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CELLULAR AND MOLECULAR

THE DISTRIBUTION OF UBIQUITIN-PROTEIN CONJUGATES IN VARIOUS REGIONS OF RAT BRAINS

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In normal aging Alzheimer's disease, senile plaques containing the β /A4 fragment of the amyloid precursor protein and neurofibrillary plaques dominated by the cytoskeletal protein tau are both present. Both of these lesions also contain other proteins, including ubiquitin which is a highly conserved protein involved in an ATP-dependent proteolytic pathway. The focus of this research is to assess any alteration in the function of the Ub proteolytic pathway during aging which could be a primary contributor to the accumulations of abnormal proteins. The experiments described here involve the identification of Ub-protein conjugates which consist of a target protein covalently linked to Ub which marks the target for degradation. Brain regions from Sprague-Dawley rats (striatum, thalamus, hippocampus, cerebellum, cerebral cortex) are isolated and selected for the presence or absence of significant accumulations of age-related pathology. The tissues are processed for protein content, and the proteins are separated on SDS-PAGE electrophoresis. Immunological labeling of ubiquitin reveals a pattern of proteins which correlates with the occurrence of ubiquitinated proteins in a given population of neurons. Comparison of these patterns across brain regions and different aged samples reveals differential functioning of the Ub pathway.

ROLE OF ZEB-BINDING MOTIF WITHIN THE KAPPA B SITE OF THE IL-2 GENE ENHANCER

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A zinc-finger protein called Zeb binds to both the NRE-A site of the IL-2 gene enhancer and an E-box site of the I μ heavy chain enhancer, and through binding, represses transactivation. A motif similar to the Zeb-binding sequences of both these sites, has been found overlapping the IL-2kB site. This motif binds with recombinant Zeb protein *in vitro*. It is speculated that this kB Zeb-binding sequence represses transactivation and accounts for the weak transactivational potential of the IL-2kB enhancer element. The capacity of this kB Zeb-binding motif to repress transactivation of a strong positive enhancer element called NFAT, was tested using Jurkat cells transiently expressing luciferase reporter gene constructs containing inserts of NFAT linked to either wild-type or mutated Zeb sequences. The mutation within the Zeb site has been shown to eliminate binding to recombinant Zeb *in vitro*. Our hypothesis is that upon stimulation, cells expressing constructs with mutated Zeb sequences will show lower levels of luciferase activity due to the repressive activity of the Zeb sequence. Due to inconsistencies in preliminary data, we are currently exploring the effect of the mutation on transactivation by the IL-2 gene enhancer.

EXPLORING CATALYSIS AND REGULATION OF A MAIZE PHOTOSYNTHESIS ENZYME USING A RECOMBINANT HISTIDINE-TAGGED FORM OF THE ENZYME

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A major enzyme of the C₄-photosynthetic pathway of higher plants is pyruvate, orthophosphate dikinase (PPDK). PPDK has been demonstrated by several studies to limit the amount of CO₂ assimilated into sugars by C₄ photosynthesis in maize leaves. Our goal to fully understand how this enzyme works and how it is controlled in the maize leaf. To accomplish this,

we have developed a recombinant form of the enzyme that has been modified to include a series of six histidines on the N-terminal end of the protein. This histidine "tag" enables mutated forms of the enzyme to be easily recovered from *E. coli* extracts via rapid one-step metal affinity column chromatography. We have employed this purification system for exploring enzyme regulation and catalysis via site-directed mutagenesis of the gene. Preliminary analysis of several active-site mutants will be presented.

CLONING AND EXPRESSION OF THE MAIZE PHOTOSYNTHESIS GENE PYRUVATE, ORTHOPHOSPHATE DIKINASE IN THE BACTERIUM ESCHERICHIA COLI

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A cardinal enzyme of the C₄-photosynthetic pathway of higher plants is pyruvate, orthophosphate dikinase. We have cloned the gene for C₄-pyruvate, orthophosphate dikinase (PPDK) from maize (*Zea mays*) into an *E. coli* expression vector and produced recombinant PPDK in *E. coli* cells. Recombinant enzyme was found to be expressed in high amounts (5.3 U enzyme-activity liter⁻¹ of induced cells) as a predominantly soluble and active protein. Biochemical analysis of partially purified recombinant PPDK showed this enzyme to be equivalent to enzyme extracted from illuminated maize leaves with respect to (i) molecular mass, (ii) specific activity, (iii) substrate requirements, and (iv) phosphorylation/inactivation by its bifunctional regulatory protein. We have employed this recombinant form for exploring enzyme regulation and catalysis via site-directed mutagenesis of the gene. Our ultimate goal is to improve photosynthesis in corn by genetically engineering PPDK for improved photosynthetic performance.

