GENE-BASED THERAPIES

Getting RNAi therapies to the brain

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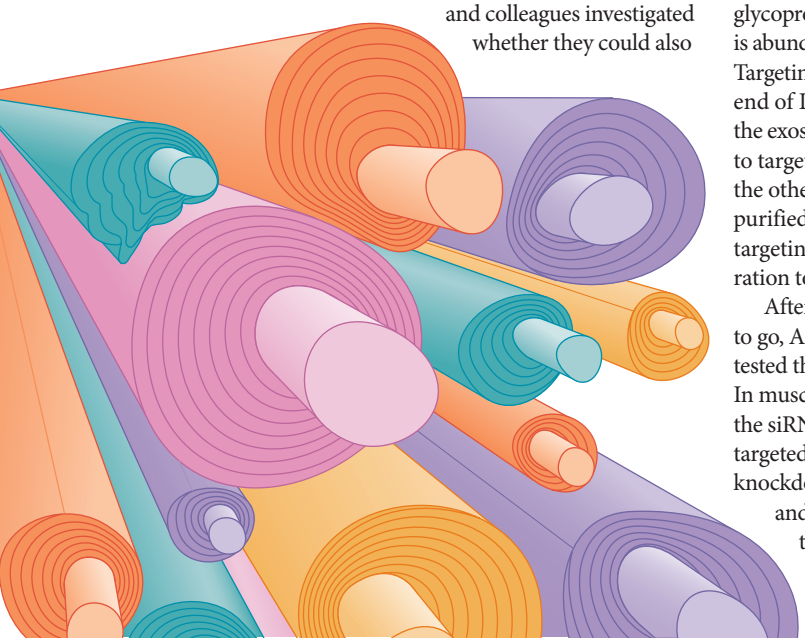
Therapies based on RNAi have great potential for the treatment of a range of diseases, but a stumbling block has been in developing suitable delivery methods. Delivery must be specific, allow efficient gene knockdown and avoid triggering an immune response. A new study in mice shows that these hurdles can be overcome by using exosomes — naturally occurring nanovesicles — to deliver small interfering RNAs (siRNAs). The authors used this approach in mice to achieve knockdown of a therapeutic target for Alzheimer's disease.

Exosomes are known to transport RNAs between cells, so Alvarez-Erviti and colleagues investigated whether they could also be used to deliver siRNAs. They purified exosomes from immature dendritic cells that were extracted from mouse bone marrow. These cells were chosen as they are less immunogenic than other cell types; precautions were also taken to avoid the triggering of immune responses by exosomes, by using donor and recipient mice that expressed the same major histocompatibility complex (MHC) proteins.



The next step was to use exosomes to target siRNAs to specific tissues. Alvarez-Erviti and colleagues transfected dendritic cells with constructs that encode modified versions of lysosome-associated membrane glycoprotein 2B (LAMP2B), which is abundant in exosome membranes. Targeting peptides were attached to the end of LAMP2B that lies outside the exosome; one version was designed to target exosomes to the brain and the other to muscle. The authors then purified exosomes carrying these targeting proteins and used electroporation to load them with siRNAs.

After the exosomes were ready to go, Alvarez-Erviti and colleagues tested them in cell culture and *in vivo*. In muscle and neuronal cell cultures, the siRNA-loaded exosomes achieved targeted delivery, efficient gene knockdown and low levels of toxicity and immunogenicity. The authors then injected the exosomes into mice that were of the same strain as those from which the dendritic cells were harvested. Importantly, unlike some other delivery methods, the exosomes did not show nonspecific targeting to the spleen, liver and kidney. Furthermore, the brain-targeted exosomes (although not those targeted to muscle) led to significant tissue-specific gene knockdown by the siRNAs they carried.

Finally, the authors used brain-targeted exosomes in wild-type mice to deliver siRNAs that are targeted against β-site APP-cleaving enzyme 1 (*Bace1*) — a candidate target for the treatment of Alzheimer's disease. Again, they saw tissue-specific knockdown of the target and, encouragingly, a significant decrease in the levels of a major component of the amyloid plaques that are associated with Alzheimer's pathology.

This demonstration of specific, systemic delivery, without evoking toxicity or an immune response, is an exciting step forward for RNAi-based therapies. Important next steps will include determining the optimal cell type from which to purify exosomes and, of course, extending this approach to the use of human exosomes.

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ORIGINAL RESEARCH PAPER Alvarez-Erviti, L. *et al.* Delivery of siRNA to the mouse brain by systemic injection of targeted exosomes. *Nature Biotech.* 20 Mar 2011 (doi:10.1038/nbt.1807)
FURTHER READING Davidson, B. L. & McCray, P. B. Jr. Current prospects for RNA interference-based therapies. *Nature Rev. Genet.* **12**, 329–340 (2011)