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ABSTRACTS

Abstracts are listed alphabetically by the last name of the first author listed.

ALTERNATIVE PEDAGOGIES IN INTRODUCTORY SCIENCE COURSES SYMPOSIUM

ORGANIZING COURSE CONTENT AROUND THEMES IN AN INTRODUCTORY NON-MAJORS BIOLOGY COURSE

Susan Chaplin
Univ. of St. Thomas, St. Paul, MN

Introductory courses in biology can be overwhelming for non-majors in the sciences because they survey vast amounts of information and may not connect that information to students' personal experiences and knowledge base. As a result, students complete the course feeling that they have memorized a lot of perhaps interesting, but somewhat useless, information. Theme-based courses approach learning in a completely different manner than survey-based courses. In a theme-based course, basic principles of biology are woven into a central topic, which might be a contemporary issue with which students are very familiar (e.g., diabetes, HIV, athletic fitness, cloning, infertility). The theme gives students a context for learning, and the concepts introduced allow students to build on their foundation knowledge, expanding their understanding as the instructor reveals more of the linkages between principles and applications of those principles. The conduct of the course might involve problems, cases, group discussions, or role play, as students progress through the material. The choice of topics can be such that the same basic principles can be introduced in a theme-based course that were included in a survey-type course.

WHY TRY SOMETHING NEW?—PART 1

Susan Chaplin and Mary Walczak
Dept. of Biology, Univ. of St. Thomas, St. Paul, MN

A new paradigm has emerged in college science education that emphasizes an active, learner-centered environment in place of a passive, lecturer-centered classroom. Classroom activities such as case studies, role play, jig saw, peer lead team learning (PLTL), just-in-time-teaching (JiTT), guided inquiry (POGIL), and problem-based learning (PBL) aim to increase engagement of students with the material while enabling them to construct new knowledge. Although there have been many published reports of "what works" among these many alternative pedagogies by some of their innovators, a thorough evaluation of their implementation by early adaptors and of the learning gains of students in this alternative environment has not been done. This session and the following one will focus discussion around the use, implementation, and assessment of alternative pedagogies in a variety of disciplines and courses.

WHY TRY SOMETHING NEW?—PART 2

Susan Chaplin and Mary Walczak
Dept. of Biology, Univ. of St. Thomas, St. Paul, MN

A continuation of the discussion from "Why try something new?—Part 1" on the use, implementation, and assessment of alternative pedagogies in a variety of disciplines and courses.

DOES THE USE OF ACTIVE LEARNING TECHNIQUES IN CHEMISTRY REALLY HELP STUDENTS LEARN?

Lynn Hartshorn
Univ. of St. Thomas, St. Paul, MN

The author will provide a description of active learning techniques used in general chemistry classes of various sizes. The technique described begins with hands-on work done by students or student observations of experiments or demonstrations given by the instructor. This is followed by student group work culminating in a worksheet that is handed in for grading. Some evidence is presented showing how the active learning influences student learning, as shown by responses in tests.

REFORMING THE INTRODUCTORY PHYSICS COURSE FOR BIOLOGY MAJORS

Leon Hsu and Ken Heller
General College, Univ. of MN, Minneapolis, MN

The biological sciences are changing dramatically and the education of the majors in those fields must change as well. At the University of Minnesota School of Physics and Astronomy, we are trying to meet this challenge by examining the utility of the introductory physics course for biological science majors and premedical students. The first step in this process is choosing a small set of achievable goals for the course. Since virtually all students who take this course are required to do so by faculty in their disciplines, the chosen goals should reflect the needs of those disciplines. In this talk, we will discuss one of the primary tools that enables us determine a set of goals for the course, a questionnaire for faculty in other departments, a brief discussion of the results, and our first attempts to restructure the course to meet the desired goals.

BUILDING A LEARNING COMMUNITY IN THE CHEMISTRY CLASSROOM

Anna McKenna and Kate Graham
College of Saint Benedict/St. John's Univ., Collegeville, MN

Much of the recent research about science pedagogy emphasizes that underrepresented groups in science, such as minorities and women, benefit from a

classroom atmosphere that encourages community. Much of this "feminist pedagogy" was developed to empower women in non-traditional fields and to encourage equal participation by women. However, many of these strategies are beneficial to all students and are becoming incorporated into commonly used good teaching practices in science.

In this presentation, we will review the literature on community-building approaches to teaching science. We will also present some of our findings on several of these approaches, including Peer Led Team Learning, a nationally recognized program in science and mathematics.

A NOVEL COURSE FOR IMPROVING PERFORMANCE OF AT-RISK STUDENTS IN GENERAL CHEMISTRY

Jack F. McKenna and Tamara Leenay
Dept. of Chemistry, St. Cloud State Univ., St. Cloud, MN

After several years of requiring students to take Preparatory Chemistry (CHEM 140) when scoring poorly on the departmental placement exam for General Chemistry 1 (CHEM 210), an alternative course sequence was developed. In 2001, a pilot program called Essential Skills for Chemistry (CHEM 110) was developed. Borderline performance on the placement test allowed students to enroll concurrently in both CHEM 110 and CHEM 210. The benefits of CHEM 110 are twofold: students are not set back a semester in their general chemistry requirement, and problem-solving skills are addressed to help students succeed in CHEM 210. CHEM 110 is a one-credit (pass/fail) intensive review course that meets three times a week for the first five weeks of the semester. This presentation will describe the contents of CHEM 110, use of the placement test to determine course placement, and how CHEM 110 prepares students for CHEM 210.

ACTIVE LEARNING IN THE INTRODUCTORY PHYSICS COURSE

Thomas Tommet
Univ. of St. Thomas, St. Paul, MN

I have introduced a number of active learning teaching techniques into the introductory physics course. I will describe some of these, and discuss the pros and cons that I perceive in their use.

USING CASE TEACHING TO INTRODUCE NUCLEAR CHEMISTRY IN THE GENERAL CHEMISTRY CURRICULUM

Wayne Wolsey and Graham Peaslee
Macalester College, St. Paul, MN, Hope College

The use of case studies for introducing content material may be of aid to the instructor who does not have a solid background in that area. One such area is nuclear chemistry, frequently a topic at the end of the text which is often not covered.

The use of a Radon case, to which the students can identify via the story line, makes this an easy entree into nuclear science. In order to properly discuss the facets of radon, it is necessary to have a basic understanding of nuclear decay processes and nuclear energetics.

Usage of a case study should culminate by forcing some kind of decision, such as where the student should live or what to eat.

Two other nuclear-related topics will be presented which have relevance to student lives.

Business and Economics Symposium

ETHNIC-BASED CREDIT UNIONS IN THE UNITED STATES

M. Anaam Hashmi
MN State Univ., Mankato, MN

According to the National Credit Union Administration's Federal Credit Union Handbook, a credit union is a financial institution which, like a bank, permits groups of persons to save, borrow, and obtain financial services, but unlike a bank, which operates in the open market, allows those same persons to participate in its management. A credit union is a not-for-profit cooperative that is member-owned and member-controlled.

Most of the credit unions still remain very small compared with other types of depository institutions, but worldwide membership in credit unions has not stopped its upward climb. Around the globe, nearly 90 million people belong to one or more of about 36,000 credit unions scattered among nearly 90 nations. In 1996 there were 36,244 credit unions belonging to the World Council of Credit Unions with a worldwide membership of 89,685,210 and total assets (in U.S. dollars) adding up to $379.3 billion.

As long as there have been immigrants in the United States, there have been immigrant banks. East European Jewish immigrants in New York, Chicago, Boston, even Altoona, Pennsylvania, established Hebrew loan societies in the late nineteenth and early twentieth centuries to finance their early business ventures. Japanese and Chinese immigrants did something similar. And Bank of America, today the nation's third-largest bank, began in San Francisco in 1904 as the Bank of Italy.

This is an exploratory study of the ethnic-based credit unions in the United States. A number of secondary and primary sources dealing with credit unions were studied. Total number of credit unions in the United States where membership is based on an ethnic group was located on the National Credit Union Association (NCUA) website. Due to lack of availability of the complete data on ethnic-based credit unions, the author decided to look into about all the ten thousand credit unions, federal and state charters, state by state, and find

the ethnic-based credit unions by name. The author is in the process of tabulating all ethnic-based credit unions in the United States and their impact on each ethnic community.

A DECADE OF TRANSITION: LONGITUDINAL EVIDENCE ON RUSSIAN STUDENTS' ATTITUDES TOWARD MARKETS, 1991-2003

Michael Hemesath, Yelena V. Morozova, and Irina V. Samarkina
Carleton College, Northfield, MN, Kuban State Univ., Krasnodar, Russia

A survey, originally developed and used by Shiller, Boycko, and Korobov, is used to compare Russian undergraduates' attitudes toward markets at the beginning of the economic transition and approximately a decade into the reforms. Though some attitudes appeared to change somewhat over the two-year period from 1991 to 1993, the changes were generally in favor of a transition to the market.

As economic performance improved unevenly in the decade from 1993 to 2003, students became more confident about the permanence of the transition but became more ambivalent about the fairness of market outcomes and appeared to be more supportive of government interventions in the market economy. The past decade certainly did not cause Russian students to become enthusiastic free marketers, but they do seem to somewhat grudgingly accept a market-based economic system.

