

Targeting Talk: Product Questions

by Dr. Douglas Lappi

Q: *In the Targeting Trends Newsletter, Oct-Nov-Dec 2006 you mentioned mixing anti-DBH-SAP with a tracer, which tracer would you recommend? We were thinking of using FluoroGold. If we do not use a tracer, we were thinking of using a neutral red solution to dilute the stock of anti-DBH-SAP in order to be able to visibly see the toxin being injected into the spinal cord. Could there be an issue of pH if we used neutral red with anti-DBH-SAP? Our concern is that the toxin is not being ejected from the pipette tip or that it is not being taken up into the pipette tip as we can not see it (it's the same color as the mineral oil). We are confident in the targeting of the spinal area for injection as we have previously used FluoroGold only and then were able to visualize it in the area of interest.*

A: Our Scientific Advisor, Dr. Ronald G. Wiley, uses Fast Green dye (0.01-0.1% w/v) in the toxin injection solutions. He originally chose Fast Green because intracellular electrophysiologists had long used it while doing intracellular recordings and shown it was non-toxic. Fast Green has more

contrast than Neutral Red (easier to see) and does not affect pH significantly. He has used it with many saporin-containing toxins with success.

Dr. Wiley says, "There are two issues when you talk about using "tracers" with targeted toxins: 1) tracing the acute injection volume to be sure it goes into the animal correctly, and 2) tracing the neurons that projected to the injection site and were therefore susceptible to being killed by the toxin.

Dr. Wiley does not use separate anatomic tracers for the immunotoxins, the only agents taken up and retrogradely transported efficiently. Since ATS immunotoxins are so efficient you have to use a high efficiency tracer such as cholera toxin B (but not WGA since it may not play well with saporin).

Dr. Wiley does not favor FluoroGold (a tin compound) because he has seen some local toxicity at FluoroGold injection sites which might impair uptake and/or transport of a targeted toxin, and it is not clear if it is compatible with saporin-containing toxins.



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performed using an NK-1 receptor antibody (discontinued). The data show that although NK-1 receptor density decreases as the animal matures, substance P (the NK-1 receptor ligand) remains an important part of these networks.

Orexin-B-saporin lesions in the lateral hypothalamus enhance photic masking of rapid eye movement sleep in the albino rat

Ocampo-Garces A, Ibanez F, Perdomo G, Torrealba F
J Sleep Res Epub, 2010.

Photic masking occurs when photic input to the retina interferes with REM sleep. Rats

that received 200 ng of orexin-SAP (discontinued) into the lateral hypothalamus experienced dramatically less REM sleep during normal light cycles. Placing them in a skeleton photoperiod (brief pulses of light, one in the morning and one in the evening), however, caused REM sleep during the rest phase to return to normal. These data suggest that photic masking may explain some effects of narcolepsy and cataplexy.

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