COMPARISON OF FOUR METHODS OF ISOLATING GENOMIC DNA FROM TILAPINE FISH

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DNA was isolated from the muscle tissue of Tilapine fish using four different methods: A fish DNA extraction protocol using methylene chloride, a phenol/proteinase K protocol, a protocol using phenol and chloroform without proteinase K, and a commercially available kit utilizing silica powder (QUICK-Geno Kit, Clontech, Palo Alto, CA). We compared the four methods in terms of yield and purity of isolated DNA, time required, and cost. Our results to date show that the commercial kit gave the highest yield and purity of DNA in the shortest time period. However, this technique is the most expensive at \$3.40 per extraction from 100 mg of tissue, compared with \$2.07 per extraction for the next most expensive procedure.

POTENTIAL PITFALLS OF USING MACCONKEY AGAR TO IDENTIFY RECOMBINANT *E. COLI*

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Genetic transformation by α -complementation involves the coexpression of a chromosomal, mutant *lacZ* gene and a plasmid-borne *lacZ* fragment. These two inactive gene fragments encode protein products that can combine to form a functional β -galactosidase protein. This technique is widely used for screening recombinant bacterial colonies. Cells transformed with non-recombinant plasmids acquire color when grown on the appropriate medium, whereas cells transformed with recombinant plasmids remain colorless. LB agar with X-gal has traditionally been used for colony screening; however, X-gal is expensive and uneven distribution of X-gal on the medium surface may result in false-positive identification of *lac*⁻ colonies. MacConkey agar was compared to LB agar for the screening of α -

complementing *E. coli* transformants. *E. coli* strains XL1-Blue, TG1, and DH5 α were transformed with pGEM5zf(+), or with an equal molar mixture of pGEM5zf(+) and pGT3. We discovered two potential pitfalls of using MacConkey agar: 1) a 2-3 fold decrease in transformation frequency, and 2) the inability to distinguish between recombinant and non-recombinant colonies after 48 hours. Both media provide a means for the identification of recombinant and non-recombinant colonies. The suitability of either type of media ultimately depends upon the researcher's objective.

IDENTIFICATION OF GENES REQUIRED FOR THE FUNCTION OF CYTOPLASMIC DYNEIN IN *DROSOPHILA MELANOGASTER*

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Using a genetic approach we have attempted to uncover gene loci that encode products necessary for the proper function of cytoplasmic dynein in *Drosophila*. Several mutations of the *Drosophila* cytoplasmic dynein heavy chain gene, *Dhc64C*, have previously been identified and characterized. Of these mutants, some act as dominant suppressors or enhancers of the rough eye phenotype caused by the dominant *Glued*¹ mutant (McGrail et al., JCB in press). It has been demonstrated that this *Glued* gene encodes a polypeptide sharing sequence homology with the p150 component of vertebrate dynactin which serves to regulate the function of dynein (Holzbaumer et al., Nature 1991; Gill et al., JCB 1991). Complementation tests between the *Dhc* allele and the *Glued* mutant indicate a functional interaction between the encoded gene products. A series of screens using deficiencies spanning the entirety of the second and third chromosomes, approximately 2/3 of the *Drosophila* genome, was conducted to uncover additional loci that exhibit dominant interactions with either the *Gl* mutant or a *Dhc* mutant. Several deficiencies that enhance or suppress the roughness of the *Glued* eye phenotype have been identified. Transposons (P-inserts) in the regions of these deficiencies are being used to aid in the molecular characterization of the genes causing the interaction.

GROWTH FACTOR INFLUENCES ON FORELIMB REGENERATION OF ADULT NEWTS

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Regeneration of amphibian forelimbs is regulated by the nervous, endocrine, and immune systems and likely is mediated by growth factors and other bioactive agents. This study examined the influences of growth factors (the insulin-like growth factors [IGF-I and -II] and transforming growth factor beta [TGF β]) on forelimb regeneration of adult newts, *Notophthalmus viridescens*. Growth factor-impregnated Dowex beads were implanted into forelimbs proximal to the site of amputation either on the day of amputation or 12 days later and the progress of regeneration followed. Initial blastema formation was not affected by any of the treatments. IGF-I appeared to accelerate blastema growth (i.e., cone stages was achieved earlier) but may have impaired differentiation or morphogenesis. IGF-II did not affect early stages of regeneration, but may have impeded later stages as did IGF-I. IGF effects did not differ with time of administration. In contrast, regeneration appeared to be accelerated when TGF β was administered on the day of amputation but arrested at the bud stage when it was administered within 5 days of bud emergence. These results are consistent with differential effects of growth factors on forelimb regeneration. Further studies will explore the role and mechanism(s) of action of these growth factors. [This work was supported in part by a grant from the Minnesota Medical Foundation.]