DOLLARIZATION, THE IMF, AND THE INDEBTEDNESS OF DEVELOPING NATIONS

Tauiilelagi Langkilde
College of St. Catherine, St. Paul, MN

With the onset of globalization, the appeal of dollarization has settled into the agendas of a number of developing countries as a means to suppress the difficulties of a volatile national currency, as well as to alleviate the hardships of foreign debt repayments, among other things. Throughout the course of history, however, the implementation of a dollarization policy by a developing country as a measure to espouse global economic integration, as well as to smooth out a developing country's balance-of-payments scheme, has not only hindered economic growth, apparently, but has also been linked to Third World poverty and international indebtedness for developing nations, namely Ecuador. How is this so? What drives countries to adopt a foreign currency? Is dollarization to blame? What are the social, economic, and political implications of following such policies? My research paper will cover three areas, which include a cost-benefit analysis of dollarization in regard to other currency stabilization devices, Third World indebtedness and poverty spurned by economic development policies, and last, the role of the International Monetary Fund in the implementation and facilitation of the first two areas.

What I expected to uncover was the workings of dollarization as a cause for Third World indebtedness. Based on my initial perspective of dollarization, which was largely influenced by my time spent in Ecuador, I perceived dollarization as a major hindrance to economic growth and stability relative to the livelihood of its people. It seemed that the inflation rate associated with the dollar was so much that daily wages could be categorized as below extreme poverty. Based on my initial perception of dollarization, I expected to find the results of dollarization as the cause of balance-of-payments deficits.

During the progression of my research, the concept of dollarization was clarified, and thus, my perception of its purpose was changed. Readings and studies have come to show that by adopting a foreign currency, namely the currency of a major trading partner, highly indebted countries would be able to alleviate balance-of-payment deficits in the long run. If such a measure is implemented correctly with the corresponding domestic reforms (labor market, financial sector, public financing, and employment), dollarization may prove to be successful in bringing highly indebted developing countries out of their foreign indebtedness. I also found that dollarization is NOT the cause of Third World indebtedness. Rather, it is through accumulation of foreign debt by developing countries, and namely through IMF loans, that Third World indebtedness persists. IMF structural policies and austerity programs tend to erode social development affiliated with economic growth through the slashing of funds for government programs, such as health care, that would have benefited the social interest. Also, it is through such programs that developing nations may tend to lose their financial sovereignty and independence. Unlike dollarization, where governments enjoy the liberty of decisions regarding tax revenues, tariffs, subsidies, and other trade policies, IMF policies are underwritten with a literal "appropriation" of a country's financial sector, such as that of in Korea post-1997. Therefore, dollarization affords some sort of sovereignty rather than eliminating those that are beneficial to a country.

In the end, my perception of dollarization has changed, and my ability to find the interconnectedness of all three areas has helped me to broaden my understanding of different international economies, as well as to learn a thing or two about exchange rate regimes and their implications in currency stabilization. I also was able to see the direction in which we need to go in terms of international lending, which is one that is less political and more in tune with social responsibilities.

A STRATEGY FOR COPING WITH SUPPRESSED VALUES IN RETAIL CENSUS SALES DATA

Lorman Lundsten, Dave Brennan, and Kevin Knutson
Marketing Dept., Univ. of St. Thomas, St. Paul, MN

Retail sales data provided by the U.S. Census Bureau in the Census of Retail Trade are increasingly incomplete because of the disclosure rule prohibiting the

release of data that would reveal the sales for individual retail establishments. For example, suppressed sales values at the county level in the three-state area of Iowa, Minnesota, and Wisconsin grew from 1.9% in 1948 to 28.9% in 1997. Only 33 of the 258 counties had complete data in 1997. Suppression is most prevalent for sparsely populated counties. Eighteen counties had over half their sales categories suppressed. The problem is most acute for the miscellaneous and general merchandise retail categories. These categories had 60% or more of their sales values suppressed. The increased incidence of suppressed data is largely the result of the rapid decline in the number of stores, the rapid increase in the number of "big box" retailers (especially general merchandise discount stores), and the census's increase in sales categories from 10 to 12 in 1997.

The Exploratory Data Analysis technique median polish was used to estimate the value of sales in suppressed categories. A random sample of 204 category values was withheld from the analysis and used to estimate the predictive value of the model. Median polish produces a robust, additive model that estimates sales in a category as a function of the general importance of the category and the importance of the county over all retail categories. Since total retail sales for the county is available even when some of the categories are suppressed, this can be used to force the estimates to sum to this total. The technique is generally applicable for all states and could be extended to deal with data suppression in other census data.

FAMILY MIGRATION DECISIONS: A HISTORICAL PERSPECTIVE

Charles A. Rambeck
St. John's Univ., Collegeville, MN

Migration is part of the labor market adjustment process in a geographically disperse population. The traditional human capital view of the migration decision emphasizes both the costs and benefits of moving. As most potential movers are members of multi-person households, the structure and demographic characteristics of those households are likely to affect both the cost and the benefits of relocation. For example, families with two or more market workers face more complex choices than households where only one market wage is affected by moving. The fact that two workers are affected by one move means that households where both spouses do market work are less likely to move than households with only one worker in the labor force. Therefore, structure and demographic forces of family structure can be expected to affect mobility decisions.

This paper uses a valuable new data source to examine how these structural and demographic influences might have changed since the 1930's. The *Integrated Public Use Microdata Series* (IPUMS) from the University of Minnesota makes it possible to examine migration rates for various types of households over a comparatively long time period. The IPUMS data provide individual records from every decennial census for which migration information was collected.

One significant finding of this paper is that the effect of multiple job holders on family mobility is quite different before WW II from its effect after the War.

WORDS, STORIES, AND ECONOMIC EDUCATION

Steven D. Soderlind
Dept. of Economics, St. Olaf College, Northfield, MN

This paper is dedicated to two favorite teachers, Walter Heller and Erik Brofoss, both of whom emphasized the playful use of words and stories in teaching economics. Not to belabor issues of language and knowledge, the presentation will focus on harnessing the power of words to enliven the teaching and discernment of economics.

Addenda: Heller, Regents Professor of Economics at the University of Minnesota and Chair of the Council of Economic Advisors under Presidents Kennedy and Johnson (1961-1964), taught brilliantly in classrooms, boardrooms, and halls of government. Brofoss (cand. econ, JD) served as Norway's Minister of Finance (1945-1947), Minister of Commerce (1947-1957), and Central Bank Director (1957-1970) before leading the International Monetary Fund (1970-1973). Over distinguished careers, both of these men learned to overcome barriers in understanding and advocacy with the persistent cultivation of economic terms–demonstrating that effective teaching necessarily involves helping others to expand their vocabularies and repertoires.

GEOGRAPHY SYMPOSIUM

CLIMATE AND ICE-OUT DATE: ANALYSIS OF THE MINNESOTA ICE-OUT RECORDS DATABASE

Virginia M. Card
Dept. of Natural Science, Metropolitan State Univ., St. Paul, MN

The timing of the loss of ice cover from lakes in the spring (i.e. ice-out) is a sensitive indicator of late winter temperatures. Globally, average ice-out dates are about 7 days earlier than they were 100 years ago (Magnuson et al. 2000). The Minnesota Ice-out Records Database held by Metropolitan State University contains over 104 records from 100 lakes across the state, including 5 records starting before 1900 (Minnetonka, Osakis, Winsted, Detroit, and Clear), 3 records starting between 1900 and 1925 (Minnewaska, Vermillion, and Fountain), and 11 records starting between 1926 and 1950 (Diamond, White Bear, Fall, Sisseton, Leech, Gunflint, Itasca, Waconia, Bone, Lizzie, and Mitawan). Analysis of the records shows very strong correlation within groups of neighboring lakes. Analysis of long records shows that

the trend of changing average ice-cover dates within the state is consistent with the global trend. The trend to increasingly early ice-outs dates has accelerated since the 1960s, with most lakes in the databases experiencing their earliest ice-out date on record in 2000. Statewide, the years of earliest average ice-out dates of the second half of the century occurred in 2000, second earliest in 1987, and third earliest in 1981. Timing of ice-out is strongly correlated with cumulative melting degree days during late winter and early spring, and is therefore important as a climatic indicator that is independent of the instrument record.

THE AGING POPULATION OF MINNESOTA: A SPATIAL ANALYSIS

Ann M. Fossum
Dept. of Geography, Gustavus Adolphus College, St. Peter, MN

The objective of this study is to spatially analyze the 65-and-over population of the state of Minnesota. The data are from census tracts of the 2000 census. Various spatial methods, such as the location quotient, were used to determine which geographic parts of the state have a greater aging population. Regression and correlation analysis were then used to test the hypothesis, namely that there is a relationship between household income levels and percentage of urban/rural population, and percentage of 65-and-over population. More specifically, both lower income and a higher percentage of rural population positively correlate with a higher percentage of 65-and-over population. Preliminary results indicate that: (a) there is an uneven spatial distribution of the elderly population in Minnesota, and (b) the rural areas of the state, as well as inner city Minneapolis and Saint Paul, have a higher proportion of 65-and-over population.

WHERE ARE ALL THE "JESUS FREAKS"? THE GEOGRAPHY OF CONTEMPORARY POPULAR RELIGIOUS MUSIC IN THE UNITED STATES

Brent Hecht
Geography Dept., Macalester College, St. Paul, MN

As Christian-rooted bands such as Evanescence, Jars of Clay and Creed climb the pop music charts, it is clear that popular religious music is a growing force in secular music markets in the United States.

