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**Pretecto-tectal influences**

**I. What the toad's pretectum tells its tectum:**
an antidromic stimulation/recording study

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**Abstract** Optic tectum and pretectum are two main structures for parallel processing of contralateral retinal inputs. Since it is known from anatomic studies that pretectum projects to ipsilateral tectum, the present investigation focuses on the physiological properties of this information transfer. (1) Extracellular single cell recordings from pretectal thalamic nuclei in cane toads *Bufo marinus* revealed different classes of neurons, TH1 to TH10, comparable to the ones described previously in *B. americanus*, except TH9. A further class TH11 responded specifically to the temporonasal direction of movement of a large object or textured surface. (2) Most neurons belonged to the classes TH3 and TH4, sensitive particularly to large moving objects and responsive to moving textured surfaces independent of the horizontal direction of movement. Nearly one third could be antidromically activated to electrical stimulation of the ipsilateral optic tectum, by the criterion of the collision test, that is ultimate proof of their pretectotectal projective character. (3) Among the remaining neurons, one tonically discharging TH1 cell fulfilled the collision test. (4) It is suggested that TH3 cells inform topographically corresponding structures of the tectal visual map about large moving objects and their extension perpendicular to the direction of movement, hence contributing to the discrimination between prey and non-prey. Class TH4 pretectotectal projection cells with inputs from the entire monocular or binocular visual field inform the tectum about large objects moving anywhere in the visual field and about a moving textured surrounding that fills the visual field. The latter would allow the tectum to discriminate object motion from self-induced motion.

**Key words** Pretectum · Neuronal classes

**Abbreviations** *A* stripe in antiworm configuration, oriented perpendicular to the direction of movement

*W* stripe in worm configuration, oriented parallel to the direction of movement

*S* square *C_E, C_T* empirical and theoretical collision interval, respectively

*ERF* excitatory receptive field

*IRF* inhibitory receptive field

*L, R* neuronal response latency and absolute refractory period, respectively

*Lpd, Lpv* lateral posterodorsal, lateral posteroventral, and posterior pretectal thalamic nucleus, respectively

*NT, TN* nasotemporal or temporonasal direction of stimulus movement in the visual field of the eye

*OT* optic tectum · *TH* classes of pretectal thalamic neurons

**Introduction**

The question "What the frog's eye tells the frog's brain" (Lettvin et al. 1959) promoted the investigation how retinal input is processed in the amphibian brain for visually guided behaviors. The release of prey-catching, for example, involves a retinotectal network (Grüsser and Grüsser-Cornehls 1973; Ingle 1983a,b; Ewert 1987; An der Heiden and Roth 1989; Arbib 1989) that has access to the bulbar/spinal motor coordinating systems (Satou et al. 1985; Ewert et al. 1994). This processing structure is necessary, but not sufficient for adequate behavior. Further influences are required, e.g. involving pretectum (visual tuning), hypothalamus (behavioral motivating), and striatum (response gating) (Ewert 1992). The present investigation focuses on ipsilateral pretectotectal information transfer which originally was suggested from the disinhibition of prey-catching in toads following caudal dorsal thalamic lesions (Ewert 1968, 1984).

It is known that the anuran retina projects topographically both to contralateral pretectal and tectal...
jections (Wilczynski and Northcutt 1977; Neary and Northcutt 1977; Neary and Northcutt 1983; Merchenthaler et al. 1989; Kozicz and Lázár 1994) are involved in the decision to avoid or to approach a visual target (Ewert et al. 1983; Ingle 1983). However, there was as yet no proof about the physiological properties of pretectotectal projecting cells. The present study in cane toads *Bufo marinus* investigates this question by means of the antidromic stimulation/recording technique. Part II concerns modes of pretectotectal interaction (Schwippert et al. 1995).

### Material and methods

#### Experimental animals

Fifty-nine *Bufo marinus* (L.) were kept in aqua-terraria under standard laboratory conditions at a room temperature of about 24°C. Since European amphibians are protected by federal laws, the experiments initially started with *Bufo bufo spinosus* were conducted with the South American species *B. marinus* which is not protected regarding its distribution and massive propagation.

#### Presentation of visual stimuli

Different configural moving or stationary visual stimuli of black card bord against a white background (contrast C = 0.95) were presented to the toad’s eye centered in a perimetric apparatus (Ewert 1984). Stimuli were moved in horizontal direction at a visual angular velocity of 8°/s at a distance to toad’s eye of 25 cm. The following standard stimuli were used: a 2°×16° rectangular stripe, whose longer axis was oriented either parallel or perpendicular to the direction of movement, called W- [worm]- or A- [antworm]-configuration, respectively; a square of 4°×4° (S4) or 16°×16° (S16) in size, S-configuration; a Julesz texture, 60°×60° in size, consisting of rectangular black and white patches in random distribution (pixel size 0.69°). The onset and the offset of the movement of a stimulus was registered by photo cells.

