Winter 2010

Adrenocorticotropin Hormone Expression in the Developing Chicken Limb

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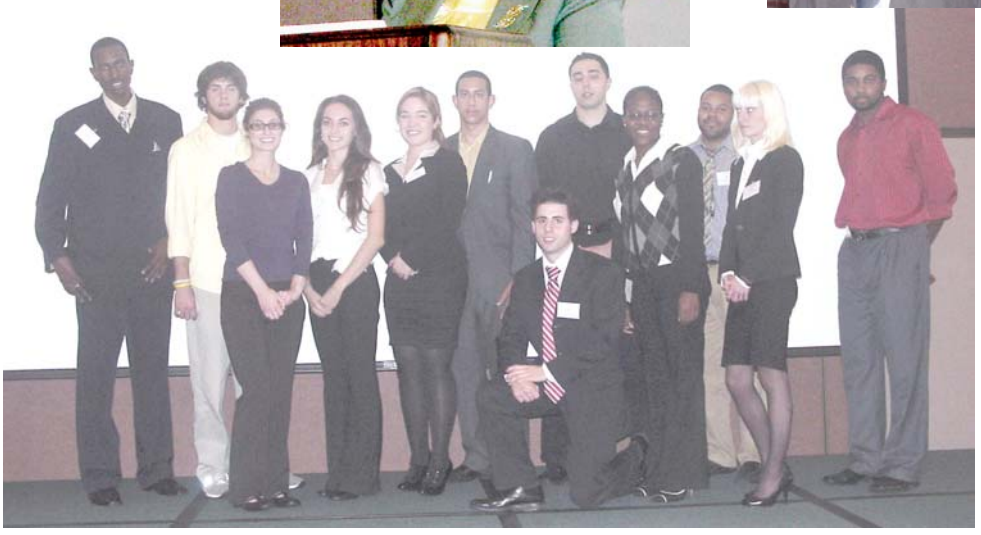
MACUB

IN VIVO

The Publication of the Metropolitan Association of College and University Biologists

Winter 2010 Volume 31, Issue 2

42nd Annual MACUB Conference Hosted By Kingsborough Community College October 24, 2009



A Publication of the Metropolitan

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Articles can be submitted electronically to *invivo@mec.cuny.edu* or mailed as a printed copy (preferably with a diskette that contains the file) to the Editorial Board at Medgar Evers College. All submissions should be formatted double spaced with 1 inch margins. The title of the article, the full names of each author, their academic affiliations and addresses, and the name of the person to whom correspondence should be sent must be given. As a rule, full length articles should include a brief abstract and be divided into the following sections: introduction, materials and methods, results, discussion, acknowledgments and references. Reviews and short communications can be arranged differently. References should be identified in the text by using numerical superscripts in consecutive order. In the reference section, references should be arranged in the order that they appeared in the text using the following format: last name, initials., year of publication. title of article, journal volume number: page numbers. (eg. - [1]Hassan, M. and V. Herbert, 2000. Colon Cancer. *In Vivo* **32:** 3 - 8). For books the order should be last name, initial, year of publication, title of book in italics, publisher and city, and page number referred to. (eg. - Prosser, C.L., 1973. *Comparative Animal Physiology*, Saunders Co., Philadelphia, p 59.). Abbreviations and technical jargon should be avoided. Tables and figures should be submitted on separate pages with the desired locations in the text indicated in the margins.

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Optimization of Fatty Acid and Cholera Toxin Concentrations for Treatments of Epithelial Cells: Can Fatty Acids Provide Mucosal Immunity against Cholera Infections? Joanna Tychowski[1], Paula Cobos[2], Laura Lorentzen[1] and Farshad Tamari[3], [1]New Jersey Center for Science Technology Mathematics Education, [2]Kean University, Union, NJ and [3]Kingsborough Community College, Brooklyn, NY.

Cholera is caused by infection of the small intestine by *Vibrio cholera*. In developing countries it can be severe or fatal. The two submit Cholera Toxin (CT) binds to the cell surface allowing one subunit to enter the cell. In response, cyclic AMP levels are elevated, influencing electrolyte and cytokine balances. Fatty acids (FAs) such as oleic, linoleic, and linolenic acids, found in flax (*Linum*) seeds, have medicinal properties. Our ultimate objective is to explore whether metabolites of the above FAs can provide any degree of mucosal immunity, as determined by cytokine dynamics in response to CT challenge. Our first goal is to determine the maximum and minimum concentrations of FAs and CT, respectively, that murine and human epithelial cell can be exposed to. The following control (C) and experimental (E) treatments will then be performed and cytokine levels will be quantified and compared using ELISA: 1. No FA or CT treatment (C), 2.FA treatment only (C), 3. CT treatment only (C), and 4. Pre- and simultaneous treatments with both FAs followed by CT challenge (E). Thus far, using mouse macrophages and MTT assays, the optimum concentrations for oleic, linoleic, and linolenic have been determined at 5-50ng/µL.

Genome-wide Distribution of G-quadruplexes in the Transcribed Regions of Human Genes. Viktor Vasilev, Lawrence D'Antonio, and Paramjeet S. Bagga, Ramapo College of New Jersey, Mahwah, NJ.