In order to understand the geography of popular religious music in the United States, it is helpful to examine the spatial trends with an eye to both the small scale and the large scale. Popular music of all religions, including that of the minority religions, displays definite trends in small-scale geography. Specifically, the issues of place and placelessness and sacred space repeatedly arise in the study of where popular religious music is sold and played live.

In this paper, I analyze the small-scale geography of religious popular music using primary research in the form of fieldwork and secondary research by means of popular media articles on the religious music phenomenon.

While small-scale geographic trends are identifiable in most religions' contemporary popular music, Christian popular music is the only popular religious music that demonstrates large-scale geographic trends, particularly regionality.

Using a data set of over 1800 concerts, I identify four major regions of disproportionately large concert/person ratios. I then postulate several theories as to what causes such large concentrations of Christian music popularity.

GEOLOGIC INTERPRETATION OF THE LAS VEGAS VALLEY BASED ON INDUSTRY SEISMIC REFLECTION DATA

Aaron C. Hirsch[1] and Catherine M. Snelson[2]
[1]*College of St. Benedict/St. John's Univ., Collegeville, MN;* [2]*Univ. of Nevada, Las Vegas, NV*

Las Vegas Valley, NV, is located in the southern Basin and Range Province where the basin was formed by the Las Vegas Valley Shear Zone as well as by several thrust and normal faulting events that occurred by Cenozoic time. The geology and tectonic setting in the Las Vegas region is poorly understood given the fact that many structures have been covered by the constant growth of the city. National studies of ground motion and amplification of seismic energy placed Nevada third in the list of states having the potential for loss of life and property due to earthquakes. The Las Vegas area has a high potential for strong ground shaking due its thick basin fill and associated amplification. Due to the amplification effects within the valley, moderate nearby quakes or large distant quakes will produce a large amount of damage in the valley. Las Vegas, though not known for its earthquakes, has numerous microquakes and an active seismic history. In a study using HAZUS to predict damage associated with a M6.9 earthquake, the loss would be billions of dollars with thousands of lives lost. Long-term economic loss would be in the several billions of dollars. Recently, several normal faults, which have the potential to produce an M6.5-to-7.0 earthquake, were reclassified as active tectonic faults with Quaternary movement. As a result, there has been increased effort to understand the Las Vegas Valley and to assess its potential for seismic hazards. One such effort included acquiring industry reflection profiles that cross the valley. In the 1980s, north-south and east-west trending reflection lines with intersecting tie points were placed between Frenchman Mountain to the east and Spring Mountains to produce seismic profiles using Vibroseis.

The profiles, which are over 200 km in length and extend down to 5 s in time or approximately 15 km depth, will provide a tie between the surface work that is currently being conducted and the crustal velocity models that are being calculated to produce a seismic hazard potential for the Las Vegas Valley. The quality of the Vibroseis data is very high. We have been able to locate

the basin/bedrock contact as well as several faults that cut the sections, all of which have been mapped on the shot point map. Most of the faults appear to be normal faults listric in character that trend north-south, paralleling the structure of the southern Basin and Range Province. Basin geometry and associated minor folding, which have been truncated by minor faulting, can be seen across the profiles. Some faults can be seen as tectonic in origin while others are merely subsidence faulting from basin settling.

With these new data we will identify structures that could potentially focus energy in the subsurface, adding to our growing knowledge of the basin geometry. In addition, these data will be incorporated into the seismic hazard model that is being developed for the Las Vegas Valley and will provide very detailed geologic information that has not been previously available.

NATURE, NARRATIVES, AND INTEGRATED CONSERVATION AND DEVELOPMENT PROJECTS: A CASE STUDY OF MADAGASCAR'S RANOMAFANA NATIONAL PARK PROJECT

Erika Jermé (Advisor: Bill Moseley)
Geography Dept., Macalester College, St. Paul, MN

Conservation projects to protect biodiversity, particularly in the developing world, have proliferated in the last several decades. In recognition of the need of support from local people for these projects to succeed, a recent trend has been the introduction of Integrated Conservation and Development Projects (ICDPs). Based on the sustainable development discourse, these projects seek to help communities promote economic development that will reduce pressures on the land.

This presentation uses a political ecology approach to critically assess the ICDP model, by means of a case study of Madagascar's Ranomafana National Park Project. Madagascar, well known for its diverse flora and fauna, is considered one of the world's top biodiversity hotspots and is therefore the site of many recent conservation efforts. This study then seeks to critique both the ICDP model and the western conceptualization of nature.

A LOOK AT MINNESOTA FAMILY FOREST OWNERS: IMPLICATIONS FOR RESOURCE SUSTAINABILITY

Earl C. Leatherberry
USDA Forest Service, North Central Research Station, St. Paul, MN

About 4 of every 10 acres of Minnesota's 16.3 million acres of forest are owned by families. In the spring of 2002, mail questionnaires were delivered to 144 Minnesota family ownerships. The response rate was 73 percent. The objective of this paper is to present findings from an initial analysis of select data elements to highlight social factors that may influence sustainable forest management.

Presently, there are an estimated 183,511 family forest ownerships holding 6.7 million acres of forestland in Minnesota. Nearly half of the forestland has been owned for at least 25 years, and 27 percent of ownerships are headed by persons 65 years or older. About half (49 percent) hold fewer than 100 acres. Most own forestland because it is part of home site, for recreation, or as a place to leave to heirs.

Nineteen percent of ownerships have a written management plan, and 35 percent have received advice about forestry. The most common sources were consultants, forestry agencies, and loggers. At least one-third of ownerships have had a timber sale.

An important concern for resource sustainability is the extent to which owners of smaller holdings practice forestry management. A challenge is the effective use of assistance programs, including cost-sharing. Programs with the greatest potential for success should target owners whose objectives are most consistent with sustainable forestry management. In terms of sustainable timber management, incentives should be directed to the larger-tract owners because they are more likely to harvest timber and make improvements to their land.

"COLLATERAL VICTIM: AFRICA"—A POLITICAL ECOLOGICAL STUDY OF THE PROSPECTS OF THE BIOTECHNOLOGICAL REVOLUTION WITHIN AN AFRICAN CONTEXT

Samuel Thomas Ledermann (Advisor: William Moseley)
Geography Dept., Macalester College, St. Paul, MN

The Biotechnological Revolution has been widely heralded in the popular press as the "next Green Revolution." While the latter has largely bypassed the African continent, the former has been heralded by many as an African panacea to end the devastating effects of starvation, malnutrition, and food insecurity. As reflected in the recent trade war at the WTO (World Trade Organization) between the US and the EU over GM (Genetically Modified) crops, this interpretation has been highly disputed on a global scale. It is within this context that the African continent has been struggling to define its own course.

This paper critically analyzes to what extent Africa has become a collateral victim to this trade war and whether or not the Biotechnological Revolution is able to provide the substantive gains advocated by the neo-liberal discourse. Through the analysis of three specific case studies of sub-Saharan countries (Zambia, Kenya and South Africa) on different stages on the development ladder, the author finds varying approaches, reaching from preventive to promotional. Consequently, the author concludes that while African nations have not become a direct collateral victim to the trade war, they should neither focus a majority of their investments on biotechnology. Rather, they should attempt at "harvesting" their newly gained attention and use their agency to direct investment toward more sustainable,

indigenous traditional farming practices, whose long-term costs and benefits are known to a greater extent.

GIS ANALYSIS OF FORECLOSURES IN THE TWIN CITIES

Lindsey Lund, Martin Lacayo-Emery, Ben Mearns, Ari Ofsevit, and Anna Sokol (Advisor: Laura Smith)
Geography Dept., Macalester College, St. Paul, MN

While homeownership rates continue to reach record highs in the U.S., foreclosure rates continue to rise as well. This becomes especially problematic when foreclosures concentrate in specific areas, leading to other social and economic problems within those neighborhoods. Spatial analysis, particularly through GIS, is becoming a useful tool in examining community and urban problems.

Using socioeconomic and housing data, our project aims to assist the Community Affairs department of the Federal Reserve Bank of Minneapolis in exploring the context in which foreclosures take place, and assessing progress toward their goals of fair and equal access to credit and financial services for all populations (especially low-income, minority, and disadvantaged populations). We will present the results of our GIS analysis of foreclosures in the Twin Cities region, and discuss the implications of our findings.

THE GEOGRAPHY OF WOLVES IN MINNESOTA

Michelle Maley
Dept. of Geography, Gustavus Adolphus College, Saint Peter, MN

Factors influencing the density of wolves (*Canis lupis)* in Minnesota were studied using a geographical information system. Wolf density was mapped using the Minnesota Department of Natural Resources winter wolf survey database and compared with human population density, road density, deer density, and land use management intensity. Each of these factors exhibits a spatial pattern that appears to be correlated, either positively or negatively, with wolf density. Thus, the results are consistent with those of other studies. Different methods for calculating road density were compared in terms of accuracy and usability. Further work may be directed toward modifying road density calculation. More accurate road density calculations could help researchers identify the most suitable areas for wolf habitat and develop other conservation strategies.