#### Preparation

For surgery, toads were anesthetized with Ketavet® at a dose of 100 mg/kg body weight. The dorsal brain surface was exposed in the region of the diencephalon and the mesencephalon. For oxygen supply, the toad’s body skin was kept moist with a thin wet tissue. During the recording studies, the animal was lightly paralyzed after intralymphatic injection of alpha-tubocurarine at a dose of 4 mg/kg.

#### Electrophysiology

Standard electrical stimulation and recording techniques were used. Recording electrodes were stainless steel needles, electrolytically sharpened to a tip of 2-3 μm diameter and insulated – except for a free tip – by means of Insl-x lacquer. Electrodes were tested according to their galvanic properties with a method developed by Dr. Evelyn Schürg-Pfeiffer (see Schürg-Pfeiffer et al. 1993). The recording electrode was advanced with the micro-manipulator toward the caudal thalamic structures at an oblique angle in order to save the dorso medial optic tract. Action potentials were fed to an oscilloscope (Tektronix D13) and stored on VHS video tape after PCM modulation. Data were processed on a 486 PC by means of a CED 1401-plus interface with the CED-internal programmable signal analyzing software Spike 2.

To determine whether a recorded pretectal thalamic neuron projects its axon toward the ipsilateral optic tectum, the antidromic electrostimulation/recording technique was applied. After classification an isolated pretectal neuron (criteria listed below), its action potential triggered – via a square wave generator (HSE-T stimulator with isolation unit) – an impulse of 0.1 ms duration and about 15 μA. This electric stimulus e was applied by means of bipolar electrodes to the ipsilateral tectum in which the axon of this pretectal neuron was suggested. The tip separation of the stimulation electrodes, oriented parallel to the sagittal brain axis, was about 500 μm. Since the visual ERFs of the pretectal neurons under investigation in this study were mostly located in (or encompassed) the frontal visual field, the tectal stimulation electrode was positioned in the corresponding tectal visual map.

#### Criteria of antidromic activation

Antidromic activation criteria were: constant latency response, following ability to repetitive electrostimulation, and collision test (Lipski 1981). In the collision test, an orthodromic traveling spike *s*0 (recorded from a pretectal neuron, e.g., visually elicited) triggered the electrical stimulus e that evoked an antidromic traveling spike *s*a. At a critical delay between *s*0 and e, both spikes collided and extinguished each other, provided both spikes were traveling along the same axon. Theoretically, the delay *Ct* between *s*0 and e, at which the antidromic spike *s*a disappears on the oscilloscope screen, is *Ct*= latency [L] + refractory period [R]. After Fuller and Schlag (1976), the collision criterion for the empirical delay *Ct* is still fulfilled, if (Ct – Ct) Ct< 0.5.

In an elaboration of this experimental paradigm, *s*0 triggered two sequential 0.1-ms square wave impulses e-e' separated by an interval >2R. The delay between *s*0 and the first electrical impulse e was variable. In the collision case, the antidromic spike *s*a was abolished to the first impulse e, but – serving as control – not to the second impulse e’. The positive collision test is ultimate proof of the antidromic activation of a cell and its projective character; however, the negative outcome of this test does not necessarily exclude the possibility of antidromic activation (Fuller and Schlag 1976).

#### Criteria for the classification of pretectal thalamic neurons

The response properties were investigated according to the criteria applied to pretectal thalamic TH-type neurons of *B. americanus* (Ewert 1971): (i) size, shape, and location of a neuron’s visual excitatory receptive field (ERF), measured with a 8° black disc moving manually by means of a transparent stick against a white background; (ii) mon- or binocularity, investigated in questional cases by covering one eye with a tiny thin opaque paper bag; (iii) on/off responses to rapid change of diffuse illumination of the visual field tested by turning the room lights off and on; (iv) responses to moving configural objects, investigated with the described W-, A-, and S- stimuli for movements (v=8°/s) both in nasotemporal and temporonal directions; (v) disc-shaped object, moving manually in the z-axis toward, or away from, the toad’s eye; (vi) responses to a stationary object, tested with the S16-stimulus; (vii) object moving continuously or jerkily in order to investigate influences of movement dynamics; (viii) object traversing ERF sections repetitively in...