

Short Communications

Direct ipsilateral retinal projections in goldfish (*Carassius auratus*)

ALAN D. SPRINGER* and GARY E. LANDRETH

Neuroscience Laboratory, University of Michigan, Ann Arbor, Mich. 48109 (U.S.A.)

(Accepted December 30th, 1976)

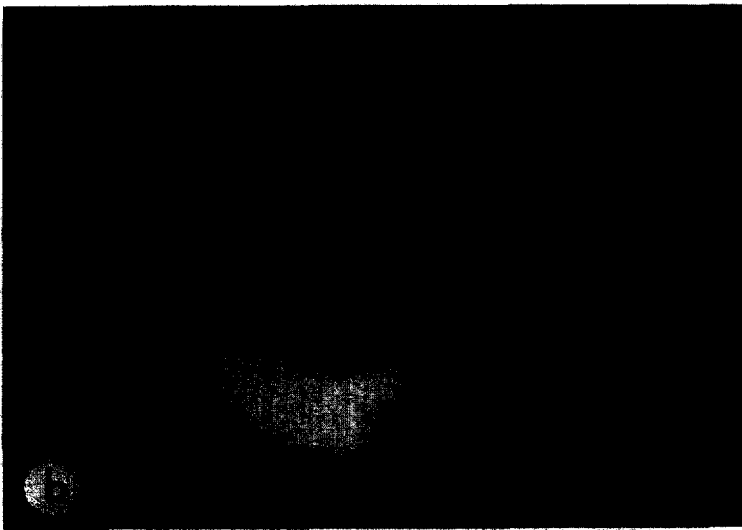
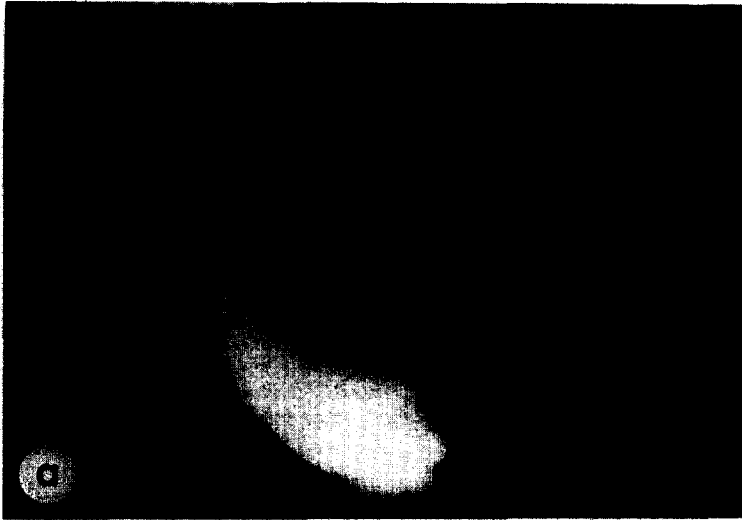
The goldfish visual system has been extensively used in biochemical, electrophysiological, anatomical and behavioral studies^{3,7,10,11,18,19}, as well as in those concerned with neuronal plasticity and regeneration^{4,6}. Conventional silver-degeneration methods^{10,18,19} and radioautography^{8,12} have been used to map retinofugal projections in goldfish. The optic nerves are described as completely decussating at the chiasm and projecting to the contralateral optic tectum, as well as to the diencephalon and preoptic area^{10,18,19}. In addition to the known contralateral projections, we have found direct retinal projections to the ipsilateral diencephalon and preoptic area using [³H]proline radioautography.

Common goldfish (*Carassius auratus*), weighing 8–11 g, and 6–7 cm in body length, were obtained from Ozark Fisheries, Stoutland, Mo.; the fish were maintained at 20 ± 1 °C. Six fish were injected intraocularly with 25 μ Ci of L-[4,5-³H]proline (46 Ci/mmol; New England Nuclear) in 5 μ l of saline. Fish were sacrificed either 24 h or 35 days after injection. Following fixation, the brains were processed for radioautography^{8,12}. Brain sections were coated with Kodak NTB2 emulsion, exposed for 34 days, developed and stained with hematoxylin–eosin.

Silver grains were heavily concentrated over the contralateral optic tract, optic tectum and retinal targets of the diencephalon and preoptic area. Retinofugal fibers were also found to terminate in several ipsilateral regions by three routes: an uncrossed pathway via the ipsilateral optic tract, fibers that decussate at the chiasm but subsequently recross the midline through the posterior commissure and fibers which recross through the minor commissure. A direct projection to the ipsilateral optic tectum was not seen.

In all specimens examined, decussation of the optic nerve at the chiasm was incomplete. A small number of fibers are distributed throughout the ipsilateral optic tract near the chiasm. As the tract courses dorsally, the fibers collect and follow its

* Present address: Department of Physiology, University of Illinois Medical Center, Chicago, Ill. 60680, U.S.A.



medial boundary (Fig. 1a). Two separate fascicles of these fibers can be distinguished as they exit the ipsilateral optic tract and distribute to diencephalic targets. The rostral fascicle courses dorsomedially and terminates in the region of the dorsal thalamic nuclei, nucleus dorsolateralis and nucleus dorsomedialis, and area prepectalis (Fig. 1b). Its course and region of termination are comparable to that of the contralateral dorsomedial fascicle of the optic tract¹⁹. The caudal fascicle passes through the diencephalon and terminates in the prepectal nucleus¹⁶ (Fig. 1b).

