Investigations of ephrin ligands during development

A thesis submitted for the degree of Doctor of Philosophy
Molecular Biosciences, Adelaide University

By
Paul Toseh, B.Sc (Hons.)

Department of Molecular Biosciences,
Adelaide University, Australia,
Adelaide, South Australia, 5005
May 2002.
Abstract

The Eph receptors and their ligands, the ephrins, are the largest subfamily of receptor tyrosine kinases (RTKs). Functional studies of this kinase family have demonstrated their importance in various aspects of embryonic development. There include migration of precursor neural crest cells (Krull et al., 1997; Krull, 1998; Koblar et al., 2000), vascular development (Wang et al., 1998), axonal guidance and bundling of nerves in pathway formation in the nervous system, and hindbrain segmentation in embryonic development (Wilkinson, 2001). The overall aim of this thesis was to isolate ephrin ligands from Drosophila melanogaster, and to analyse their involvement in Drosophila development. In addition, the potential of ephrin-B1 as a causative gene in the human condition Aicardi's syndrome was also investigated.

The release of the Drosophila genome in 1999 revealed one full length EST coding for a gene that contained an ephrin core domain, designated d-ephrin. In situ analysis shows that d-ephrin mRNA is restricted to the CNS of Drosophila embryos at the time of axon pathfinding, suggesting that d-ephrin could play a role in nervous system development.

Bioinformatics analysis on d-ephrin was used to assign d-ephrin to either of the recognised ephrin-A or ephrin-B subclasses. Tissue culture experiments demonstrated that d-ephrin has an affinity for the only currently known Drosophila Eph (Dck). Analysis of misexpressed d-ephrin in Drosophila embryos showed axon guidance defects in the ventral nerve cord of Drosophila.

Detailed evolutionary analyses of all the currently known ephrin genes are in agreement with the current system of nomenclature derived by structural and functional studies. Furthermore, these evolutionary analyses placed the invertebrate ephrins equidistant from the ephrin-A and ephrin-B subclasses, respectively.

Aicardi's syndrome is an X-linked dominant disorder. Patients with this disorder typically present with a distinct collection of symptoms, including callosal agenesis, and retinal pigmentary defects (Aicardi et al., 1969). Genetic evidence taken from mouse models suggests that ephrin-B1 is involved in callosal formation (Henkemeyer et al., 1996). DNA sequence analysis of the h-ephrin-B1 genomic region in six known Aicardi's patients was performed to determine if h-ephrin-B1 is the principal disease causative gene of this disorder, however no mutations in the five exons of h-ephrin-B1 were found.
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