WHERE ARE THEY NOW? A SPATIAL ANALYSIS OF GUSTAVUS ALUMNI RESIDENCES: 1940-2000

Justin M. Nelson
Dept. of Geography, Gustavus Adolphus College, St. Peter, MN

The objective of this study is to describe the changing residential mobility of college alumni over the past 60 years. It is the general hypothesis of this research that because of improvements in transportation and communication, the residential choices of college graduates will be expanding spatially over time. Alumni residential data were collected from Gustavus Adolphus College, a small private liberal arts college in St. Peter, Minnesota, for the years 1940 through 2000. A variety of cartographic and statistical methods was employed to analyze the data, including Location Quotients and Weighted Mean Centers displayed on ArcView maps. These were then employed to answer the following questions: Were some states more preferred over time than others as residential locations for Gustavus alumni? Did state destinations of alumni grow larger? Were some cities more preferred over time than others as residential locations for Gustavus alumni? The results of this study conclude that rather than spatially expanding residential choices over time, certain states and certain cities have been preferred residences of Gustavus alumni over the last 60 years.

FIRE ON THE LANDSCAPE: A POLITICAL ECOLOGY OF FIRE MANAGEMENT IN OREGON FORESTS

Robert Spurlock (Advisor: Bill Moseley)
Geography Dept., Macalester College, St. Paul, MN

Forest fires have been systematically suppressed throughout the United States for a century. Removing fire from the landscape has damaged the ecology of Oregon's forests. Trees rely on fire for regeneration, disease control, and as a natural means of thinning. But the U.S. Forest Service and other federal land management agencies, driven by corporate interests, use fire suppression propaganda to obtain power and profits. In and around Oregon, this disruption of natural fire regimes has created forest ecosystems with high fuel loads, resulting in a greater occurrence of catastrophic fires. Fire ecology research shows that prescribed burning is the best method for controlling fires, while cultural ecology research shows that historically fire was the primary land management tool for Oregon's inhabitants. The Bush Administration, however, favors fuel removal (i.e., logging) as the preferred method of fire control. This plan ignores the local knowledge of foresters and small-scale loggers, while it benefits national corporate timber companies. It also endangers and marginalizes the local communities and ecosystems that it purports to protect.

No substantial body of work exists within political ecology on the topic of fire and timber production. A political ecology framework is most appropriate for this study because it incorporates issues of scale between local and national land management practices, as well as the political and economic weight of the timber industry in fire management policies.

NORTHSTAR COMMUTER RAIL: HOW HAS THE COMMUTER RAIL POLICY PROCESS UNIQUELY AFFECTED DEVELOPMENT ALONG HIGHWAY 10?

Anne Taff (Advisor: Laura Smith)
Geography Dept., Macalester College, St. Paul, MN

The objective of this research project is to examine the connection between transportation and land use in the context of community response to a transit line–specifically, the proposed Northstar Commuter Rail between Minneapolis and St. Cloud along Hwy. 10. The Northstar project has faced both the "normal" obstacles inhibiting transit projects in metro areas, and a new, more conservative political agenda. An increased focus on cost efficiency and personal transit has drawn attention away from the project and simultaneously forced the process through unforeseen hoops.

Throughout the research, I discovered that the policy process driving/impeding the installation of the rail has uniquely influenced decisions regarding land use change along the corridor. This will be shown by analyzing general transportation land use connections, the progression of the Northstar project, and the corresponding land use decisions along the corridor. The information to be presented was gathered through interviews with commuter rail officials, state legislators, and city planners and developers. To supplement this, current media coverage, city comprehensive plans, Metropolitan Council blueprints, Northstar status reports, and geographical texts were used as background resources.

ENVIRONMENTAL CONSEQUENCES OF THE "IRON CURTAIN" REMOVAL IN EASTERN AND CENTRAL EUROPE: THE ANALYSIS OF KUZNETS' ENVIRONMENTAL CURVE

Jovana Trkulja (Advisor: Bill Moseley)
Geography Dept., Macalester College, St. Paul, MN

As Eastern Europe's heavy manufacturing industries strived to meet the planned output levels set by their central governments, they neglected to consider the environmental consequences caused by the implementation of inefficient production methods. This paper examines the environmental effects of economies in transition as they lift the "iron curtain" and adapt to the free market system. The applicability of Kuznets' environmental curve is put to question in the case of the former eastern bloc countries. The Danube River is the focus of the study because it flows through three countries of differing social standing and economic growth: Austria, Hungary and Romania.

To quantify the pollution levels caused by each country, this paper conducts a comparison of nitrogen and phosphate usage in agricultural fertilization. These two variables are chosen since they are Danube's primary pollutants. The findings of the paper point to the fact that countries in transition do not fully follow Kuznets' environmental curve. My results suggest that as Romania and Hungary experienced their economic transition, their levels of pollution rose. The data, however, show that the altering levels of environmental degradation are not solely the cause of economic factors, as the core theory states, but also of political factors. These factors include governmental transparency and openness, implementation of pollution controls, and motivation of industries to engage in more environment-friendly production methods.

HOME DEPOT, CHIPOTLE, AMERICAN EAGLE, AND BEST BUY: THE GROWTH OF MANKATO AS A RETAIL DESTINATION

Jeff Turrittin
Dept. of Geography, Gustavus Adolphus College, St. Peter, MN

The counties of Sibley, Nicollet, Le Sueur, Blue Earth, Brown, Watonwan, Waseca, Martin, and Faribault have undergone a concentration of retail space in their regional center, Mankato. The study uses a spatial interaction model based on Reilly's Law of Retail Gravitation to examine the changing retail trade areas for the surrounding communities. The study will assess trade area capture for electronics, furniture, gas, food, and dining. The growth of Mankato into a regional destination is evidenced in the movement of its retail core from downtown farther up Mankato's retail spine, Madison Avenue. Consequently, the main streets of surrounding communities have floundered in retail decline.

Molecular and Cellular Biology

Symposium, Sponsored by the American Society for Biochemistry and Molecular Biology Undergraduate Affiliate Network

ADRENERGIC MECHANISMS IN ADOLESCENT DEPRESSION

David Bylund
Dept. of Pharmacology, Univ. of Nebraska Medical Center, Omaha, NE

Tricyclic antidepressants are an effective treatment for adults with major depressive disorder, but in children they are no different from placebo. By contrast, the selective serotonin re-uptake inhibitors (SSRI) are as effective in children and adolescents as they are in adults. A potential reason for this difference is that maturation of the noradrenergic system is slower than that of the serotonergic system, and that the noradrenergic system is not fully developed in humans and rats until adolescence. Because tricyclic antidepressants have a major noradrenergic component, an immature noradrenergic system in young humans and animals may respond differently to this class of drugs as compared with adults. The alpha-2 adrenergic receptor, which down-regulates in the brain following chronic desipramine administration to adult rats, up-regulates in juvenile animals. An

understanding of the adrenergic mechanisms in juvenile depression should lead to better therapeutic modalities for the treatment of affective disorders in children and adolescents.

FUNCTION OF ACTIN CAPPING PROTEIN SUBUNITS IN THE HEART

Marilyn Hart
Dept. of Biological Sciences, MN State Univ., Mankato, MN

In striated muscle, the barbed ends of thin filaments are attached to Z lines. To mediate this attachment, biochemical and cell biological studies suggest that actin capping protein (CP) binds the barbed ends of actin filaments and a-actinin crosslinks overlapping filaments from adjacent sarcomeres. My previous studies in transgenic mouse hearts support the hypothesis that CP attaches thin filaments to the Z line. Defective interaction between CP and the actin thin filaments causes major structural defects in sarcomere organization and leads to cardiac hypertrophy and lethality. The goal of my research laboratory is to understand how thin filaments are attached to Z-lines and how defects in this attachment can affect the structure and function of the heart. We are currently characterizing an existing mouse model for human cardiomyopathy caused by defects in a sarcomere component and identifying interacting proteins of the Z-line.

PHOSPHORYLATION-DEPENDENT REGULATION OF THE GUANYLYL CYCLASE-LINKED NATRIURETIC PEPTIDE RECEPTOR FAMILY

Lincoln Potter
Cellular and Molecular Biology Program, Univ. of Minnesota, Minneapolis, MN

Atrial natriuretic peptide (ANP), brain natriuretic peptide, and C-type natriuretic peptide compose a family of endocrine/paracrine factors that are known to regulate sodium and water excretion, smooth muscle relaxation, vascular remodeling, bone growth, and cell proliferation. The primary signaling molecules for these factors are the guanylyl cyclase-linked natriuretic peptide receptors A and B (NPR-A and B). They consist of an amino-terminal extracellular ligand binding domain, a single membrane spanning region, an intracellular kinase homology domain (KHD), and a carboxyl-terminal guanylyl cyclase catalytic domain (see figure). The activation of these enzymes results in the synthesis of the second messenger, cyclic GMP. We have shown that these receptors are highly phosphorylated and that dephosphorylation mediates both homologous (natriuretic peptide-dependent) and heterologous (PKC-dependent) receptor deactivation, a process known as desensitization. We identified the exact location of five and six phosphorylation sites within the KHD of NPR-A and B, respectively, and discovered that phosphorylation is absolutely required for their activation. These data suggest that the enzymes that add the phosphate to (protein kinase) and remove the phosphate from (protein phosphatase) NPR-A and B are critical regulators of the natriuretic peptide signaling pathway. We are currently using a number of biochemical and molecular genetic approaches to identify these molecules because we anticipate that they may be useful drug targets for the treatment of diseases such as hypertension, atherosclerosis, and dwarfism.

REGULATION OF DOPAMINERGIC NEUROTRANSMISSION BY DOPAMINE TRANSPORTER PHOSPHORYLATION.

