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Early embryonic administration of xenoestrogens alters vasotocin system and male sexual behavior of the Japanese quail

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Abstract

The copulatory behavior and the parvocellular vasotocin (VT) system of the nucleus of the stria terminalis (BST) are sexually dimorphic in the Japanese quail. Embryonic administration of estradiol benzoate (EB) induces an organizational effect determining the disappearance of such a dimorphism (male shows behavior and cerebral phenotype of the female). The VT parvocellular system can therefore be considered an accurate marker of the sexual differentiation of brain circuits and a very sensitive indicator of the activity of estrogen-like substances on neural circuits. To test this hypothesis we administered diethylstilbestrol (DES), a powerful synthetic xenoestrogen, genistein (GEN), a phytoestrogen produced by soy, and bisphenol A (BPA). After 3 days of incubation, quail eggs were injected with vehicle, EB, DES, GEN or BPA. Administration of BPA caused an early blockage of development and no further analyses were done on the BPA groups. At puberty, the copulatory behavior of EB- or DES-treated male quail was totally abolished, whereas only the highest doses of GEN determined a significant decrease of the behavior. After the tests, the animals were sacrificed and perfused. The fractional area (FA) covered by VT immunoreactivity was analyzed in BST, medial preoptic nucleus, and lateral septum by computerized image analysis. The FA was significantly reduced after treatment with EB, DES and GEN at high doses. These results confirm that the sexually dimorphic VT system

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of the Japanese quail is a sensible indicator of the effects of xenoestrogens at the level of the central nervous system.

Keywords: Limbic system; Hypothalamus; Diethylstilbestrol; Genistein; Bisphenol A

1. Introduction

In quail and chicken, vasotocin (VT) neurons of the bed nucleus of the stria terminalis, pars medialis (BSTm)—medial preoptic nucleus (POM) system (Fig. 1F–G) represent a population of brain cells with a clearly identifiable neurochemical phenotype, undergoing a profound and irreversible sexual differentiation during ontogenetic development (for a review see [1]). These parvocellular VT-immunoreactive (-ir) elements have similar characteristics in all avian species investigated so far. The extension of their dendritic arborization is simpler (generally bipolar), and their immunostaining is weaker than in the magnocellular population (Fig. 1D–E). A population of parvocellular VP-ir neurons has similarly been clearly identified in the BST and in the medial amygdala of mammals [2]. Thin VT-ir fibers have been observed in several brain regions outside of the hypothalamo-neurohypophyseal tract also in different oscsine avian species (canary [3], zebra finch [4], junco [5]). VT-ir fiber endings were observed in the telencephalon (lateral septum, BSTm), in the POM, in broad areas of the diencephalon, the mesencephalon, the pons, and the medulla (see [6] for a complete list). The origins of all these thin VT-ir fibers have not been fully identified, but results collected in our laboratory demonstrated that in male quail the BSTm and POM project to the lateral septum [7] (Fig. 1A–C), and that the VT neurons of the BSTm project to POM [8].

2. The sexually dimorphic parvocellular vasotocin system

A few studies have revealed a robust sexual dimorphism of the parvocellular VT-ir cell groups in the avian BSTm (for a review see [9]). In particular, in galliforms, VT-ir neurons are present in the BSTm of males only and cannot be visualized in females [10,11]. In male quail, the VT-ir population of BSTm merges cranially and ventrally with a population of scattered neurons observed within the boundaries of the POM, a sexually dimorphic region controlling male reproductive behavior [12]. VT-ir fibers also show a sexually dimorphic distribution in several regions of the quail brain. In particular, the two regions that are connected with the BSTm, the lateral septum and the POM of males contains a denser VT-ir innervation than in the females (see [6] for a review).

VT-ir fibers are distributed in large portions of the diencephalon and brain stem. In the telencephalon, they are present only within the limbic system. In many places [e.g. the lateral septum, the POM, the ventromedial nucleus (VMN), the substantia grisea centralis (GCt), the intercollicular nucleus (ICo), and the locus coeruleus (LoC)], these fibers are present in higher density in males than in females. All these regions are directly or indirectly related to the control of reproduction. Therefore, these anatomical data support the notion