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Research report

Monocular sleep following passive avoidance learning in chicks

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Abstract

Monocular sleep following passive avoidance learning was investigated in young chicks. One group of animals (Experimental Group) was presented with a bead coated with an aversive substance; a second group (Control Group 1) underwent bead presentations as in the Experimental Group but with the bead coated with water rather than with the aversive substance, and a third group (Control Group 2) did not undergo to any bead presentation. Binocular and monocular sleep was recorded during the 8 h subsequent the learning (or the control) event. The main results were that (1) the percentage of time spent in binocular sleep and the percentage of episodes of binocular sleep were lower in the Experimental Group that in the Control Groups; (2) the times spent in monocular sleep was higher in the Experimental Group and progressively lower in Control Group 1 and Control Group 2; (3) chicks slept predominantly with their left eye closed, but there was no evidence of specific changes in the opening of their right or left eyes associated with the learning event. The results seem to be compatible with the hypothesis that monocular sleep serves to provide the animal with the possibility to monitor periodically the environment to look for important events (e.g. predators, presence of the mother hen and siblings) and any arousal-producing event that would occur before a sleep episode tends to increase the likelihood of monocular sleeping.

Keywords: Sleep; Unihemispheric sleep; Domestic chick; Learning; Passive avoidance

1. Introduction

Together with aquatic mammals and other bird species (review in [18]), domestic chicks exhibit a unique behavioural and electrophysiological state dubbed monocular or unihemispheric sleep [2,11,15,20,26]. During normal sleep, chicks show brief periods of time in which one eye is opened and the contralateral hemisphere shows a EEG pattern typical of wakefulness (fast and low voltage waves), whereas the other eye remains closed and an EEG pattern of slow wave sleep (slow and high voltage waves) can be recorded in the contralateral hemisphere [2]. Chicks showed also binocular or bihemispheric sleep during which closure of both eyes is associated with slow wave sleep (SWS) or rapid eye-movement (REM) sleep in both hemispheres.

Few studies have looked at the function of unihemispheric sleep in birds. During unihemispheric sleep birds keep one eye open allowing them to monitor the environment for potential predators whilst at the same time gaining the benefits of sleep. This predator detection hypothesis was tested by Rattenborg et al. [16,17] by manipulating the level of risk perceived by Mallard ducks (*Anas platyrhynchos*). Individuals at the edge of a group (a position which animals perceive as dangerous) showed a 150% increase in unihemispheric sleep compared to those in the center. Moreover, animals showed a strong preference for directing the open eye away from the center of the group to the surrounding environment from where a threat would be most likely to appear [16,17].

Some recent findings suggest, however, that the anti-predatory function of unihemispheric sleep could be not the only explanation of the phenomenon or that, at least, the hypothesis has to be modify to account for these novel findings. Although in principle opening of the left or right eye would be indifferent for the anti-predatory hypothesis (being the actual location of a source of danger more important than the particular eye to be opened), several evidence have been collected that in the domestic chick lateralization at the population level can be observed in the direction of eye opening.

Mascetti et al. [11] reported that during the first week post-hatching, female chicks reared with an imprinting object showed

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a pattern of monocular sleep with either right or left eye closure occurring with approximately the same frequency, except on day 5 in which right eye closure dominated. During the second week, there was a clear bias towards more monocular sleep with left eye closure [11]. By contrast, monocular sleep in female chicks reared without any imprinting object showed a predominant left-eye closure pattern in both weeks except on day 8 in which monocular sleep was recorded equally from each eye. Comparing the two rearing conditions, it was apparent that during the first week imprinted chicks showed more right eye closure compared to chicks reared without any imprinting object; this effect was tentatively associated with consolidation of imprinting memories in the left hemisphere (see [1]). During the second week, both groups of female chicks showed a preference for left eye closure. A removal of the imprinting object or a change in the colour of the imprinting object on day 8 caused a striking shift towards a predominant right eye closure during monocular sleep [11]. This was tentatively associated with right hemisphere involvement in response to novelty (see e.g. [27,29]).

Evidence for lateralization in the pattern of monocular sleep is not, in principle, directly opposed to a vigilance function for unihemispheric sleep. It is likely that birds use monocular sleep to allow the monitoring of their surroundings not only for predators but also for other pertinent events (e.g. position of conspecifics, food availability, changes in the weather, etc.). However, it is likely that predator detection would be the most important function given the potential cost of not detecting an approaching predator. In some circumstances, however, other visual stimuli could be very important as well, an example being the need for young domestic chicks to monitor the presence of the imprinted hen and siblings [11]. Nonetheless, the existence of such a precise relationship between opening of one particular eye and the associated functions of the contralateral hemisphere makes one to suspect that unihemispheric sleep may serve even other functions, for instance that it could be involved in memory consolidation.