CLONING OF THE *TRICHODERMA HARZIANUM* CBH 1 GENE'S 5' AND 3' ENDS BY INVERSE PCR

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Trichoderma harzianum FP108 is a fungal strain able to bioconvert cellulose into glucose. The CBH1 gene (exocellobiohydrolase) was amplified by polymerase chain reaction (PCR) using primers based on the DNA sequence of the *Trichoderma reesei* QM9414 CBH1 gene. Because the PCR primers overlapped the 5' and 3' ends of the gene, the flanking regions of the CBH1 gene were not amplified. However, following our sequencing of the CBH1 gene, we were able to clone the flanking regions using inverse PCR. Using this technique, we were able to clone both the 5' and 3' flanking regions of the *T. harzianum* CBH1 gene.

AN ANALYSIS OF THE EFFECTS OF THE PROTEIN KINASE INHIBITOR KT5926 ON NERVE CELL GROWTH CONE ACTIVITY

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For abstract, see Winchell Papers.

INTERACTION OF THE THYROID RESPONSE ELEMENT AND A NUCLEAR FACTOR I BINDING SITE IS REQUIRED FOR ENHANCED MYELIN BASIC PROTEIN GENE EXPRESSION

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For abstract, see Winchell Papers.

PH SENSITIVITY THRESHOLDS IN MEMBRANE ATPASES OF THE YEAST *SACCHAROMYCES CEREVISIAE*

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For abstract, see Winchell Papers.

CHEMISTRY/BIOCHEMISTRY

SYNTHESIS OF POLY(PHENANTHRYL METHYLENE)

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This research involves the preparation and characterization of poly(phenanthryl methylene). The poster will describe the synthesis of the monomer, 9-chloromethyl phenanthrene. This begins by reducing 9-phenanthrene carboxyaldehyde to 9-phenanthrene methanol. The alcohol is then chlorinated by substitution to produce 9-chloromethyl phenanthrene. Obtaining a high yield of this product has proven to be very difficult and much work has been done to improve yields. The polymer is then synthesized by Friedel-Crafts self condensation of the 9-chloromethyl phenanthrene to produce poly(phenanthryl methylene). Characterization data of the monomer and the polymer will also be described.

THE EFFECT OF LOW STRENGTH MAGNETIC FIELDS ON ASCORBATE DEPENDENT LIPID PEROXIDATION

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Numerous types of cytotoxicity have been proposed from exposure to low strength magnetic fields, such as those occurring under high voltage transmission lines. One such toxicity is lipid peroxidation, which is mediated by free radical mechanisms. The peroxidation of the membrane lipids has been shown to cause the destruction of many subcellular organelles and cytoplasmic constituents such as oxidizable small molecules, certain enzymes and proteins. The effects of low strength magnetic fields on ascorbate dependent lipid peroxidation were examined by measuring

thiobarbituric acid reactive substances (TBARs) in a non-enzymatic system. Non-enzymatic lipid peroxidation was carried out in liposomes prepared from total lipids extracted from pig liver microsomes. Lipid peroxidation required ascorbate and the prooxidant iron complexes ADP-Fe⁺³ and EDTA-Fe⁺³. Our results show that a 10G magnetic field significantly increases in vitro lipid peroxidation.

THE EFFECTS OF HYDROXYLATION AND MUTATION ON COLLAGEN STABILITY

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Collagen proteins are the major fibrous elements in connective tissues such as bone, tendons, skin, and blood vessels in the human body. Although the 3-dimensional structure (tertiary structure) of a number of collagen-like proteins is well known, a detailed understanding of structure and stability, as it relates to amino acid sequence, has not yet been achieved. Using fragment models of the collagen triple helix (composed of 3 left hand helical chains wound in a right-hand superhelix), substitutions to Y position in the repeat sequence (glycine-X-Y)_n have been studied on a structural basis. Computer simulation techniques and X-ray data have been applied to determine how these substitutions modify thermal stability. In particular, the relative free energy of triple helix formation for Y=proline versus Y=hydroxyproline has been calculated using free energy simulations. Molecular dynamics calculations of these structures are also performed to determine the role hydration plays in stabilizing triplex formation. The results are compared and contrasted with all available experimental data for known collagen structures and sequences.

