5-Hydroxyindoles compounds and nitric oxide voltammetric detection in the rat brain: changes occurring throughout the sleep-wake cycle

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Summary. The release of serotonin may occur throughout the sleep-wake cycle according to 2 different modalities: – by the axonal nerve endings during waking; – by the dendrites and/or the soma of the nucleus raphe dorsalis (nRD) during sleep. Neuronal nitric oxide (NO), synthesised by constitutive NO synthase (NOS), is colocalized with neurotransmitters such as GABA, acetylcholine, somatostatine, serotonin, etc. In order to evaluate its modalities of release throughout the rat sleep-wake cycle, a sensor allowing its specific detection in freely moving animals was prepared. In the cortex, the highest NO signal occurs during the waking state (W = 100%) versus slow wave sleep (SWS = −6%) and paradoxical sleep (PS = −9%). The mild variations observed might reflect a mean of the individual sleep-wake cycle variations attached to each NO source (GABAergic interneurons, cholinergic and serotoninergic axonal nerve endings, etc.).

Keywords: Serotonin, nitric oxide, voltammetry, sleep, wake.

Introduction

The involvement of serotonin (5-HT) in sleep has been suggested for more than 20 years (Jouvet et al., 1969). Since then, the hypothesis concerning the modalities through which this transmitter might influence sleep has evolved owing to continuously improved technical approaches (McGynty and Harper, 1976; Cespuglio et al., 1981b, 1992, 1994). Recently, nitric oxide (NO), a diatomic low molecular weight compound first considered as a toxic gas, has been reported to be present within neuronal elements (Snyder and Bredt, 1992; Bredt and Snyder, 1994). Contrary to serotoninergic neurons which innervate the entire central nervous system from cell bodies located in the limited area of the raphe system, NO is synthesised within several neuronal sets widespread throughout the entire brain (Vincent and Kimura,
1992; Vincent, 1995; Rodrigo et al., 1994). These sets of neurons co-synthesize different well-known neurotransmitters like GABA, acetylcholine, somatostatine, NPY. As regards monoaminergic neurons, those expressing tyrosine hydroxylase do not contain NOS except a limited neuronal population located in the periaqueductal grey area and the rostro-ventral tegmentum (Johnson and Ma, 1993). A significant proportion of the serotonergic neurons located in the rostral raphe expresses, however, a NOS activity (Wotherspoon et al., 1994; Wang et al., 1995).

According to the well-known involvement of 5-HT in sleep triggering and maintenance (Jouvet, 1969; Petitjean et al., 1981; Cespuglio et al., 1990), if the co-storage 5-HT-NO underlies a functional reality, it appears likely that NO might play a part in the sleep-wake regulatory processes. In this report, the current knowledge about the involvement of 5-HT and NO in sleep processes is considered.

**Serotonin and sleep**

*The paradoxical role of serotonin*

Historically, 2 main contributions suggested first that 5-HT released at postsynaptic targets might act as a hypnogenic neurotransmitter: – the destruction of the raphe system of the cat, containing 80% of serotonergic perikarya, is followed by a total insomnia lasting several days; such an insomnia ends in a progressive recovery of normal sleep amounts without any sleep rebound (Jouvet, 1969); – the inhibition of 5-HT synthesis by p.chlorophenylalanine (PCPA) also produces in the cat a secondary total insomnia together with the occurrence of ponto-geniculo-occipital (PGO) waves; progressive restoration of slow-wave-sleep (SWS) and paradoxical sleep (PS) also occurs without any sleep rebound in this situation; during the period of total insomnia, systemic administration of 5-hydroxytryptophan (5-HTP), the precursor of 5-HT, immediately suppresses the spontaneous occurrence of PGO waves (1–2 min.) and restores, within 30 to 60 minutes, a normal alternation of SWS and PS for 6 to 8h (Jouvet, 1969; Petitjean et al., 1981). These observations at the basis of the serotonergic sleep theory have been, however, exposed to some criticisms: – moderate and localised cooling (+10°C) of the nucleus raphe dorsalis (nRD) with adequate thermodes are sleep-inducing (Cespuglio et al., 1976, 1979); – unitary studies performed in the raphe nuclei indicate that serotonergic neurons are active during waking and that their neuronal discharge decreases continuously during SWS and PS (McGinty and Harper, 1976; Cespuglio et al., 1981a; Jacobs and Fornal, 1991). These facts, while based only upon neurophysiological evidence reveal a manifest inconsistency with the PCPA-5-HTP paradigm postulating a need of 5-HT for sleep as well as a possible direct role for it in this state. On the basis of the above experimental data the nature of the part played by 5-HT in sleep thus appears paradoxical.

*Modalities of serotonin release throughout the sleep-wake cycle*

In order to clarify the above contradictory aspects, methodologies attempting to measure 5-HT release were developed, i.e. the “push-pull” canula (Puizill-