Research Briefings

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Genetic dissection of Amyotrophic Lateral Sclerosis pathogenesis in Drosophila

HVAPB IS THE CAUSATIVE GENE OF AMYOTROPHIC LATERAL SCLEROSIS8 (ALS8) IN HUMANS. WE GENERATED A MODEL FOR VAP-INDUCED ALS8 IN DROSOPHILA THAT RECAPITULATES MAJOR HALLMARKS OF THE HUMAN DISEASE. WE INTEND TO USE THIS MODEL TO DISSECT THE MOLECULAR MECHANISMS UNDERLYING ALS8 IN HUMANS. OUR APPROACHES SHOULD PROVIDE NEW INSIGHTS INTO THE PATHOGENESIS OF ALS AND IDENTIFY POTENTIAL NEW TARGETS FOR THERAPEUTIC INTERVENTION.

AMYOTROPHIC LATERAL SCLEROSIS

ALS, the most common Motor Neurone Disease (MND), is a fatal neuromuscular degenerative disease characterized by selective dysfunction and death of motor neurons leading to spasticity, muscle atrophy and paralysis. Since its first description by the French neurobiologist Jean-Martin Charcot more than 130 years ago, the mechanisms underlying the characteristic selective degeneration and death of motor neurons in ALS have remained a mystery. Although modern genetics has identified mutations in a few genes as a primary cause of the disease and implicated other ones as potential contributors, there is still no effective treatment for this progressive, fatal disorder.

The main objective of our research is to obtain new insights into the molecular mechanism of ALS pathogenesis by using the fruit-fly Drosophila melanogaster as an experimental model.

WHY STUDY HUMAN NEURODEGENERATION IN FLIES?

The small size, rapid generation time and low cost for maintenance of flies as compared to mammalian models, have made them an attractive model system to study human neurodegeneration. More importantly, recent evidence have shown that Drosophila is a powerful system to study human biology as more than 75% of human genes causing a disease have a Drosophila counterpart. In addition, entire genetic pathways are conserved between flies and humans. Over the last decade, a growing number of neurodegenerative diseases, including Parkinson's disease, Alzheimer's

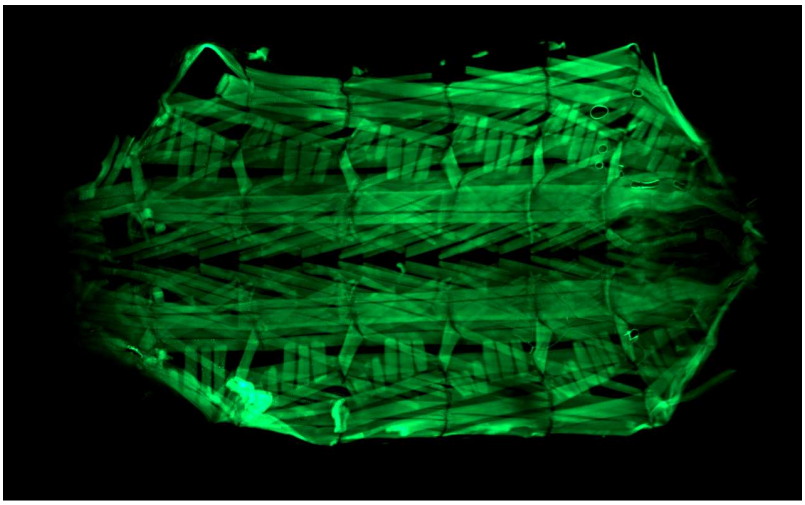

Figure 1: Phalloidin staining showing the musculature of a third instar Drosophila larva.

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disease and Huntington's disease have been modelled in Drosophila. These studies have provided important insights into the molecular pathogenesis of these diseases and have led to the identification of compounds with potential therapeutic effects that have been subsequently validated by preclinical trials in vertebrate models.

We have recently developed a ALS model in Drosophila and we intend to exploit it to dissect the genetic pathways underlying neurotoxicity in ALS.

MODELLING ALS IN DROSOPHILA

In 2004, hVAPB (human VAMP-associated protein B) was shown to be the causative gene of ALS8. To understand the patho-physiology underlying VAP-induced ALS in humans, we undertook a detailed functional characterization of VAP proteins in flies by using both transgenic and loss-of-function approaches. Drosophila VAP (dVAP), the stuctural homologue of hVAPB in flies, regulates structural remodelling at the Neuromuscular Junction (NMJ).

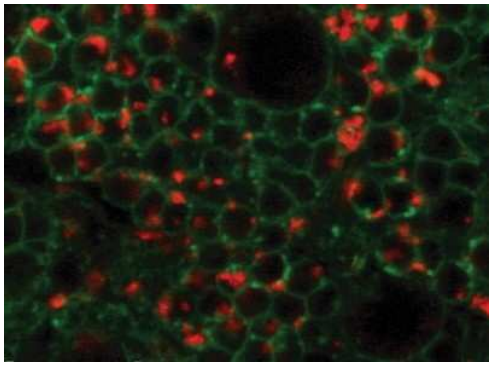

Figure 2: Aggregates immunoreactive for dVAP (red) in axons (right) and neuronal cell bodies (left) of larvae expressing dVAP mutant

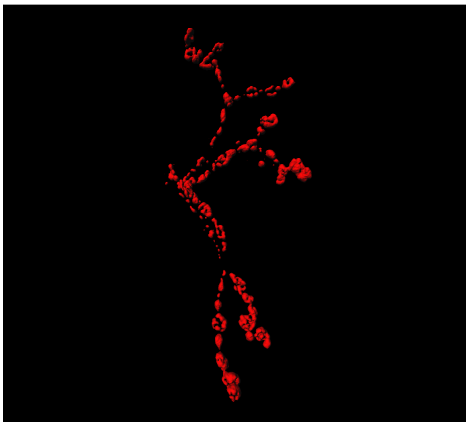

Figure 3: Volume renderings of clusters immunoreactive for glutamate receptor subunit III at the third instar larval NMJ.

Associated with these structural alterations, are compensatory changes in the physiology and the ultrastructure of NMJs which maintain synaptic activity within functional boundaries. More importantly, we have shown that hVAPB and dVAP are functionally interchangeable making results in flies relevant for the function of the human protein as well. Furthermore, we have shown that neuronal expression of the protein carrying the pathogenic mutation recapitulates several hallmarks of the human disease including locomotion defects, neuronal death and aggregate deposition.

DISSECTING THE PATHOMECHANISM OF ALS IN DROSOPHILA

We have undertaken biochemical and genetic approaches to identify new proteins interacting with dVAP. Several genes have been identified as physical interactors of dVAP and their functional characterization is an ongoing project in our lab. We have also initiated a genome-wide screen for an unbiased search of new genes that show interaction with dVAP. The function of these genes and their disease-related role is assessed by using a variety of techniques ranging from molecular genetics to electrophysiology and electron microscopy. This research will significantly contribute to our growing understanding of the molecular basis of ALS and will accelerate the progress of testing therapeutic strategies in mice and ultimately, in humans.

SELECTED REFERENCES

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