Several studies provide support to the hypothesis that sleep would play a crucial role in brain plasticity and particularly in the processes of memory organization and consolidation (i.e. [3,8–10,24,31]). The domestic chick has been one of the preferred experimental models for the neuropharmacological studies on memory consolidation processes (see [22]). It has been reported that after imprinting there is an increase of REM sleep either in the total amount or in the number of events [25]. Furthermore, it has been proposed that sleep is not exclusively a process involving the entire brain but it may show also regional aspects dependent on the activation of specific brain regions [5,7,12,14,30]. The domestic chick is an interesting experimental model in this regard, because it makes available to direct investigation changes in the pattern of monocular-unihemispheric sleep following learning tasks which are known to be lateralized in this species.

Here we investigated one such tasks, i.e. the passive avoidance learning task. The task takes advantage of the tendency of newly hatched chicks to peck at small visually conspicuous objects. When a coloured bead is presented, chicks repeatedly peck at it, even in the absence of any obvious reinforcement. When the bead is coated in a strong bitter substance, chicks peck at once, but soon exhibit a characteristic disgust response and subsequently avoid pecking again beads of similar appearance [4]. The underlying neuronal mechanism for the formation of passive avoidance memory have been extensively investigated (reviews in [22,13], as well as cerebral lateralization effects that have revealed a prominent involvement of structures in the left hemisphere (reviews in [1,19,21,27]). Here we investigated the pattern of binocular and monocular sleep during the first 8 h following passive avoidance learning.

2. Materials and methods

2.1. Subjects

The subjects were 106 females Hybro (a local hybrid variety derived from the White Leghorn breed) domestic chicks (*Gallus gallus*). Subjects were divided in two groups tested in different days (Day 2 and Day 3 of life). Day 2: Experimental Group ($n = 16$), Control Group 1 ($n = 16$), Control Group 2 ($n = 15$); Day 3: Experimental Group ($n = 22$), Control Group 1 ($n = 18$); Control Group 2 ($n = 19$).

2.2. Apparatus

The apparatus used for observation of sleep consisted in two glass home-cages (40 cm × 40 cm × 30 cm) with semi-transparent cloths along the wall that served as one-way screens. Each cage was illuminated continuously from above with a 60 W electric-light bulb and contained two identical small transparent glass containers (5 cm × 5.5 cm), one for food and the other for water, available ad libitum throughout the overall period of observations. A red plastic cylindrical ball (4 cm × 3 cm) was suspended freely in the middle of the cage at about head height for the chick, providing an artificial social companion (see [28]).

For the PAL procedure chicks were tested in their home-cages (below) that contained two identical small transparent glasses, one for food and the other for water, and the imprinting object, the same as described above.

2.3. Procedure

Chicks arrived in the laboratory from a commercial hatchery (“Agricola Berica” di Montegalda, Vicenza) in the early morning of day 1 after hatching and were immediately placed singly into metal cages (28 cm × 31 cm × 40 cm) with food and water ad libitum as described above. In order to reduce isolation stress, each chick was kept in its home cage with an imprinting object (a red plastic cylindrical ball, 4 cm × 3 cm). According to the condition, on their second or third day of life (see [4,21]), chicks underwent to passive avoidance learning.

It has been reported that even pecking at a water-coated bead is itself an appetitive experience and has brain sequelae that differ from those showed by control animals held in equivalent conditions but without any pecking bead experience [6,23]. We decided, then, to use two Control Groups, labelled respectively as “*Control Group 1*”, in which chicks underwent bead presentations as in the Experimental Group but with the bead coated with water rather than with the aversive substance as in the Experimental Group (see below), and “*Control Group 2*”, in which subjects did not undergo to any bead presentation, but only to sleep observations with the same timing as the Experimental and Control 1 chicks.

The experimental design comprised four phases:

2.3.1. PAL pre-training

A small dry red bead was presented to the chicks for three times, spaced 5 min apart. Chicks that did not peck at the bead for at least two of the three trials were discarded.

2.3.2. PAL training

During training a bead coated with water was presented to chicks of the Control Groups, whereas a bead coated with an aversive substance, Methylanthranilate (MeA), was presented to chicks of the Experimental Group. Following