

Morpholino-Based Gene Targeting in the Zebrafish Embryo for the Modeling of Human Diseases

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We are utilizing advances in genomic information from vertebrate genome projects and in techniques for gene targeting to model human diseases in the simple vertebrate, *Danio rerio* (zebrafish). This organism has many advantages to offer for these studies. First, the zebrafish genome contains the rich complement (50-100,000) of vertebrate genes, a complexity lacking in simpler model systems such as *Drosophila* and the nematode. Second, zebrafish are vertebrate organisms and develop all major organs and organ systems using the same, conserved protein products found in higher vertebrates such as mice and man. Third, the transparent zebrafish embryos develop externally, a powerful combination of traits that allows unparalleled access to high quality developmental information using a simple dissection microscope. Fourth, zebrafish embryos develop rapidly – embryogenesis is complete by the end of 2 days of development, a rate that greatly facilitates the accumulation of data by the investigator in a short period of time. Finally, we developed morpholino-based antisense gene targeting tools for use in F0 embryos to enable very rapid loss of function tests based on sequence information using this model organism¹. We have used this tool to develop this organism for the direct study of holoprosencephaly. We have shown that similar to mice and men, zebrafish use products of the *sonic hedgehog* gene for critical midline signaling events during development¹. We are continuing the development of this model system to facilitate and enhance the molecular, biological and genetic advances in the understanding of this common human birth defect.

1. Nasevicius, A. and Ekker, S.C. (2000) *Nature Genetics*, **26**, 216-220.