In addition to those fibers in the ipsilateral optic tract, other retinal fibers also enter the ipsilateral side of the brain. These fibers decussate at the chiasm and then recross at the midline at two sites. The first group of recrossing fibers originates from the dorsomedial fascicle of the contralateral optic tract. It distributes to the contralateral dorsal thalamic nuclei and to adjacent nuclear groups at the level of the posterior commissure. A number of fibers continue in the posterior commissure to terminate in the corresponding area ipsilaterally (Fig. 1b). The ipsilateral dorsal thalamic nuclei and area prepectalis appear to receive fibers from the ipsilateral optic tract and from the contralateral dorsomedial optic fascicles that course over the posterior commissure. However, since both tracts project into the area, we could not determine whether either tract projects exclusively to a given nucleus or whether the projections overlap.

A second group of recrossing fibers follow the medial optic fascicle as it exits from the contralateral optic tract. Fibers in this fascicle distribute to the contralateral nucleus preopticus pars magnocellularis¹⁹. Fibers also pass through the minor commissure to terminate in the same nucleus ipsilaterally (Fig. 1c). Apparently, a small number of retinal fibers comprise the ipsilateral projections since fewer grains were located over ipsilateral as compared to symmetric contralateral retinal targets. This difference in degree of labeling is consistent with observations in the gar¹⁵.

It is possible that the labeling observed in the ipsilateral diencephalon does not represent primary afferent terminals but may be the result of either non-specific incorporation of blood-borne precursor or transneuronal transport of precursor. Systemic labeling with [³H]proline could not account for the localized ipsilateral grain densities. It is difficult to rule out the possibility that our results can be accounted for by transneuronal movement of labeled precursor from primary, contralateral terminal fields to adjacent second order cells, which then project ipsilaterally⁵. This alternative also seems unlikely since the ipsilateral terminal fields were evident in radioautographs of fish killed 24 h after [³H]proline injection. We would not expect a significant amount of radioactivity to appear in second order terminal fields over this in-

Fig. 1. Coronal section of a normal goldfish brain at three different levels: (a) caudal chiasm, (b) rostral tectum and (c) mid-tectum. The fish was injected with [³H]proline into one eye and was allowed to survive for 5 weeks. Microphotographs were taken with both transmitted and incident light to illustrate both the tissue section and emulsion grains. Scale, 1 mm. COT, contralateral optic tectum; IOT, ipsilateral optic tectum; COTr, contralateral optic tract; IOTr, ipsilateral optic tract; OC, optic chiasm; PC, posterior commissure; AP, area prepectalis; NP, nucleus prepectalis; NDL, nucleus dorsolateralis; NDM, nucleus dorsomedialis; MC, minor commissure; NPM, nucleus preopticus pars magnocellularis.

terval. In a related study using frogs, labeling was not found in second order targets until 11 days after intraocular injection of 60–80 μCi of [^3H]proline³. In addition, if the source of the ipsilateral labeling were via secondary cells, it is unlikely that these cells would project ipsilaterally directly through the optic tracts.

An ipsilateral retinofugal projection has not been previously described in the goldfish despite a number of extensive anatomical investigations using conventional silver methods. The sensitivity of [^3H]proline radioautography^{8,12} in defining small projections has allowed the detection of additional primary terminal fields. Our observation of primary, ipsilateral terminal fields and the course taken by fibers projecting to these areas was facilitated by the use of multiple postinjection survival periods which permitted us to take advantage of the differential distribution of labeled protein carried by fast and slow components of axonal transport. Rapidly transported proteins preferentially distribute to the region of the nerve endings, while those carried by slow flow principally contribute to axonal constituents².

Ipsilateral retinofugal projections have been considered to be absent in fishes, based on findings in a variety of teleosts^{1,17}. Examination of the retinal projections in the holosteans *Lepisosteus osseus*¹⁵ and *Amia calva*¹⁴, the two surviving members of holostean fish, however, revealed direct ipsilateral retinal projections to the diencephalon and rostral optic tectum. Holosteans represent an intermediate level of organization that gave rise to the radiation of teleosts monophyletically¹³. This led to the suggestions that the ipsilateral projections were lost in the teleost radiation or that these projections may have arisen independently in holosteans¹⁵. In view of the present report, it would appear that only the ipsilateral retinotectal projection was lost, and that the ipsilateral projections in holosteans represent retention of ancestral characteristics.

Our conclusion is supported by a recent radioautographic study which found an uncrossed ipsilateral projection to the dorsal thalamic nucleus and area pretecalis in *Astyanax mexicanus*²⁰. The findings with *Carassius* and *Astyanax* suggest that ipsilateral retinal projections are not an aberrant condition and are likely to be detected in other teleosts as well.

We thank B. W. Agranoff, R. G. Northcutt, R. E. Davis and A. M. Heacock for their helpful comments and T. Calloway for her technical assistance.

This research was supported by Research Grants NSF-BMS75-03810, MH12506-10 (B.W.A.) and by Predoctoral Training Grant MH-142719 (G.E.L.).

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