

ALAN E. LEVITON STUDENT RESEARCH AWARD REPORT

Fluorophore Labeling of Two Temporally Defined Protein Populations in Mammalian Cells

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Kimberly Beatty is recipient of a 2007 AAAS, Pacific Division Alan E. Leviton Student Research Award.

She has since completed her Ph.D. and is starting a post-doc at the University of California, Berkeley.

David Tirrell was her faculty advisor.



The proteome can undergo complex dynamic changes in response to disease or environment. While there are many methods for identifying proteins, a more complete examination of the proteome would include complementary, time-resolved images of proteins in their natural environment. In 2006, Beatty and coworkers demonstrated selective dye-labeling of a subset of the proteome in mammalian cells. Cells were pulselabeled with a reactive methionine (Met) analogue, which permitted labeling of newly synthesized proteins with a fluorescent dye using an azide-alkyne ligation. The application of two reactive Met analogues, azidohomoalanine (Aha) and homopropargylglycine (Hpg), to dye-label two distinct protein populations inside fixed cells has now been developed, and will enable changes in the proteome to be tracked over time. Reactive lissamine rhodamine LR, 7-dimethylamino coumarin (DMAC), and Bodipy-630 (BDPY) dyes were synthesized and examined for selective dye-labeling of newly synthesized proteins. The LR and DMAC, but not BDPY, fluorophores were found to enable selective, efficient labeling of a subset of the proteome. Next, distinct and temporally defined protein populations were tagged by simultaneous and sequential pulse-labeling with Aha and Hpg. After pulse-labeling, cells were two-dye labeled using the reactive LR and DMAC dyes. Two-dye labeled cells were imaged using fluorescence microscopy and analyzed by flow cytometry, revealing that this new method can be used to selectively dye-label and image two distinct protein populations inside cells. The manuscript describing this work is in preparation (2008). The funds provided by AAAS, Pacific Division Alan E. Leviton Student Research Award were used to purchase and prepare the reactive LR, DMAC, and BDPY dyes used to dye-label newly synthesized proteins.

to help defray their costs to the meeting. See page 4 for additional information.

Field trips: Pre-registration for all field trips is required due to limited seating in the vehicles and the need to inform some destinations of the number of people arriving. If you are interested in one or more of the excursions, it is recommended that you register early. At least one member of a family group requesting field trip reservations must be a paid meeting registrant.

Please send your Advance Registration Form and accompanying payment to AAAS, Pacific Division, Southern Oregon University, 1250 Siskiyou Blvd., Ashland, OR 97520. Alternatively, you may phone (541-552-6869) or fax the information (541-552-8457 – a dedicated fax line).

ACCOMMODATIONS AND FOOD SERVICE

Housing and meals at the Hawaii Preparatory Academy (HPA). The Pacific Division has a very attractive arrangement with HPA to provide several nights lodging and meals at an affordable price for those attending the meeting. Please keep in mind that the lodging is in typical dorm rooms, and is priced either as singles (one person per room) or doubles (two persons per room). Bathrooms, including shower rooms, are

in common facilities, either down the hall or on an adjacent floor. Bed linens and a washcloth and towel are provided. The rooms have internet access and telephones. Washers and dryers are available for use at no cost (bring your own detergent or purchase at the bookstore) for those staying in the dorms.

We have negotiated the following room and board and meals-only packages.

Lodging with Meals (see exception in C. below):

- A. Four nights lodging** (Sunday, 15 June – Wednesday, 18 June) **and ten meals** (Sunday: reception/dinner; Monday: breakfast, lunch, Hapuna Beach picnic dinner; Tuesday (field trip day): breakfast only; Wednesday: breakfast, lunch, Division Luau-type Banquet; Thursday: breakfast, lunch). Cost: \$410 single; \$300 double (per person); \$140 per child (up to two children age 15 or younger with at least one parent in a room).
- B. Five nights lodging** (Sunday, 15 June – Thursday, 19 June) **and twelve meals** (as package “A” but with dinner Thursday and breakfast Friday added). Cost: \$501 single; \$363 double (per person); \$166 per child (see A. above)
- C. Additional nights lodging** (only as additions to packages A or B above) – Friday, 13 June, Saturday, 14 June, and/or Thursday, 19 June: Cost \$68.50 per night single; \$42.50 per night double (per person); \$10.50 per night per child