Roxanne A. Vaughan
Dept. of Biochemistry and Molecular Biology, Univ. of North Dakota School of Medicine and Health Sciences, ND

The dopamine transporter DAT, is a neuronal plasma membrane protein that serves a vital function in the reuptake and control of synaptic levels of the neurotransmitter dopamine. DAT is also a target for abused and therapeutic drugs, and may play a role in neurological disorders such as depression, schizophrenia, and Parkinson's disease. There is increasing evidence that some activities of DAT are subject to acute control by treatments that affect phosphorylation pathways, but the molecular basis for this is not understood. Recent work from our laboratory and others suggests that these regulatory processes may involve phosphorylation of the transporters by protein kinase C and other kinases, and may occur by affecting intrinsic transport activity or by controlling transporter cell surface expression. Phosphorylation-mediated regulation of DAT provides the potential for acute presynaptic control of neurotransmitter levels during normal neurophysiologic events, and its dysregulation may lead to inappropriate transmitter clearance that contributes to the etiology of neurological disorders.

WINCHELL UNDERGRADUATE RESEARCH SYMPOSIUM

AN INNOVATIVE METHOD TO PURIFY DNA

Amanda Y. Anania, Mark A. Wallert, and Joseph J. Provost
MN State Univ., Moorhead, MN

Plasmid DNA can be described as being a self-reproducing element that houses DNA in its small structure and is located outside of the chromosome. A familiar example would be its existence outside of the genome of particular types of bacteria. Due to its value in biotechnological applications, the demand for pure and inexpensive DNA easy to produce is high. However, at

the given moment, purifying DNA takes around 8 hours to complete and is an expensive process because the kits available at this time are not reusable. Currently, we are working with a new material that can decrease the amount of time involved with plasmid purification, can cut costs, and is reusable. Endotoxin is a bacterial protein that commonly co-purifies with DNA and is a potential problem for using the DNA with mammalian cells and for gene therapy. This new kit will decrease the amount of endotoxin without the use of detergents or other potentially dangerous compounds used in many kits that are currently available. By using this new material, we hope to achieve equivalent yields and purity obtained by the popular conventional methods/kits that exist at this time. Ultimately, the process can be beneficial in the aid of production of important substances such as insulin, gene therapy, DNA vaccination, or other biotechnologically produced proteins.

DIFFERENTIATING INATTENTIVE BEHAVIORS IN THE COLLEGE CLASSROOM UTILIZING A SIMPLIFIED WIRELESS EEG

Christopher J. Aura, Matthew R. Stanton, Teresa Kutchma, Corey Perez, Jennifer Seim, Angela Markham, Abbigale Pitts, Edison Perdomo, and Allison Jones
Dept. of Psychology, MN State Univ., Mankato, MN

A significant amount of research has been devoted to the behavioral correlates of inattentive behavior in children. Utilizing a well-developed index, teachers and instructors can accurately gauge the attention level of students in their classrooms, allowing valuable feedback in their development of effective lesson plans. College students, however, in their several years of experience, are much more capable of masking these trademark behaviors. When a child loses interest, they will begin to openly look around the room, wiggle around, or chat with their neighbors; college students are more liable to candidly fidget, shift in their seat, or even maintain eye contact with their instructor while daydreaming. As these behaviors are difficult to distinguish from those of an attentive state, an additional aid is required to better assess attentive and inattentive periods. Given the invasive nature of past devices commonly used, such as the Electroencephalograph (EEG), accurate measurement proved to be very difficult. Bearing a close resemblance to a fluorescent-yellow and blue bike helmet, the Attention Trainer is a simplified, wireless EEG providing active feedback of localized brain activity. Focusing on the structures such as the reticular formation and parts of the frontal cortex, commonly associated with both focus and the switching of attention, it allows accurate distinction of inattentive periods. Pairing the frequency of many hypothesized classroom behaviors with the brain wave activity recorded by the EEG, significant correlations could potentially be established for the predicted behaviors operationally defined in our methodology.

DESIGN AND SYNTHESIS OF NEW COMPOUNDS BASED ON CAPSAICIN AND N-ARACHIDONOYL DOPAMINE: POTENTIAL ANALGESIC AGENTS

Sarah A. Barfknecht and Patrick M. Traynor (Advisor: David B. Rusterholz)
Univ. of Wisconsin–River Falls, River Falls, WI

Capsaicin, the chemical which is responsible for the pungency of chili peppers, also desensitizes the nerve after the initial hot sensation. This observation has led to the inclusion of capsaicin in some over-the-counter pain-relieving commercial products; however, it still has the disadvantage of initially eliciting a burning pain sensation before it provides any pain relief. The natural receptor activated by capsaicin, called VR1, has been cloned and N-arachidonoyl dopamine has been proposed as the natural endogenous ligand. Previous structure-activity relationship (SAR) studies of capsaicin have identified several major regions of its structure that are thought to be involved in receptor interaction

Several target structures based on both capsaicin and N-arachidonoyl dopamine have been designed. It is hoped that appropriate structure modification will minimize the pungent qualities and enhance pain-relieving effectiveness. These structures contain: (a) an amide of a hydrophobic acid, (b) a substituted benzene ring, and (c) a variable distance of separation between these two. The chemical syntheses of these new structures will be presented.

THE EFFECTS OF A SELENIUM-DEFICIENT DIET ON BLOOD GLUCOSE LEVELS IN MICE

Abbie Bennett
Bethel College, St. Paul, MN

Selenium (Se) is a trace mineral which, when deficient in soils, has been shown to contribute to white muscle disease in sheep, particularly those with a vitamin E deficient diet. Additional research has also shown Se to contribute to insulin-mimetic effects on blood glucose regulation in rats.

The purpose of this experiment was to investigate what effects a Se-deficient diet might have on blood glucose levels in mice. The experiment was conducted on two groups of mice: the experimental group was fed a Se-deficient pellet for 6 weeks and regular chow the last 3 weeks, and the control group receiving regular chow for the duration of the experiment. The experimental mice came from parents that were on a Se-deficient diet for 3 weeks prior to mating. Blood glucose levels were tested at 3, 6, and 9 weeks of age. The results showed statistically significant lower glucose levels in the experimental group for the duration of the experiment. Even after being fed regular chow the last 3 weeks, glucose levels of the experimental group increased slightly but were still significantly lower than in the control group. Results were in contrast to what was expected from the insulin-mimetic properties of Se previously reported in rats. Since Se is important for thyroid hormone production, perhaps the Se-deficient

experimental mice had lower blood glucose levels as an indication of lower metabolic rate due to decreased thyroid activity. Another possible explanation for the lack of insulin-mimetic properties may be that the effects of Se-deficiency are often linked with a simultaneous deficiency of vitamin E, which was not included in this experiment.

SCA-1 EXPRESSION USING TRANSGENIC MICE IN A TETRACYCLINE-REGULATED SYSTEM

Michael Berlinger; Harry Orr, Ph.D.; and Tao Zu, M.D.
Univ. of Minnesota, Twin Cities, MN

Spinocerebellar ataxia type 1 (SCA 1) is a progressive neurodegenerative disease caused by an expansion of an unstable trinucleotide (CAG) repeat within the respective gene. Subsequent mutant polypeptide aggregation into the Purkinje cells results in abnormal CNS function and is demonstrated by the observed phenotype of transgenic mice. Pathology indicates that the increase in mutant protein (ataxin-1) aggregation is directly proportional to age, as well as the reduction of dendritic arborization and the increase of nuclear inclusions regarding the Purkinje cells of the diseased rodents. By incorporating a tetracycline-regulated system into the existing mouse model, the ability to "turn off" the gene was achieved. Using this system, it is suggested that Purkinje cell recovery is possible in the early to middle stages of aggregation. While the later stages of the disease are still under investigation, preliminary data demonstrate the possibility to halt the progression of the disease even if complete cell recovery is not possible.

IDENTIFICATION OF INTERACTING PROTEINS VIA A YEAST GENETIC SCREEN

Aja Bjerke, Noah Sutton, and Marilyn Hart
Mankato State Univ., Mankato, MN

Actin, a component of all eukaryotic cells, contributes to cell motility, shape, and organization. In striated muscle, actin capping protein (CP) binds to the barbed end of the actin filament at the Z-line, directing and maintaining proper organization. CP is a heterodimer composed of alpha and beta subunits. Three isoforms of the beta subunit have been identified: beta1 (β1), beta2 (β2), and beta3 (β3). The beta isoforms are produced via alternative splicing of one gene and share 90% sequence identity. Previous studies revealed that the beta isoforms have distinct localizations in cardiac myocytes, with β1 localizing to the Z-line and β2 localizing to the cell periphery and intercalated discs. In transgenic studies, the β1 isoform was unable to functionally replace the β2 isoform and vice-versa. These studies suggest that the unique function of the beta isoforms may be due to interactions with additional cellular components.

The purpose of this research is to identify proteins that interact with the β1 and β2 isoforms of actin CP using a yeast two-hybrid genetic screen. The genetic screen relies on identifiable gene expression induced by protein interactions. The necessary plasmid constructs have been generated and their orientations confirmed. Western blot analysis revealed that the constructs express the β1 and β2 proteins. A small-scale preliminary screen is underway.

AN INVESTIGATION INTO THE OCCURRENCE OF ICE JAMS

Amanda Brandt
St. Cloud State Univ., St. Cloud, MN

Ice-jam flooding is a serious problem causing over $100 million in damages annually in the United States. A river ice jam is a stationary accumulation of ice that restricts the normal flow of water. Ice jams may form during winter freeze-up or during mid-winter or spring thaws and often cause a rapid rise in water levels and significant flooding. Unfortunately, many National Weather Service (NWS) River Forecast Centers can only react to ice jams in their service areas each winter because the NWS has no procedures in place for forecasting ice jam occurrence.