ECOLOGY

BIRCH SAP TREE UTILIZATION BY YELLOW-BELLIED SAPSUCKERS, RUBY-THROATED HUMMINGBIRDS AND RED SQUIRRELS

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Many animals such as hummingbirds and squirrels may benefit from yellow-bellied sapsucker (*Sphyrapicus varius*) wells. This research examines the extent to which ruby-throated hummingbirds (*Archilochus colubris*), red squirrels (*Tamiasciurus Hudsonicus*), and sapsuckers utilize birch sap trees. Intraspecific comparisons of male, female, and juvenile sapsuckers and interspecific comparisons of durations on the sap trees were made to see how much time these animals spend on the trees. Data were collected from early July through mid-August of 1995 in Itasca State Park, MN. Durations on the sap trees by male, female, and juvenile sapsuckers varied considerably, and juveniles spent the longest intervals of the sapsucker classes at the trees. At two of the three sites, squirrels spent the most time at the sapsucker wells. Hummingbirds made the most visits at all three sites, but spent less total time on the trees than sapsuckers or squirrels. Yellow-bellied sapsucker activity, therefore, appears to affect the availability of this food source to these species.

SYNCHRONIZING GOLDFISH REPRODUCTIVE BEHAVIOR: EFFECTS OF HORMONAL PROSTAGLANDIN ON FEMALES AND DETECTION OF PHEROMONAL PROSTAGLANDIN BY MALES

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In goldfish (*Carassius auratus*), it is believed that blood-borne prostaglandin (PGF_{2α}) elicits females' reproductive behavior, while water-borne PGF_{2α} triggers males' reproductive behavior. I sought to determine: 1) if the

concentration of PGF_{2α} in female plasma corresponded with the activity level of reproductive behavior and 2) if blocking either olfaction or vision affected the ability of males to court a PGF_{2α} releasing female by hindering their ability to find, follow, or nudge her.

In both the presence and absence of males, the level of females' reproductive behavior increased rapidly from 0-45 minutes after PGF_{2α} injection, paralleling the rise of plasma PGF_{2α} levels, but the decline in behavior after 45 minutes was slower than the decline in plasma PGF_{2α}. Behavior levels over time more closely matched the levels of an unidentified PGF_{2α} metabolite. For the second study, blocking vision reduced males' ability to find and follow females more than blocking olfaction. However, nudging behavior was greatly reduced when olfaction was blocked, but was minimally influenced by absence of vision. Unilaterally anosmic males performed equal to those with full olfaction. This suggests that spawning is a complex social behavior requiring more than one sense.

THE EFFECTS OF SHADE, SOIL NITROGEN, AND HABITAT TYPE ON LEAF HERBIVORY BY INSECTS IN TWO OAK SPECIES

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Previous studies indicate that plants under environmental stress will experience less herbivory because they put more energy into protecting their leaf tissue. A study was conducted to test these findings in *Quercus ellipsoidalis* and *Q. macrocarpa*. Herbivory levels in both species were compared in seedlings planted in different light and soil nitrogen conditions. These results were compared with herbivory levels in natural conditions. While there were many discrepancies between these two study groups, differences were found in plants undergoing light and nutrient stresses, as well as differences in herbivory levels between the two species. Herbivory levels in *Q. macrocarpa* increased with added nitrogen, while herbivory levels in *Q. ellipsoidalis* decreased with added nitrogen. Also, seedlings in light areas were eaten more than those found in the shade. We also compared herbivory levels between the two species in different habitat areas. Data collected from seedlings in three oak savanna sites and three old field sites showed that *Q. ellipsoidalis* seedlings were eaten more than *Q. macrocarpa* seedlings.

POSTCOPULATORY/PREZYGOTIC ISOLATING MECHANISM BETWEEN *DROSOPHILA SIMULANS* AND THEIR SIBLING SPECIES, *DROSOPHILA MAURITIANA*

Jasper M. Simon, Macalester College, 1600 Grand Ave., St. Paul, MN 55105

Studies suggest the existence of postmating but prezygotic reproductive isolation between sibling species *D. mauritiana* and *D. simulans*. Preliminary data on double-mated *D. simulans* females suggest that when males of the conspecific species are mated second, their semen causes a reduction in heterospecific sperm from the first mating. Data also suggest "resistance," or first-male-advantage, when the first mating was conspecific. The effect on the existent sperm was determined from the proportions of hybrid to *D. simulans* male progeny after the second mating. Two predominant ideas seem plausible: 1) conspecific sperm have evolved some adaptive advantage over heterospecific sperm, or 2) some physiological mechanism in the female has evolved which can differentiate between con- and heterospecific sperm, and thus 'chooses' the conspecific species' sperm. A number of additional mechanisms could also explain this postmating but prezygotic reproductive isolation.

