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Neuritic guidance by polyornithine-attached materials of ganglionic origin ☆

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Abstract

Dissociated ciliary ganglionic neurons do not survive in culture unless supplied with an appropriate source of ciliary neuronotrophic factors (CNTF). Moreover, previous work from this and other laboratories has shown that polyornithine (PORN) substrata per se do not allow neuritic development from such CNTF-supported neurons. Neuritic formation, however, would take place if the PORN substratum is coated with a neurite-promoting factor (NPF) from heart conditioned medium. Undissociated chick embryo ciliary ganglia explanted on PORN substrata will also produce a lavish *radial* neuritic outgrowth only if they are CNTF supported and the substratum is coated with NPF. In the absence of exogenous NPF neurites grow radially but only to a limited extent. At a distance of approximately 120 μm from the explant, neurites switch from a radial to a circular or tangential growth pattern, thus giving rise to a neuritic ring almost concentric with respect to the explant. Time-lapse studies show that the neuritic ring is generated through a combination of several behaviors that neurites display upon reaching the radial-to-circular "transition zone," including branching and turning either clock- or counterclockwise. Contributions are also made by interneuritic bridges arising from fusion of tangential filopodia from adjacent growth cones. The overall behavior of neuritic outgrowth suggests that the explanted ciliary ganglia are able to produce materials endowed with neurite-promoting properties, which coat the PORN substratum in the immediate vicinity of the ganglia. Autoradiographic experiments using ganglia prelabeled with either [^3H]glucosamine or [^{14}C]leucine demonstrated the existence of these materials. A periganglionic territory of PORN-attached materials (PAMs) of ganglionic origin can be recognized a few hours after explantation, before the onset of neuritic emergence from the ganglion. This PAM territory grossly corresponds to the area that a few hours later will be occupied by the neuritic ring. If two ganglia are explanted at a distance approximately equal to twice the distance normally covered by a dense periganglionic PAM halo, a continuity between the PAM territories of both ganglia is established before the onset of neuritic outgrowth. This overlapping generates an "avenue of preferential terrain" that provides guidance to neurites between the neighboring ganglia. The possibility is considered that a similar mechanism of neuritic guidance could also occur *in vivo*.

Abbreviations

CG, ciliary ganglion; CNTF, ciliary neuronotrophic factor; EE, embryo extract; HCM, heart-conditioned medium; HEBM, modified Eagle's basal medium; NPF, neurite-promoting factor; CG-NPF, ciliary ganglion-derived NPF; CM-NPF, conditioned medium-derived NPF; PAM, polyornithine-attached material; PORN, polyornithine

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