Therefore, an investigation into the occurrence, severity, and likelihood of ice jams on the Kankakee River near Wilmington, Illinois was undertaken. The main objective was to develop threshold indicators to act as a tool in forecasting devastating floods cased by ice build-up. It was discovered that certain values of the accumulated freezing degree days, flow statistics, and rate of rise of the river are promising ice-jam predictors, along with the mean flow being a reasonable predictor of flood stage.

This research will greatly impact the affected NWS and RFC offices, along with the emergency management and disaster relief teams in the nearby communities. Not only will these findings allow the NWS to better monitor the areas that are at high risk for ice jams, but these offices will be able to offer advanced lead time for flood watches and warnings, allow additional opportunities for controlled break-up of ice, and ultimately reduce flood damages by increasing the preparedness for the community.

THE ROLE OF CHLOROPHYLL FLUORESCENCE METHODOLOGY IN Fv/Fm RECOVERY FROM LIGHT STRESS

Margaret Broeren and Dr. Amy Verhoeven (Advisor)
Dept. of Biology, Univ. of St. Thomas, St. Paul, MN

In researching the affects of light stress on plants, chlorophyll fluorescence parameters reveal important information about the physiological state of the plant. The fluorescence parameter F_v/F_m is a measure of the maximum efficiency of PSII photochemistry, and is often used to indicate whether a plant is under stress. In order to draw meaningful conclusions, F_v/F_m must be measured in dark-acclimated plants, which is optimally done predawn. Many studies have reported F_v/F_m values from plants after shorter dark acclimation times, or after using leaf clips to darken portions of a leaf in the light. The goal of this study was to compare techniques for

assessing F_v/F_m, in order to determine which methods are most reliable (i.e. give results closest to predawn values). A Hansatech fluorometer was used to collect F_v/F_m and PSII data for four species located at the University of St. Thomas in St. Paul, Minnesota. Predawn data was collected for all four species, and three methods were applied to monitor the F_v/F_m recovery time after 10, 20, 30 and 60 minutes of darkening leaves collected in the light. Leaf clips were placed on the four species in the afternoon and darkened until measurements could be taken the next morning. The outdoor, indoor and cloudy methods were not sufficient to allow complete recovery in all species, but the clipped method was successful in reaching a predawn fluorescence equivalent.

POLYCLONAL ANTIBODY PRODUCTION IN RABBITS USING A PURIFIED GREEN FLUORESCENT PROTEIN ANTIGEN

Derick S. Burgad, Alicia J. Levorsen, Amanda M. Lipp, and Joseph J. Provost
Departments of Biology and Chemistry, MN State Univ., Moorhead, MN

The gene that encodes the green fluorescent protein (GFP) comes from a jellyfish, *Aequorea victoria.* This gene was transformed and expressed in *Escherichia coli.* A lysate solution of the GFP expressing *E.coli* was then purified and injected subcutaneously into rabbits for production of polyclonal antibodies. The purpose of this experiment was to produce rabbit specific polyclonal antibodies toward a highly purified preparation of the antigen GFP. The purification process involved dialysis to reduce the salt concentration and the use of two different chromatography columns. In the first step, an IEC column containing DEAE Sephacel was used to fractionate the sample based on charge; in the second step, a SEC column using Sephadex S-100 beads was used to fractionate by size. Following the purification through each column, fractions were collected and analyzed by the Bradford method to determine protein concentration. The final protein sample was then concentrated using a Centriprep YM-10 centrifugal filter unit. Analysis was performed on the final sample to determine purity by an SDS-PAGE gel. The final sample was then emulsified using complete freunds adjuvant and a boost with incomplete freunds adjuvant. Four weeks post-injection sera was isolated. The titer of the sera was tested by both Western blot and an ELISA.

IS THE BUNDLING OF ACTIN BY eEF1A ESSENTIAL IN YEAST?

Jessica Burtness
Dept. of Biological Sciences, St. Olaf College, Northfield, MN

Many studies have suggested a role for the actin cytoskeleton in translation, but the molecular details of this role are unknown. One link between the actin cytoskeleton and yeast is through eukaryotic elongation factor 1A (eEF1A), which plays a central role in protein synthesis by delivering aminoacyl-tRNAs to the elongation polypeptide at the ribosome. eEF1A has been shown to bind and bundle actin in vitro, and previous work has shown that overexpression of eEF1A disrupts actin morphology. However, the role of actin bundling by eEF1A in vivo has not been investigated.

We undertook a study to look specifically at the role of actin bundling by eEFIA. We turned our attention to bacterial elongation factor IA (EFIA), a homolog of eEFIA which functions similarly in translation. We have purified EFIA from *Escherichia coli* and found that it does not bundle actin. We are now in the process of determining whether EFIA can functionally replace eEFIA in yeast. If EFIA can functionally replace yeast eEFIA, bundling of actin is not essential to viability, although more subtle defects in translation may be observed.

CHARACTERIZATION OF SiO MASER FEATURES USING AN AUTOCORRELATION FUNCTION

William Cox and Gordon McIntosh
Univ. of Minnesota, Morris, MN

We have been collecting spectra of the SiO maser emission of the stars Mira and R Cassiopeia. We intend to examine the linewidths of the maser features that make up these spectra. Each feature should be described by a Gaussian distribution. However, in our spectra individual maser features are blended together. This makes it impossible to fit a Gaussian distribution to each feature. Thus, we will be using an autocorrelation function to characterize the linewidths. An autocorrelation function provides a measure of how the intensity at one velocity affects the intensity at another velocity. This autocorrelation function, being derived from a set of Gaussian distributions, should itself be described by a Gaussian distribution. Therefore, we will be using the width of these autocorrelation functions to characterize the width of the maser features found in the spectra.

RESOURCE LIMITATION AND PATTERNS OF SEX ALLOCATION IN PRAIRIE LARKSPUR *Delphinium virescens*

Nicole Duxbury, Simon Emms, and Stephanie Churchill
Univ. of St. Thomas, St. Paul, MN

Like most Angiosperms, prairie larkspurs *Delphinium virescens* are insect-pollinated hermaphrodites. Thus, to maximize reproductive success they must optimally divide their resources among three types of floral structure: stamens, through which they achieve male success; pistils, through which they achieve female success; and attractive structures (petals, nectar, and scents), through which they attract pollinators. Moreover, the optimal division of resources may change through the blooming season as the opportunities for male and female success change.

Previous research in 2000 and 2001 found that larkspurs always decreased their allocation to female

structures in late-blooming flowers, but that changes in male allocation were more variable. The decrease in female allocation appeared to be due to the fact that ovules were less likely to mature into seeds in late-blooming flowers. However, the reason for the decline in seed maturation rates was not identified. In 2003, we confirmed that male allocation typically increases in late-blooming flowers. This is a very unusual pattern, and the reason why it occurs has not yet been identified. We also carried out an experiment to determine the cause of declining seed maturation rates. Early-blooming flowers were removed from some inflorescences and the seed set in late-blooming flowers was compared with that of control inflorescences that were left intact. Control inflorescences produced fewer seeds in late blooming flowers than experimental inflorescences, supporting the hypothesis that resources were limiting seed set and explaining why female allocation declines through the blooming season in this species.

CHEMICAL CHARACTERIZATION OF OSTARIOPHYSAN ALARM SUBSTANCE

Beth Ehlers, Joseph Provost, Mark Wallert, and Brian Wisenden
Dept. of Biology, MN State Univ., Moorhead, MN

The Ostariophysi is a group of freshwater fishes that include the minnows, tetras, catfishes, and suckers. Collectively, they compose about 64% of all freshwater fish species in the world. One feature that all of these fish have in common is the presence of specialized club cells in their skin that contain an alarm substance. When one of these fish is attacked by a predator, club cells are ruptured and alarm substance is released. Nearby fishes smell the chemical and adopt antipredator behaviors that reduce their probability of being captured by the predator. The chemical nature of alarm substance is not well understood. Some evidence points toward a small molecule, other evidence suggests that it is a large molecule such as a protein.

In this study, we used dialysis tubing to separate skin extract of zebra danios into large and small molecules. We found that the fraction of skin extract that contained only small molecules retained full biological activity. We conclude that although large (proteinaceous) molecules may serve as carriers, the active ingredient(s) of minnow alarm cue is a small molecule. Understanding the chemical nature of this signaling system will contribute to greater understanding of the ecology of this dominant group of fishes.

SEXUAL REPRODUCTION OF THE DIATOM *Cyclotella bodanica* IN FOY LAKE, MONTANA

Robert P. Ellis
Metropolitan State Univ., St. Paul, MN

Foy Lake is dominated by the planktonic diatom *Cyclotella bodanica*. By examining annual slides prepared from a sediment core that date from 1917 to 1997, my goal is to determine the length of time between sexual reproduction events of this species. My initial prediction was that these events are not as frequent as assumed and may be on the order of decades apart. Comparison of size profile graphs produced using data from each slide examined has revealed the reproduction cycles as well as the cardinal sizes of this population of *C. bodanica*. I have observed cells ranging in size from 14 to 45 µm, whereas the described range for *C. bodanica* is from 20 to 80 µm. Data of yearly C/N ratios, %N, and TOC are compared with the reproductive cycles to determine whether nutrient levels are linked to sexual reproduction events or if such events are solely dependent.