HEAVY METALS IN BLUE-GREEN ALGAE AS FOUND IN ST. PAUL'S WATER SUPPLY: A SCANNING ELECTRON MICROSCOPE (SEM)/X-RAY MICROANALYSIS STUDY

James M. Hafner, Jr., University of Minnesota, College of Natural Resources, 2003 Upper Buford Circle, St. Paul, MN 55108-6146

For abstract, see Winchell Papers.

THE EFFECTS OF VARYING PRESSURE ON THE GROWTH OF A METHANOGEN, *METHANOBACTERIUM WOLFEEI*

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For abstract, see Winchell Papers.

SUCCESSIONAL DEVELOPMENT OF WETLANDS: A YEAR STUDY

Alycia Klunenber, Hamline University, 1536 Hewitt Ave., St. Paul, MN 55104

For abstract, see Winchell Papers.

THE ABUNDANCE AND DISTRIBUTION OF *TRIDACNA GROCEA* and *T. MAXIMA* ON FIVE PATCH REEFS IN PALAU

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For abstract, see Winchell Papers.

MATH

CREATING ENVIRONMENT ADAPTED STRUCTURES USING GENETIC ALGORITHMS (CEASGA)

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Genetic Algorithms are search algorithms that utilize concepts of evolutionary biology. CEASGA intends to show the usefulness of Genetic Algorithms in finding structures that are adapted to a simulated environment without giving the algorithm more than the general survival criteria of the environment. My specific goal was to "evolve" two-dimensional "plant-like" structures to fit a simulated environment.

I set up a simple world consisting of simulated water, soil and sun supply. I defined the survival criteria for a "plant-like" structure to be 1) connection to water in the soil, 2) exposure to sun rays, 3) a bonus for static coherence. The algorithm had a limited number of cells from which to build a structure well adapted to the simulated environment.

Two test cases were run, in which 50 structures were evolved for 1 00 generations. The structures grew towards the "sunlight", developed "leaves" (horizontal planes) and displayed visual similarities to "real world" plants. In test case II 2/3 of the simulated world was left in the shade. The evolved structures tended to grow out of the shaded area. From the limited amount of information given in the test world, the GA developed complex structures which seemed to display functionality that was advantageous in the given environment

MEDICAL SCIENCE/PHYSIOLOGY

OPIOID INDUCED HOMOTYPIC ADHESIONS IN HUMAN LYMPHOCYTES

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Cell-to-cell adhesion plays a pivotal role in physiological and pathological processes. These adhesions are regulated by chemical signals that are instrumental in the growth and metastasis of tumor cells, necessary for normal development, required for wound healing and essential to the immune response. Opioid peptides, an important class of neurohormones which were initially described for their analgesic activity, have now been shown to modulate other physiologic functions. These agents participate in the regulation of autonomous rhythms, reproduction, learning and memory and the immune response. In this study we show that DADLE (D-alanine-D-leucine-enkephalin), a synthetic opioid peptide, stimulates homotypic (cell-to-cell) adhesions of cultured human lymphocytes. We have incubated NALM 6 cells (human pre-B acute lymphoblastic leukemic cells) and Jurkat cells (human T leukemic cells) with and without DADLE. Following fixation and staining, the cells were visually scored with the aid of a light microscope. Our results show that when these cells were treated with DADLE, they formed homotypic aggregations. Our findings point to a role for opioid peptides as signals for the induction of B and T lymphocyte adhesion.

NA-H ANTIPORT ACTIVITY IN NEONATAL VENTRICULAR MUSCLE CELLS

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The intracellular pH (pH_i) in ventricular muscle cells isolated from 2 to 5 day old rat pups was measured using the pH-sensitive fluorescent dye 2',7'-bis(2-carboxyethyl)-5(6)-carboxyfluorescein (BCECF). The resting pH_i for neonatal heart cells two to three days following culture ranged from 7.0 to 7.2. The ammonium chloride prepulse technique was used to acidify pH_i . Following acidification, pH_i recovered to resting levels in a sodium-dependent, amiloride-sensitive manner which confirmed the presence of the Na-H antiporter in these cells. Two different agonists, phenylephrine and adenosine triphosphate(ATP), were tested to determine their ability to alter pH_i through activation of the Na-H antiporter. Phenylephrine and ATP each stimulated an increase in pH_i of 0.11 to 0.15 units. In both cases, the addition of amiloride blocked the agonist induced increase in pH_i indicating that Na-H antiport activation was responsible for the alkalization.

