Targeting Talk

by Dr. Douglas Lappi

Q *Your targeted toxin data sheet gives the following instruction for disposal: "Care in disposal is mandatory; autoclaving or exposure to 1 M sodium hydroxide will inactivate the material. All labware that comes into contact with this material should be likewise treated." I am wondering if I can deactivate saporin by using 10% bleach or if everything has to be autoclaved?*

A Yes, you can use bleach to deactivate saporin prior to disposal or reuse of labware. If you are using nanogram quantities, these are too low to be toxic, so you can discard as you do your other non-hazardous laboratory materials without fear.

Q *I was wondering if there is any indication that intrastriatal administration of 192-IgG-SAP (Cat. #IT-01) will lesion the cholinergic neurons of the striatum. My sense from reviewing the literature is that these cholinergic neurons are not susceptible to the toxin, but I thought I'd ask to see if you had any information / experience regarding this point.*

A No, I don't think it will work because the target of 192-IgG-SAP is p75, LNGFr, which is only expressed on the rat basal forebrain cholinergic neurons. Those striatal neurons don't express p75 in the adult. The NK1r is often expressed in the striatum, and you can use SSP-SAP for them, but that's the best we can do right now.

Q *I have a few questions about the Alexa488-labeled affinity purified NGFr antibody (Cat. #FL-03).*
Is it specific to extracellular p75?
Can you use it on live cells? Does it work on fixed cells?
Does it cause activation of the p75 receptor (i.e., result in apoptosis or changes in axon outgrowth in neuronal cells)?

A This product does recognize extracellular p75 in both live and fixed cells. As for the activation, that's an interesting question. There is no evidence of 192-IgG either causing apoptosis or neurite outgrowth as far as I can see. Chandler *et al*. (1984) report that the antibody "partially inhibits the regeneration of neurites from primed PC12 cells," and it enhances NGF binding. But that's about it, despite several studies being done with PC12 cells and *in vivo*. We assume all this holds upon treatment with 192-IgG-SAP — until the cell dies from saporin poisoning.

Chandler CE, Parsons LM, Hosang M, Shooter EM (1984) A monoclonal antibody modulates the interaction of nerve growth factor with PC12 cells. *J Biol Chem* 259(11):6882-6889.

Q *I purchased your secondary conjugate, Mab-ZAP (Cat. #IT-04). I am preparing to do a cytotoxicity assay and I'm wondering if my primary antibody should be sterilized prior to combining with Mab-ZAP?*

A Depending on the conditions of your lab in which you are using your antibody, it is possible that within a 72-hour period, you may see bacterial growth in your plates if the antibody was accidentally exposed to bacteria. It is recommended that, if you feel comfortable with the antibody, you can just go ahead and try it without sterilizing it, and if you do see bacterial growth, you can certainly filter sterilize the material through a 0.2 micron filter before using.

Society for Neuroscience
November 3-7, 2007
San Diego, CA

American Society for Cell Biology
December 1-5, 2007
Washington, DC