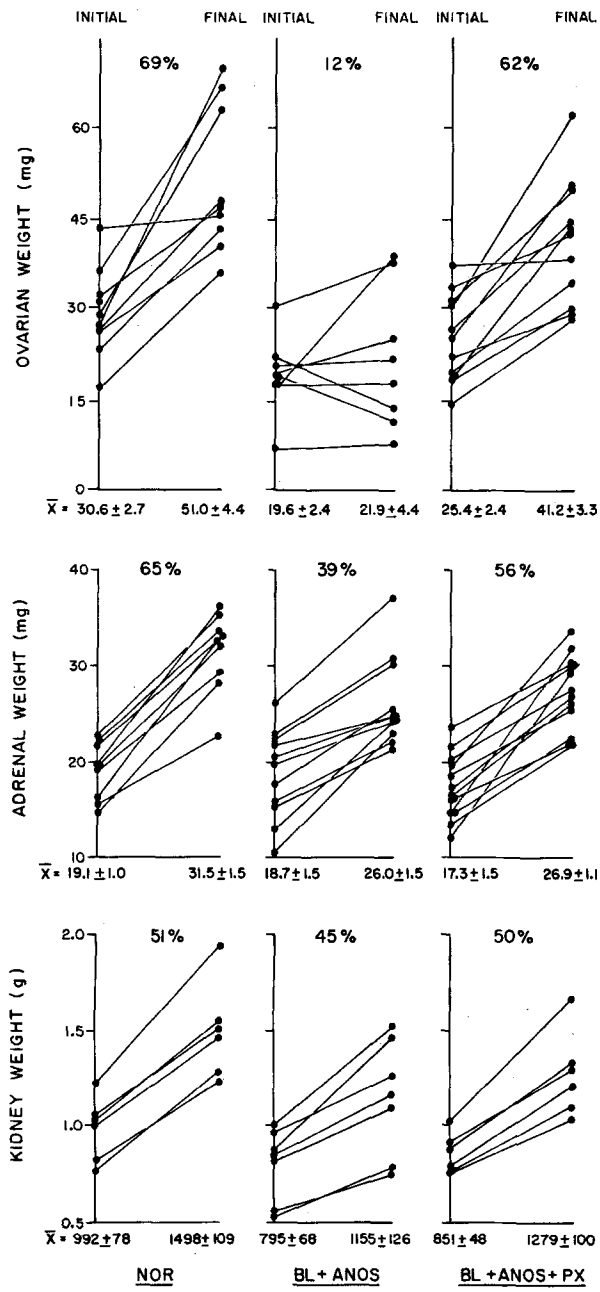
Compensatory Growth of the Ovaries, Adrenal Glands and Kidneys in Blinded, Anosmic Rats[1]

The combination of removal of the eyes and of the olfactory bulbs is seriously detrimental to the development of the reproductive systems in rats[2,3]. This impairment of genital development in blinded, anosmic animals is due to a hypofunctioning of the neuroendocrine axis. The following study was designed to determine whether another aspect of pituitary function, namely the synthesis and secretion of adrenocorticopin (ACTH), could also be affected by blinding and anosmia. Additionally, compensatory growth of a visceral organ, the kidney, whose primary growth does not depend on a single pituitary trophic hormone, was studied.



Compensatory growth of the ovaries (upper), the adrenal glands (middle) and kidneys (lower) in untreated (NOR) rats, in blinded anosmic (BL + ANOS) rats and in blinded anosmic rats that were also pinealectomized (BL + ANOS + PX). Lines between 2 points represent the growth of the respective organ in a single animal. Mean (± standard error) initial and final organ weights are given. Percent hypertrophy is given in the upper portion of each graph.

Materials and methods. 21-day-old male (52 animals) and female (27 animals) rats were either left intact or had their eyes and olfactory bulbs surgically removed; one half of the latter group were also pinealectomized. 3 weeks later each of the 3 groups of female rats were unilaterally ovariectomized. The weight of the removed ovary was recorded. At the same age, the male rats were unilaterally adrenalectomized or had one kidney removed. The weights of these organs were also recorded. All operations were done on animals anesthesized with ether. 2 weeks after the operations, when the animals were sacrificed and the single remaining organs, i.e., the ovary, the adrenal gland and the kidney, were retrieved and weighed. The percent hypertrophy was calculated. Data were statistically evaluated using an analysis of variance and a t-test between related means.

Results and discussion. Ovaries in normal rats exibited a 69% hypertrophy within 14 days (Figure, top). This enlargement was depressed ($p < 0.01$) to 12% in rats that were deprived of both their eyes and olfactory bulbs. In rats that were pinealectomized, however, combined blinding and anosmia were unable to restrain compensatory growth and in these animals the remaining ovaries enlarged an average of 62%. It is thus apparent that the restrictive influence which blinding and anosmia exert on compensatory ovarian hypertrophy is mediated by some substance deprived from the pineal gland. In that compensatory growth of the remaining ovary is known to involve the release of augmented amounts of follicle stimulating hormone (FSH) the present results indicate that the pineal restricts FSH synthesis or release[4].

As with the ovaries, compensatory growth of the adrenal glands was also retarded ($p < 0.05$) in blinded anosmic rats unless they were pinealectomized (Figure, middle). The plasma titer of ACTH rises after unilateral adrenalectomy and this hormone is believed to be responsible primarily for the accelerated growth of the adrenal gland, particularly of the adrenal cortex[5]. Since pineal removal allowed for normal compensatory enlargement of the remaining adrenal gland, it is concluded that pineal secretory products also inhibit ACTH release from the anterior pituitary gland.