THE INFLUENCE OF MELATONIN ON REGENERATION IN THE LIMBS OF THE FIDDLER CRAB *UCA PUGILATOR*

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Melatonin is a hormone produced by the vertebrate pineal gland and possibly by photoreceptive structures of invertebrates. High melatonin levels have been found in invertebrate optic, brain, hemolymph and other tissues. Melatonin's influence on circadian rhythms and reproductive cycles in vertebrates have been well-established; its effects in invertebrates, however, have not been extensively studied, though melatonin has been detected in all invertebrates examined. Crabs were separated into six groups with eyestalks either removed or intact, and in sea water with no melatonin, 0.06 mg ml⁻¹ or 0.03 mg ml⁻¹ melatonin. The third

right leg of each crab was autotomized and limb bud regeneration measurements were taken for 17 days. Results show a significantly faster limb bud regeneration rate in those crabs exposed to melatonin, and the influence was dose-dependent. Mid-photophase MEL levels were measured in the eyestalks of fiddler crabs with RIA; the mean MEL concentration was 128.7 ± 22.3 pg per pair of eyestalks, or 43.2 ± 5.9 pg per mg of exoskeleton-free tissue.

ACTIVATION OF THE PITUITARY-ADRENAL AXIS DURING FASTING IN RATS

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For abstract, see Winchell Papers.

CIRCADIAN RHYTHM OF MELATONIN IN THE EYESTALKS OF THE FIDDLER CRAB *UCA PUGILATOR*

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For abstract, see Winchell Papers.

MICROBIOLOGY

SYNERGISTIC EFFECT OF SERA AND ANTIBIOTICS AGAINST *HELICOBACTER PYLORI* ON HEP2 CELLS IN VITRO

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The link between peptic/duodenal ulcer disease and the presence of *Helicobacter pylori* in the digestive system has become the center of much debate and study. Many drugs such as tetracycline, cephalothin, metronidazole, and bismuth salts are capable of suppressing *H. pylori* infection during drug therapy when used as single-agent treatment in patients with peptic ulcer. However, a single drug therapy generally has not resulted in true eradication of the organism. Using these drugs in combination with each other and in combination with a variety of animal sera, a more effective treatment for *H. pylori* infection may be found. Fetal bovine, chicken, goat, guinea pig, horse and sheep sera have been chosen to determine if they will have a synergistic effect with the drugs already used to treat *H. pylori* HEP2 cells were as an indicator for determining the results. These cells were grown in minimum essential medium (MEM) supplemented with 7% fetal calf serum and 1% pen/strep at 37°C with 5% carbon dioxide tension. The initial test was performed to determine if *H. pylori* could survive in HEP2 cells. Preliminary tests have shown a traditional triple drug therapy, metronidazole, cephalothin, and tetracycline, prevented the growth of *H. pylori* resulting in the protection of the HEP2 cells. More work is in progress to include an additional number of sera with currently used antibiotics for therapy.

IN VITRO SYNERGISTIC EFFECTS OF SIX PLANT EXTRACTS ON HSV-1 AND HSV-2 IN HEP2 CELLS

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Six plant extracts namely *Artemesia absinthium* (Absinthe), *Resin pistacia lenticus* (mastic), *Rosa centifolia* (roses), *Flores onosma brateatum* (borage), *Delphenum denodatum* (jadwar), and *Elletaria cardamomum* (cardomom) were used to attempt to inhibit Herpes Simplex virus type 1 and type 2 using human epithelial cells type 2 (HEP-2) of the larynx as indicator cells. Cytotoxicity of the plant extracts was measured by growing HEP-2 cells in Minimal Essential Media (MEM) supplemented with 5% fetal calf serum, 1% pen/strep,

and serial two-fold dilution of all combinations of plant extracts. Anti-HSV activity was tested by adding 0.5 ml of the highest dilution of extract displaying cytotoxic effects and 0.5 ml of MEM to each well containing a cell monolayer that was previously overlaid with 0.1 ml of MEM containing 5 virus particles. The infected cell monolayers were incubated at 37 degrees Celsius and 5% carbon dioxide tension and were observed for 48 hours to determine cytopathic and cytotoxic effects. None of the plant extracts showed anti-HSV activity alone. However of the 57 tested combinations, 11 showed protection against HSV- 1 and 4 showed protection against HSV-2. One extract combination (3Ab:2Ro:1Ja) was effective in protecting the HEP-2 cells from both HSV-1 and HSV-2. Thin Layer Chromatography (TLC) and protein assays have determined that no lipid and a negligible amount of protein are present in these plant extracts. The protein pattern of HSV-2 infected cultures treated with 3Ab:2Ro:1Ja were different than untreated infected cells and treated control cells when analyzed by SDS-PAGE.