G-rich DNA and RNA G-quadruplexes can play significant biological roles in important cellular processes and human disease. The goal of current studies in our lab has been to investigate the role of G-quadruplexes in post transcriptional regulation of gene expression. We have used a bioinformatics approach to study the composition and distribution patterns of G-quadruplex forming motifs in the transcribed regions of >17,000 protein coding human genes. G-quadruplex motifs were found in almost all of the >500,000 of exons and introns that were analyzed. Our studies revealed the prevalence of G-quadruplexes with high putative stability near 5' splice sites in the introns. Stable RNA G-quadruplexes in the vicinity of 5' splice site may be involved in modulating splicing via interactions with regulatory proteins that bind G-rich sequences and influence alternative and tissue specific splicing events. We also found a very strong correlation between the distribution of the positions of ESEs (Exonic Splicing Enhancers) and G-quadruplexes, especially in the exons. Further investigation revealed overlaps between the predicted ESEs and G-quadruplexes mapped near the splice sites. ESE mediated regulated splicing may in fact involve the G-quadruplex structure. Our findings suggest that G-quadruplexes play a regulatory role in splicing of the human pre-mRNAs.

Study on the Effect of Cupric Chloride and Cadmium Chloride on Cyanobacteria *Synechococcus sp*. IU 625. Vico Viggiano[1], Shyam Patel[1], Jose L. Perez[1], Tin-Chun Chu[2] and Lee H. Lee[1], [1]Montclair State University, Montclair, NJ and [2]Seton Hall University, South Orange, NJ.

Cyanobacteria, *Synechococcus sp.* IU 625, were used because they are good indicators of water contamination by heavy metals. In this experiment, the effect of $CuCl_2$ (0, 5, 10, 15 and 30 mg/L) and $CdCl_2$ (0, 10, 15, 25, and 30 mg/L) on the growth of cyanobacteria *S. IU 625* were studied. Growth was monitored by direct count using hemocytometer and turbidity study using spectrophotometer at wavelength 750 nm. The content of $CuCl_2$ and $CdCl_2$ in the cells and media was analyzed by using ICP (Inductively Coupled Plasma) spectrometer. In the cultures containing $CuCl_2$, the growths were similar except 30 mg/L, where the growth was inhibited. ICP study indicated that 87 to 100% of $CuCl_2$ stays outside of the cells. In the 5 mg/L $CdCl_2$ culture, the growth was the same as the control and in 15mg/L it was slightly inhibited. At 30 mg/L, the growth was almost completely inhibited. ICP study indicated that 70 to 80 % of the metal stays in the media. This study suggested that the cells have low permeability to $CuCl_2$ and $CdCl_2$ and permeability may be one of the reasons that the cells are able to tolerate the metal contamination.

Adrenocorticotropin Hormone Expression in the Developing Chicken Limb. Michele J. Vigliotti and Jodi F. Evans, Molloy College, Rockville Centre, NY.

In previous studies using mammalian models we have found both clinical and laboratory evidence of a role for melanocortins in endochondral ossification. The melanocortin system has remarkable conservation among vertebrates and melanocortin receptors are expressed with significant sequence homology in teleosts to mammals. The overall goal of these studies is to provide a more accessible model of melanocortin involvement in endochondral growth. We hope to determine if melanocortins play a role during endochondral ossification of the developing chicken limb. Like in mammals melanocortins are widely distributed throughout the body of chicken and participate in a wide range of physiological functions with the peripheral tissue distribution of melanocortin receptors in chicken more widespread. Our first step was to examine melanocortin expression in the developing limbs of the chick embryo. Using immunohistochemistry techniques, we detected ACTH (1-24) in the limbs of embryonic day 9 chick embryos. This initial data indicates that the chick embryo is a viable model that can be used to determine a role for melanocortin in endochondral growth. Melanocortin expression shows remarkable sequence homology, therefore results of these studies can be extrapolated to many vertebrate models.

Development of Purification of Valproic Acid and Butyric Acid for Positron Emission Tomography Studies. Khaing Win[1] and Sunny Kim[2], [1]St. Joseph's College, Brooklyn, NY and [2]Brookhaven National Laboratory, Upton, NY.

Valproic acid (VPA) and butyric acid (BA) are two epigenetic drugs used for seizures and neurocognitive disorders. While the two acids have been known to bind histone deacetylases that suppresses gene expression, their pharmacokinetics, biodistribution, and the blood brain barrier penetrability remain an enigma. Positron Emission Tomography (PET) using [^{11}C]VPA and [^{11}C]BA could potentially solve these issues. Before [^{11}C]radiosynthesis, purification methods for unlabeled VPA and BA, generated via Grignard precursors, as impure mixtures were developed. High Performance Liquid Chromatography (HPLC) with C18– Gemini column under the isocratic system (acetonitrile (MeCN) and formic acid (FA)) is used. The following optimum purification conditions were found: a 50% MeCN/50% FA for VPA and a 15% MeCN/85% FA for BA. Respective HPLC (flow rate=1ml/min) retention times for BA and VPA were 8.55 minutes and 11.76 minutes. Our preliminary radiosynthesis and purification of [^{11}C]BA was completed within 40 min after the End of Bombardment. [^{11}C]BA was obtained in moderate radiochemical yield (>40%) and high purity (>99%). Radiosynthesis of [^{11}C]VPA is still to be attempted. We have successfully developed conditions for the synthesis and purification of both unlabelled VPA and BA for preparation of the radiolabeled acids to be used for PET studies.