Growth of the kidney is determined by a gamut of pituitary and extra-pituitary hormones[6]. During compensatory renal enlargement, however, there does not seem to be a single pituitary hormone that is secreted in excessive amounts. This relative independence from the pituitary gland during compensatory growth may explain the failure of blinding and anosmia to inhibit renal enlargement after removal of the contralateral organ (Figure, bottom). On the other hand, the initial kidney weights of the blinded anosmic rats were significantly (795 vs 992; $p < 0.05$) smaller than initial kidneys taken

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from normal rats. This finding is probably attributable to the lower levels of circulating testosterone in animals that can neither see nor smell[3]. Testosterone has an important stimulatory influence on normal renal growth[6].

Résumé. Chez les rats qui ont subi une ablation de la glande pinéale, la présence simultanée de perte de vue et d'anosmie empêchent la croissance compensatoire de l'ovaire chez la femelle et celle des glandes surrénales chez la mâle. Une hypertrophie du rein provoquée expérimentalement après une néphrectomie unilatérale n'est pas modifiée par la perte de vue et l'anosmie.

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Biological Properties of Synthetic Ser[4]-Arg[8]-Oxytocin (Ile[3]-Ser[4]-Arginine Vasopressin): Role of the Residue No. 4 in the Hormone-Pressor Receptor Interaction

The isolation of oxytocin and arginine vasopressin from the neurohypophysis of several mammals[1,2] has shown that these two nonapeptides, although structurally very similar (they differ only by 2 residues in positions 3 and 8) act on different receptors, since oxytocin is responsible for the uterotonic and milk-ejecting activities of the gland while vasopressin accounts for the pressor and antidiuretic activities. A hybrid molecule with the residue No. 3 of oxytocin (isoleucine) and the residue No. 8 of arginine vasopressin (arginine), called *arginine vasotocin*, has been synthesized and this peptide displayed on mammals the 4 activities mentioned above[3]. The role of residue No. 3 for the interaction with oxytocic receptor and that of residue No. 8 for the interaction with the pressor-antidiuretic receptor were thus disclosed.

More recently the identification of a new neurohypophysial hormone in bony fishes, *isotocin* (Ser[4]-Ile[8]-oxytocin) (ref.[4]) has shown that the substitution in position 4 does not decrease strongly the rat oxytocic activity of the nonapeptide, a result in agreement with some data previously obtained with synthetic 4-substituted analogues of oxytocin[5]. Furthermore, synthetic Thr[4]-oxytocin is about twice as active as natural oxytocin[6]. Except the residues 4 and 8 which can be substituted, the other amino acids of the molecule seem necessary for the oxytocic activity although the α amino group can be removed[7].

About the amino acids involved in the pressor activity, which is associated with the antidiuretic activity, it has long been known that a basic residue (arginine, lysine, ornithine, diaminobutyric acid) is necessary in position 8 (ref.[5]). Only a few analogues of vasopressin with substitutions in other positions have so far been synthesized, so that the relative importance of these positions, in particular that of position 4, is poorly known. Ser[4]-Arg[8]-oxytocin (or Ile[3]-Ser[4]-arginine vasopressin) has recently been synthesized[8] and a study of some of its biological properties has now been carried out.

From another point of view, because Ser[4]-Arg[8]-oxytocin has a serine residue in position 4 like hormones found in fishes (such as isotocin of teleosts[4] or glumitocin of rays[9]) and an arginine residue in position 8 like arginine vasotocin found in all non-mammalian vertebrates, this peptide might be a common evolutionary precursor of these hormones in a very primitive species. Isotocin and arginine vasotocin are simultaneously present not only in Neopterygii[4,10,11] but also in some Paleopterygii such as Polypterus[12]; on the other hand all the cartilaginous fishes have at least 2 neurohypophysial hormones[9,13]. If it is assumed that a gene duplication has occurred in early vertebrates for giving 2 similar peptides in fishes and higher classes, a single peptide might be found for instance in some species of the primitive class of Cyclostomata, and Ser[4]-Arg[8]-oxytocin might be a possible candidate for this single neurohypophysial hormone.

Table I. Amino acid composition of synthetic Ser[4]-Arg[8]-oxytocin (number of residues per mole, aspartic acid being taken as reference)

Amino acid	Theoretical values	Oxidized sample		Reduced sample	
		46 nmol	38 nmol	32 nmol	52 nmol
Asp	1	1.00	1.00	1.00	1.00
Ser	1	0.89	0.90	0.95	0.85
Pro	1	1.11	1.53	1.09	0.78
Gly	1	1.03	0.92	1.10	1.09
Ile	1	1.02	1.01	0.99	1.03
Tyr	1	0.68	0.42	1.04	0.92
Arg	1	1.00	0.89	0.95	0.97
Cys	2	2.40	2.11	—	0.67

	1	2	3	4	5	6	7	8	9
Isotocin	Cys	Tyr	Ile	Ser	Asn	Cys	Pro	Ile	Gly(NH_2)
Vasotocin	Cys	Tyr	Ile	Gln	Asn	Cys	Pro	Arg	Gly(NH_2)
Ser[4]-Arg[8]-oxytocin	Cys	Tyr	Ile	Ser	Asn	Cys	Pro	Arg	Gly(NH_2)

Amino acid sequences of isotocin, arginine vasotocin and Ser[4]-Arg[8]-oxytocin

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