IDENTIFICATION OF GENE(S) RESPONSIBLE FOR REGULATION OF GENE EXPRESSION IN REOVIRUS INFECTED CELLS.

Jay - Alphonse Q Santos, University of Minnesota, Department of Microbiology Box 196, 1460 Mayo Memorial Building, 420 Delaware St. S.E, Minneapolis MN 55454

In this study, the gene(s) responsible for the regulation of gene expression in reovirus infected cells will be identified. Findings in the literature (S. Munemitsu and C.E Samuel, Virology 136: 133-143) observe a polymorphism between reoviral strains T1L and T3D When incubated at 30°C, T1L infected mouse L-cells exhibited late gene expression relative to T3D infected mouse L-cells Confirmation of these findings, and an assay in which the polymorphism is maximally demonstrated will be done through a time course assay in which cell extracts of infected mouse L-cells are harvested, immunoprecipitated with an equivalently binding monoclonal antibody 10H2, and analyzed via SDS-PAGE. Subsequently, T1L x T3D reassortants and parental strains will be analyzed in the time course assay established, with respect to the difference in gene expression By comparing the phenotype and genotype of the T1L x T3D reassortants with the parentals as controls, the gene(s) in question will be identified.

PHYSICS

STATIC DIELECTRIC CONSTANT OF SINGLE-CRYSTAL $YBa_2Cu_3O_6$

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The high-temperature superconductor $YBa_2Cu_3O_7$ may be easily converted to the insulator $YBa_2Cu_3O_6$ by an appropriate heat treatment. $YBa_2Cu_3O_6$ is interesting in its own right: the static dielectric constant of polycrystalline samples has been reported to be large, approximately 700 at room temperature.¹ We have undertaken an investigation of the static dielectric constant of single crystals of $YBa_2Cu_3O_6$. Samples were obtained by heat treating single crystals of $YBa_2Cu_3O_7$ in flowing argon at 600°C for a week. X-ray diffraction measurements and magnetization measurements were made on the crystals before and after the heat treatment to confirm that the conversion took place. We will report on preliminary measurements of the static dielectric constant by a capacitance method.

¹L. R. Testardi *et. al.*, Phys. Rev. B 37, 2324 (1988).

POINT-SPREAD FUNCTION PHOTOMETRY

Frank J. Deglman, Department of Physics, Astronomy, and Engineering Science, St. Cloud State University, St. Cloud, MN

My poster will demonstrate the merits of employing point-spread function photometry over more conventional aperture photometry in crowded-field CCD stellar photometric applications. Stellar photometry is essentially the measuring of light output (at various wavelengths) of stars. By monitoring certain characteristics of this intensity, much can be determined about the mechanics of these stars. Imaging with a CCD camera, the data are displayed in a rectangular array of values which represent stellar intensities, i.e., a digitized picture of the sky. Having a computerized array of values instead of single a data point, both aperture photometry and point-spread function photometry can be done on a given image. It has been our experience at SCSU that aperture photometry is ineffective with globular cluster images. These cluster images is where point-spread function photometry shows the most potential. It is a powerful tool for determining intensity of given stars under extremely cramped conditions. The PSF photometry method assumes that the intensity profile of each stellar image will have the same basic shape, usually that of a gaussian function. Once this shape is determined mathematically, it is then fitted, via a scaling factor, to every star in the image and their respective intensities are determined.

STABILITY ANALYSIS OF A SIMPLE NONLINEAR OSCILLATOR

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For abstract, see Winchell Papers.

PLANT SCIENCE

THE EFFECTS OF MELATONIN ON ROOT GROWTH IN ARABIDOPSIS THALIANA

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Melatonin is a hormone which plays an important role in the circadian rhythms in animals. It has also been found in relatively high amounts in several plant species. However, the role of melatonin in plants is unknown at this time. In a preliminary study, *Arabidopsis thaliana* seedlings were grown in the presence of a variety of concentrations of exogenous melatonin. Concentrations were selected to bracket physiological levels of melatonin. Several changes in growth habit were noted. The primary roots appeared to grow straighter and thicker than those of control plants (no melatonin added to the medium). Lateral root growth was partially inhibited which was noted as fewer and shorter lateral roots. Further experimentation is in progress to verify and expand these findings.

