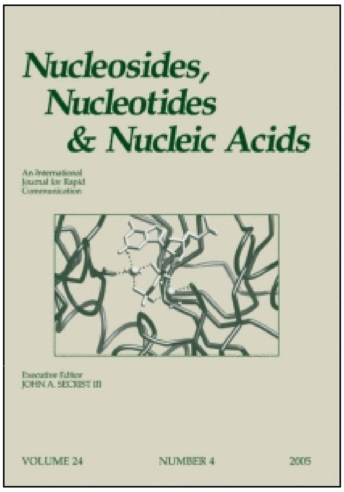

Nucleosides, Nucleotides and Nucleic Acids
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597286

In Situ Hybridization with Antisense Oligonucleotide: Estradiol Stimulates Transcription and Translation in Oxytocin Producing Hypothalamic Neurons

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To cite this Article Ramalho-ortigao, F. , Jirikowski, G. F. and Seliger, H.(1988) 'In Situ Hybridization with Antisense Oligonucleotide: Estradiol Stimulates Transcription and Translation in Oxytocin Producing Hypothalamic Neurons', Nucleosides, Nucleotides and Nucleic Acids, 7: 5, 769 — 771
To link to this Article: DOI: 10.1080/07328318808056327
URL: http://dx.doi.org/10.1080/07328318808056327

NUCLEOSIDES & NUCLEOTIDES, 7(5&6), 769-771 (1988)

IN SITU HYBRIDIZATION WITH ANTISENSE OLIGONUCLEOTIDE: ESTRADIOL STIMULATES TRANSCRIPTION AND TRANSLATION IN OXYTOCIN PRODUCING HYPOTHALAMIC NEURONS.

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Abstract: Oligonucleotide probes combined with immunocytochemistry was applied to study the transcriptional and secretory activity of oxytocinergic neuros.

The labor inducing and milk ejecting posterior lobe hormone oxytocin is produced in magnocellular hypothalamic neurons. Previous studies have shown that the number of oxytocin immunoreactive neurons in the rat hypothalamus and preoptic region is increased after treatment with physiological amounts of estradiol[1].

The aim of the present study was to determine whether estradiol enhances transcription in oxytocinergic hypothalamic neurons.

In situ hybridization with synthetic antisense oligonucleotide probes for specific m-RNA, combined with immunochemistry have been used in order to characterize the synthetic and secretory activity of these neuropeptide producing neurons. This technique has several advantages over commonly used in situ hybridization of histological specimens with genomic probes[2].

Methods

The synthesis and the structure of the oligonucleotide, as well as the hybridization protocol utilized have been described elsewhere [2].

Ovariectomized wistar rats were treated for two days with subcutaneous silastic implants providing plasma estradiol levels of 25 pg/ml. Control animals received blank implants. Serial vibratome sections of perfusion fixed brains of these animals were in situ hybridized with a (gamma ^{32}P) ATP labelled icosameric oligonucleotide probe inverse complementary to oxytocin m-RNA.

After hybridization, the sections were immunostained for oxytocin with the peroxidase-anti-peroxidase method and processed for autoradiography.

Results

In control animals immunostained and hybridized neurons were visible in the paraventricular and supraoptic nuclei and in the periventricular nucleus. While colocalization of in situ hybridization and immunostaining was apparent throughout these nuclei, several neurons with strong immunostaining remained unhybridized. Another population of cells showed intense radiolabelling and only weak immunostaining. This indicates that in ovariectomized animals, neuronal population of different levels of synthetic and secretory activity are present.

In estradiol treated animals oxytocin immunostained perikarya with intense radiolabelling appeared in the preoptic region, the perifornical region, the zona incerta and the lateral hypothalamus, in addition to the the neurons visible in control animals. The number of hybridized neurons in the supraoptic and paraventricular nuclei was increased as compared with the controls.

The combination of in situ hybridization and immunocytochemistry used in the present study provides a useful tool for studies of the synthetic and secretory activity of endocrine neurons.

Estradiol is known to stimulate the secretion of oxytocin into the peripheral circulation [3]. A subpopulation of hypothalamic oxytocin neurons has nuclear estrogen receptors [4]. Our results indicate that estradiol has the abillity of inducing oxytocin synthesis in hypothalamic sites outside the classical magnocellular nuclei, perhaps through a direct genomic effect. This activation has probably its physiological significance in the estrogen dependent stimulation of centrally projecting oxytocinergic system controlling sexual and maternal behavior[5], as well as in the stimulation of the hypothalamo neurohypophysial system[3].

REFERENCES

1. Jirikowski, G.F., Cadwell, J.D., Pedersen, C.A. and W.E. Stumpf (1988). Estradiol influences oxytocin-immunoreactive brain systems. Neuroscience (in press).
2. Jirikowski, G.F., Ramalho-Ortigao, F. and H. Seliger (1988). In situ hybridization with complementary synthetic oligonucleotide and immunocytochemistry: a combination of methods to study transcription and secretion of oxytocin by hypothalamic neurons. Molecular and Cellular Probes (in press).
3. Yamaguchi, K., Akaishi, T. and H. Negoro (1979). Effect of estrogen treatment on plasma oxytocin and vasopressin in ovariectomized rats. Endocrinol. J. 26: 197-205.
4. Jirikowski, G.F., Stumpf, W.E. and Ch. Pilgrim (1987). hypothalamic estradiol target neurons immunoreactive for neuropeptides. Verh.Anat.Ges. 81: 921-923.
5. Pedersen, C.A., Asher, J.A., Monroe, Y.L. and A.J. Prange jr. (1985). Oxytocin induces maternal behavior in virgin female rats. Science 216: 648-649.