METHOD DEVELOPMENT AND STUDY OF THE WINTER REARRANGEMENT OF THE PHOTOSYSTEMS IN CONIFERS

Leigh Farrell
Univ. of St. Thomas, St. Paul, MN

One of the major stresses that affects photosynthesis in evergreen plants is winter weather. In northern climates, plants will experience long periods of low temperatures and high light. Conifers have adapted to these stresses through down-regulation of photosynthesis. Rearrangement of the proteins in the photosystems is involved in this process of down-regulation. To study this change in plants, native green gel electrophoresis can be used which breaks apart protein complexes, but allows tightly coupled proteins to remain together with their pigments still bound.

The purpose of this study was to examine fir (*Abies*) conifers in central Minnesota for winter acclimation and rearrangement of the pigment-protein complexes in photosystem II. To find the best method of analysis, green gel electrophoresis was used following methods outlined in several similar studies. It was found that a Deriphat-160 method (Lee et al., 1995) had the best success.

WINTER ACCLIMATION AND REARRANGEMENT OF PHOTOSYSTEM II IN FIR (*Abies*) CONIFERS

Leigh Farrell
Univ. of St. Thomas, St. Paul, MN

One of the major stresses that affects photosynthesis in evergreen plants is winter weather. In northern climates, plants will experience long periods of low temperatures and high light. Conifers have adapted to these stresses through down-regulation of photosynthesis. Rearrangement of the proteins in the photosystems is involved in this process of down-regulation. To study this change in plants, native green gel electrophoresis can be used which breaks apart protein complexes, but allows tightly coupled proteins to remain together with their pigments still bound.

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complexes in photosystem II. To find the best method of analysis, green gel electrophoresis was used following methods outlined in several similar studies. It was found that a Deriphat-160 method (Lee et al. 1995) had the best success.

ISOLATION AND STRUCTURAL DETERMINATION OF PHOTOINDUCED AUTOINHIBITOR NATURAL PRODUCT PRODUCED BY *Schizophyllum commune*

Gina Fasching
College of St. Benedict/St. John's Univ., Collegeville, MN

The fungus *Schizophyllum commune* produces a photoinduced autoinhibitor that appears to be linked to sporulation (reference Keith Klein). Bioassay-guided fractionation was used to purify the natural product responsible for this autoinhibitory activity; a disk-diffusion bioassay was used to test for fungal inhibition. The fungus was grown in a CYM broth. The broth was extracted with EtOAc that was evaporated to produce the crude extract. The crude extract was dissolved into ACN and extracted with hexanes to remove lipids. The ACN fraction was separated via a Sephadex column to yield fractions A-M. (reference the cardellina paper). Fractions C, D, J, and K were shown to inhibit fungal growth of *S. commune*. Fractions J and K were pursued and separated further via reverse phase HPLC. When sufficient material has been isolated, structure determination using NMR and mass spectrometry will be done.

ORGANELLE BIOGENESIS IN *Leishmania* PARASITES

Dr. John Flaspohler, Heidi Meiers, and Lindsay Riley
Concordia College, Moorhead, MN

Members of the genus *Leishmania* are blood and tissue pathogens of vertebrate organisms. Kinetoplastid organisms such as *Leishmania* possess an unusual organelle called the glycosome, an organelle similar to peroxisomes of higher eukaryotes. This organelle has been proposed as a potential target for anti-leishmaniasis therapies.

This project is attempting to further understand the biogenesis of the glycosome organelle and its involvement with *Leishmani* pathogenesis. The project focused on the cloning of a glycosome biogenesis gene (*LmPEX2*) from the *L. mexicana* genome. Ultimately, the use of this construct to knock out *LmPEX2* in *L. mexicana* will aid in understanding the gene's role in pathogen survival/virulence. This research also focuses on the isolation of other potential glycosome biogenesis genes from two mutant *L. donovani* cell lines deficient in glycosome biogenesis.

SEASONAL PREFERENCE OF *Microtus pennsylvanicus* FOR NON-NATIVE COOL SEASON GRASS SPECIES WITH DIVERSE PLOTS IN ADJACENT FIELDS

Linnea Friberg
Bethel College, St. Paul, MN

When Minnesota was first settled in the 19th century, a considerable portion of the habitat was native prairie grasslands. At the present time, there are only a few scattered remnants of what used to be a unified expanse of prairie. In the interest of recreating the best habitats, research using conventional catch-and-release methods was conducted to determine the types of prairie restorations that small mammals prefer. Three major types of restorations, including non-native cool season grasses, native warm season grasses, and fields with a diversity of flowering plants, were sampled from restoration sites at St. Croix Valley Tree Farm (WI) in August 2003. The most frequently captured mammal was the meadow vole, *Microtus pennsylanicus,* and its unique presence was statistically greater in the non-native cool season grass fields. This is in contrast to findings from a similar study sampling the same fields in August 2002, which showed that (1) the meadow jumping mouse, *Zapus hudsonius,* was the most frequently captured mammal, and (2) its unique presence was statistically greater in the diverse fields, never in the non-native cool season grasses. Even though it has been observed that small mammals exhibit population cycles, data from these studies may indicate that a variety of restorations is necessary to ensure the ecological success of a diversity of small mammals.

***Leishmania braziliensis:* CHARACTERIZATION OF A VITAL GLYCOCONJUGATE**

Rebecca Gaalswyk
Bethel College, St. Paul, MN

Leishmania is a protozoan parasite affecting millions of people worldwide with diseases from skin lesions to permanent disfigurement. If untreated, some forms can be lethal. Current treatments, however, are expensive and can result in serious side effects. *Leishmania braziliensis* causes mucocutaneous Leishmaniasis, which leaves permanent disfigurement of the mucous membranes. The parasite has two stages in its life cycle. They are promastigotes within the sand fly and amastigotes within the vertebrate host. The parasite is covered with a glycoconjugate shield that has been shown to change its structure during the different stages of the parasite's life cycle. The glycoconjugate, lipophosphoglycan (LPG), has been shown to be necessary for evasion of the vertebrate host's immune system. Carbohydrate regions of *L. braziliensis* LPG have been isolated and separated from the core/lipid anchor via acid hydrolysis followed by size exclusion chromatography. We have initially characterized LPG from both life stages.

DISTRIBUTION OF MOSQUITO SPECIES IN CLAY COUNTY

Erin Galegher and Keith Gillis (Advisor: Dr. Bryan Bishop)
Concordia College, Moorhead, MN

We evaluated the distribution of mosquitoes in an urban environment compared with a rural environment. Mosquitoes were collected at heights of 1 meter and 4 meters in both urban and rural settings in Clay County, Minnesota. Mosquitoes were attracted using dry ice and trapped during dusk hours with CDC miniature traps. *Aedes vexan* was the most abundant species and did not show a variance in vertical distribution. However, when *Ae. vexan* was excluded from the data set, a preference was seen by *Ae. dorsalis*, which preferred the 1-meter height. *Mansonia perturbans* showed a different distribution between urban and rural settings, being more common in the rural settings. Other mosquito species did not show any differences between rural/urban settings or heights.

SURVEY OF WILD TURKEY (*Meleagris gallopavo*) DISTRIBUTION IN CASS AND CLAY COUNTIES

Katie R. Geray, Tracy Mastel, Natasha W. Gruber, and Donna M. Bruns Stockrahm
Dept. of Biology, MN State Univ., Moorhead, MN

The Minnesota Department of Natural Resources (DNR) is currently working on a wild turkey (*Meleagris gallopavo*) reintroduction program in northwestern Minnesota. To determine the current populations of wild turkeys in Cass County, North Dakota, and Clay County, Minnesota, we distributed surveys along the Red River in the Georgetown, Kragnes, Oakport, Kurtz, and Holy Cross Townships of Clay County, and along the Sheyenne River in the Harwood and Reed Townships of Cass County. Information about the survey was also placed in the Barnesville Recorder and in The Fargo Forum, and those interested contacted us. Surveys were also filled out at the annual meeting of the local chapter of the National Wild Turkey Federation. We received a total of 64 usable surveys and 11 usable e-mails. A total of 537 birds were reported sighted, with 59 identified as toms (male) and 94 identified as hens (female). However, because we have no way of knowing if some of the turkeys were sighted more than once, the actual number reported is probably inflated.

Through this survey, we have discovered a thriving population of wild turkeys in Cass and Clay Counties, and a hunting season may be opened in the area. In the future, we plan to continue surveying residents as well as using GIS techniques to predict if human interactions with wild turkeys are increasing.

A MOSQUITO POPULATION MODEL WITH A GENETIC TWIST

Christopher J. Gervais
St. John's Univ., Collegeville, MN

We develop a Leslie matrix population model for the mosquito that serves as the vector for the malarial parasite. The model simulates logistic growth and incorporates life history parameters and life cycle stages. Variation in these life history parameters is represented in the model by different genotypes. Simulation results suggest that modifications in fecundity or adult longevity would be most efficient for enabling the modified genotypes to replace the wild-type population.