MELATONIN LEVELS IN DIFFERENT PLANT TISSUES, AGE GROUPS, AND LIGHT/DARK EXPOSURES QUANTIFIED BY RADIOIMMUNOASSAY IN ARABIDOPSIS THALIANA, ZEA MAYS, AND RAPHANUS SATIVUS

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Melatonin is a molecule produced in members of both the animal and plant kingdoms. Studies have shown that production of melatonin in animals decreases with age and fluctuates with photoperiod: the presence and function of melatonin in the plant kingdom has not been well documented. We attempted to find relationships between melatonin levels and plant tissues, age groups and light/dark exposures. Melatonin was detected and quantified in three

species of plants investigated using a tritium (^3H) radioimmunoassay procedure. *Raphanus sativus* tissues were assayed for melatonin levels: relatively high levels were found in stems (105,862 pg melatonin/g tissue) and leaves (11,965 pg/g). Lower melatonin levels were found in roots (463 pg/g), cotyledons (101 pg/g), and seed coats (6419 pg/g). *Arabidopsis thaliana* was found to have higher melatonin levels at 16 days age (8014 pg/g) than at 7 days (1526 pg/g). Light exposure affected melatonin levels in *Zea mays*: plants grown in 24 hours of light and subsequently exposed to 6 hours of dark had elevated melatonin levels (460 pg/g) as compared to a control group (204 pg/g) and etiolated seedlings (204 pg/g). Further experimentation and replication is in progress by the authors.

PSYCHOLOGY

JANA SHEEDY, MANKATO STATE UNIVERSITY

This backwards masking experiment explored the relationship between the size of a target stimulus and the size of the masking stimulus. The target stimulus was a solid circle 20 mm or 45 mm in diameter. The mask was an open circle 60 mm for the first group and 90 mm for the second group. The mask was presented to the subjects either, 0 msec, 13msec, 26 msec, 39 msec, 52 msec, and 65 msec. The subjects were to state "yes" or "no" to whether they saw the solid circle or not. The results showed that the size of the target stimulus was significant in effects of backward masking. The subjects reported seeing the larger solid circle sooner than the smaller solid circle. This would conclude that metacontrast or backward masking occurs more often in stimuli presented below 60 to 90 mm in size.

WORD METHOD DIRECTED FORGETTING: A TEST OF CREDIBILITY OF RECOVERED REPRESSED MEMORIES

Dawn Mears, Mankato State University, Department of Psychology

The credibility of repressed memories is tested with a combined hypermnnesia and directed forgetting experiment. Subjects viewed a list of 44 words with instructions to rate each word as it appears on the screen, and to remember the words which are followed by the "remember" command, and forget those words followed by a "forget" command. Following the word presentation, subjects will write down as many of both the words followed by a "remember" command as well as any words followed by the "forget" command. Subjects will be instructed to recall words of both types a further two times. The number of both types of words recalled should increase across tests. However, it is expected that as recall increases, subjects may miss identify words, and report words followed by a "remember" command as being followed at study by a "forget" command, and vice versa for the "forget" words. An explanation for these errors is provided.

TEACHING SCIENCE

A CORRELATIONAL ANALYSIS OF ANTISOCIAL BEHAVIOR PORTRAYED ON TELEVISION AND ANTISOCIAL BEHAVIOR OF ADOLESCENTS

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It has been suggested by social learning theorist that observing antisocial behaviors as portrayed on television is

positively correlated with antisocial behavior of viewers (Messner, 1986). In the above titled study, 100 adolescents (50 males, 50 females) from South Washington County School District will be completing a comprehensive questionnaire assessing their television viewing habits and behavioral patterns. The participants will be asked to review a television schedule and to highlight those programs they had viewed the week prior. The selected television programs will be given quantifiable scores for their portrayal of antisocial behavior. The scores for each program will be derived from the content analyses performed by the National Coalition on Television Violence and as reported by the Surgeon General's Advisory Commission on Television and Behavior. The participants will also be providing information on their patterns of conduct. They will be asked to review a list of 55 antisocial behaviors and indicate which of the behaviors apply to their personal conduct. The list of behaviors ranges from minor social infractions to severe criminal conduct. The data generated from this portion of the study will be correlated with the television viewing composite scores. Depending on the outcome, this research may demonstrate a positive correlation between viewing antisocial behavior and antisocial conduct of those viewers.

NEURAL CORRELATES OF INFANT MEMORY DEVELOPMENT: A COMPARISON STUDY OF NICU GRADUATES AND FULL-TERM HEALTHY INFANTS

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For abstract, see Winchell Papers.

ELECTROPHYSIOLOGICAL INDICES OF CATEGORIZATION: DIFFERENTIATION BETWEEN EMOTIONAL AND NON-EMOTIONAL CATEGORIZATION TASKS

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For abstract, see Winchell Papers.

EFFECT OF PEER MEDIATION IN RESOLVING CONFLICT BETWEEN COLLEGE STUDENTS

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For abstract, see Winchell Papers.