Targeting Talk: Product Questions

by Dr. Douglas Lappi

CORRECTION: The protocol printed in Targeting Trends Volume 10, Issue 3 was incorrect. The corrected protocol is presented at right.

Q: We're interested in trying out your melanopsin antibody (Cat. #AB-N38) using immunohistochemistry in mouse retina. Do you have a recommended protocol?

A: Please see corrected protocol from Panda *et al*. This protocol is also available on our website. Just click on "Protocols" on our Home Page. ***www.ATSbio.com***

Panda S. *et al*. 2002. Melanopsin (Opn4) requirement for normal light-induced circadian phase shifting. *Science* 298(5601):2213-2216.

Targeting Teaser Winners

The solution to the puzzle was:

Jumbles: MICROGLIA
SLICE
DENSITY
VISUALIZE
COMPOUND

Answer: He provided the right . . .STIMULUS

Congratulations to the puzzle solvers from our last newsletter. Each winner receives $100 credit towards research product purchases from Advanced Targeting Systems.

WINNERS: David Looney, UCSD Pathology, La Jolla, CA * Megan Green, Univ TX Health Science Ctr Pharmacology, San Antonio, TX * Erica Edwards, Oklahoma Medical Research Fdtn, Oklahoma City, OK * Shikha Gaur, City of Hope Clinical & Molecular Pharmacology, Duarte, CA * Bruce Pappas, Carleton Univ, Ottawa, ON * Ruth Stornetta, Univ Virginia Pharmacology, Charlottesville, VA * April Price, UCSF Medicine, San Francisco, CA * Brigitta Peteri, CNRS FRE 3094, Nice, FRANCE * Glenn Kageyama, Cal Poly Pomona Univ, Pomona, CA * Jean Peduzzi, Wayne State Univ School of Medicine, Detroit, MI * Muthu Kumara gnana sammandh, National Univ Singapore, Bioengineering, Singapore * Seto Chice, SUNY-HSC, Brooklyn, NY * Cai Peng, ApoPharma Inc, Ottawa, ON * Elia Nahas, McGill Univ, Montreal, QC * Ashley Linder, UCSD Neurobiology, La Jolla, CA

Solve the Teaser online at:
http://www.atsbio.com/news/09q4_teaser.html

Anti-Melanopsin Immunostaining Protocol
*** corrected ***

Remove the corneas, and postfix eyes at 4°C for 24 hours in 4% paraformaldehyde in phosphate-buffered saline (PBS). Remove lenses.

Cryoprotect eyecups for sectioning at 4°C for 24 hours in 30% sucrose in PBS; embed the eyecups in OCT medium (Sakura Finetek, Torrance, CA), freeze, section (16-20 µm), and thaw-mount onto gelatin-coated slides.

Dissect retinas destined for flat-mounting from eyecups immediately after postfixation, stretch onto filter paper, and process in 1.5-ml microfuge tubes.

Wash tissue (slides and flat-mounts) 3 times (10 min, 4°C) in Tris-buffered saline (TBS, Quality Biological, Gaithersburg, MD) and block for 30 min at 4°C in 1.5% normal goat serum in TBS.

Incubate tissue for 24 hr at 4°C in a 1:2,500 dilution of anti-Melanopsin UF006 (Cat. #AB-N38) in a TBS-incubating buffer containing 1% bovine serum albumin, 0.25% carrageenan lambda and 0.003% Triton X-100.

Wash slides and flat mounts three times in TBS (10 min, 4°C) and incubate for 1 hour at 22°C in Cy3-conjugated anti-rabbit IgG antibody (Jackson ImmunoResearch Laboratories, West Grove, PA) diluted 1:500 in TBS incubating buffer.

Wash 3 final times in TBS (10 min, 22°C).

Remove flat-mounts from the filter paper and transfer onto glass microscope slides. Mount flat-mounts and sections in DAPI-containing Vectashield (Vector Laboratories, Burlingame, CA), coverslip, and seal with clear fingernail polish.

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October 17-21, 2009 • Chicago, IL
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American Society for Cell Biology
December 5-9, 2009 • San Diego, CA

American Association for Cancer Research
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