ESTIMATING THE RISK OF GENE FLOW TO WILD POPULATIONS BY MEASURING THE MATING SUCCESS OF TRANSGENIC VS. WILD-TYPE MEDAKA MALES

Ozge Goktepe
Dept. of Fisheries, Wildlife and Conservation Biology, Univ. of Minnesota, St Paul, MN

The development of genetically modified organisms (GMOs) has been a major achievement of modern biotechnology. Genetically engineered fish may be the first transgenic animal to gain regulatory approval (U.S.) for use in commercial aquaculture systems. The rapid production of GMOs raises ecological concerns. Of concern with this technology is fish escape from aquaculture systems, interbreeding with their wild relatives, and subsequent evolutionary consequences and risks. The estimation of this risk is currently assessed by a mathematical model to predict the ability of a transgene to spread throughout a wild population (Muir and Howard, 2001). This model estimates the likelihood, level, and consequence of gene flow between populations using six net fitness traits: (1) juvenile viability, (2) age at sexual maturity, (3) mating success, (4) female fecundity, (5) male fertility, and (6) adult viability.

In this research, I examined one of six traits used to assess fitness in this model, mating success. I used transgenic medaka fish (*Oryzias latipes*) engineered with a salmon growth hormone to enhance growth rate and final size relative to the wild-type medaka population. My hypothesis is that transgenic medaka males have a competitive advantage in mating success over wild-type males. To investigate the mating advantage, I visually observed the mating behavior of wild type and transgenic males with wild-type female medaka and recorded which male attained the first successful mating in each of five trials, of five copulations each. After every trial, the males were moved to compete with a different male and female that it had not encountered before. Statistical analyses of these results indicate that there is no difference ($p \geq 0.30$) between the mating advantage of transgenic and wild-type males in this line, and reinforce the need to test each transgenic line.

Reference: 1. Muir WM and Howard RD (2001) Fitness components and ecological risk of transgenic fish release: a model using Japanese Medaka (*Oryzias latipes*). *Am Natur* 158:1-16.

ZINC-BINDING AND Fur (FERRIC UPTAKE REGULATORY) PROTEIN AFFINITY FOR THE SUPEROXIDE DISMUTASE GENE IN *Escherichia coli*

Kathleen Graf and David Polzin
Univ. of St. Thomas, St. Paul, MN

The superoxide radical mutates the DNA of *Escherichia coli* cells. The ferric uptake regulatory (Fur) protein plays a regulatory role in the transcription of the Mn-containing superoxide dismutase (MnSOD) gene (sodA). It is thought to activate sodA, causing transcription of MnSOD. Fur exists in different forms dependent on the metals bound to it. In vivo, Fur exists either as the 1-zinc form (Zn1Fur) or the zinc-iron form (Zn-Fe2+Fur). In vitro, the 2-zinc form (Zn2Fur) is thought to act in a way similar to that in which Zn-Fe2+Fur acts. Protein shift assays using gel electrophoresis were used to determine the relative affinities of Zn1Fur and Zn2Fur. Zn2Fur has shown a higher affinity for the sodA promoter than Zn1Fur.

PHYLOGENETIC RELATIONSHIPS AMONG *Chaoborus* SPECIES

April Graves and Lauren Lucas
Dept. of Biological Sciences, St. Olaf College, Northfield, MN

The phylogenetic relationships among many *Chaoborus* species have not been described well. We reexamined morphology-based phylogenies using molecular biology to build a DNA-based phylogeny of four species of *Chaoborus* found in North America: *C. punctipennis*, *C. astictopus*, *C. americanus*, and *C. flavicans*. We extracted, amplified, sequenced, and analyzed molecular variation in an 806-827 bp region of the mitochondrial genome (cytochrome oxidase I, Leu-tRNA, and cytochrome oxidase II). Our individual samples unambiguously grouped into the four species. Two clades, *C. americanus* and *C. flavicans*, were only partially congruent with previous morphology-based phylogenies. We also found small but consistent individual variation within three of the species; *C. flavicans* was the most variable. Our future projects will expand the *Chaoborus* phylogenetic tree by including more species from around the world and will examine variation among populations of *C. flavicans*, a species with a Holarctic distribution.

COMPARING AGROECOSYSTEMS: EFFECTS OF CROPPING AND TILLAGE PATTERNS ON SOIL, WATER, ENERGY USE, AND PRODUCTIVITY

Megan M. Gregory, Kathleen L. Shea, and Gene Bakko
St. Olaf College, Northfield, MN

We compared three farms in an unusually dry farming season: conventional (continuous corn and deep tillage), rotation (corn-soybean-oats/alfalfa-alfalfa-alfalfa) and no-till (corn-soybean). Soil organic content was highest on the rotation farm, followed by the no-till farm. Nitrogen content of the soil did not differ significantly among the three farms, although the conventional farm had higher fertilizer nitrogen input. Soil compaction was lower and percent soil moisture was higher in no-till and rotation systems compared with the conventional farm. Soil macroinvertebrate abundance and diversity were highest on the no-till farm, followed by the rotation farm. No invertebrates were found in the soil of the conventional farm. The conventional farm had the highest runoff volume and N loss in runoff when compared with the rotation and no-till farms, and higher P loss in comparison with the no-till farm.

These results indicate that the perennial crops (i.e., alfalfa) used in crop rotations, and plant residue left on the surface of no-till fields, can enhance soil organic content and decrease runoff. Lower soil compaction and higher soil moisture on rotation and no-till farms show that reduced tillage can increase soil aggregation and water infiltration, thus preventing erosion. No-till also promotes diverse invertebrate populations, which play an important role in maintaining nutrient cycling and soil structure. The crop rotation and no-till farms had lower fossil fuel use than the conventional farm due to decreased use of fertilizers, pesticides, and fuel. Furthermore, they provided higher per-acre economic returns due to increased yields and decreased costs.

USE OF RAPD PCR TO DETERMINE GENETIC VARIATIONS IN MUSHROOMS

Deanna Groebner and Presely Martin
Hamline Univ., St. Paul, MN

A molecular approach was used in defining the genetic differences among commercial mushroom strains. Random amplified polymorphic DNA-Polymerase chain reaction (RAPD-PCR) was used to asses the genetic differences. Three types of mushrooms were used, Portobella, Baby bella and the button mushroom. DNA was extracted using a quick extraction method developed for mushroom PCR, and RAPD-PCR amplification was performed using Amersham RAPD-PCR Ready-to Go Beads. We found that DNA concentration was a major variable that needed to be controlled in order to get reproducible results. When equivalent DNA concentrations were used, characteristic differences between mushroom strains could be determined.

THE BEHAVIOR OF A BLOCK COPOLYMER IN THE MICELLE REGIME USING NMR TECHNIQUES

Jennifer Hagen and Dr. Ronald Fedie
Augsburg College, Minneapolis, MN

Previous studies have observed the structure and behavior of block copolymer molecules throughout the various regimes of the full-phase diagram using a number of different techniques: small-angle x-ray scattering (SAXS), rheology, static birefringence, and dynamic light scattering (DLS). One particular question remained within the micelle regime. What is the fraction of free chains not

contained within the micelle structure? The objective of this research was to answer this question employing typical ^{1}H and ^{13}C NMR (nuclear magnetic resonance) techniques as well as some specialty techniques including 2D, pulsed-gradient, and temperature studies. The primary instrument used was a Varian 300MHz NMR. Using styrene and isoprene selective solvents, such as dialkylphthalates, the poly(styrene-b-isoprene) block copolymer micelles form with the appropriate isoprene or styrene block on the inside or outside of the micelle. Spectral results and interpretations will be given about the styrene monomer, poly(styrene) homopolymer, and the block copolymer solutions to elucidate how the polymer dynamics appear in the NMR experiments, including, for example, the peak half-height-line-width broadening with polymer volume fraction increase.

QUANTIFYING THE IMMUNOREATIVITY OF POLYCLONAL IGG AND IGY

Brittney Harthaus (Advisor: Marilyn Hart)
Dept. of Biology, MN State Univ., Mankato, MN

Actin, a filament found in the cytoplasm in all eukaryotic cells, contributes to cell shape, cell mobility, and the organization of certain tissues such as striated muscle. Actin is regulated by a variety of proteins, including actin capping protein (CP). CP is composed of two subunits, an alpha and a beta subunit. In previous studies, the beta subunits have been shown to have distinct functions in murine myocardium.

The goal is to determine if the alpha subunits, reminiscent of the beta subunits, have similar or distinct functions in cells and tissues. As a first step toward accomplishing this, we will determine the location of the alpha subunits in cells/tissues using antibodies specific for each alpha isoform. The objective of this research was to characterize recently generated chicken anti-alpha 2 IgG and IgY antibodies, quantifying their immonoreactivity. Murine hearts were removed and flash frozen, and the tissue was solubilized. The proteins were separated by Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE) and transferred to Nitrocellulose (NC) for subsequent Western Blot analysis. The immobilized proteins were allowed to react with dilutions of the antibodies and visualized with a secondary antibody labeled with alkaline phosphatase. The reactive titers of both the chicken anti-alpha 2 IgG and the chicken anti-alpha 2 IgY antibodies were 10^{5}, providing an initial characterization of the newly generated antibodies and suggesting an approximate working dilution for subsequent studies.

THE EFFECTS OF ETHANOL ON CELL MEMBRANE PERMEABILITY AND ATP SYNTHESIS IN LIVER AND RED BLOOD CELLS OF *Rana pipiens*

Lindsay M. Haus (Advisor: Dr. Richard Kowles)
Saint Mary's Univ. of Minnesota, Winona, MN

Research was undertaken to determine the effects of ethanol on cell membrane permeability and ATP synthesis in liver and red blood cells of the frog *Rana pipiens*. To test cell membrane permeability, both in vivo and in vitro experiments were conducted. In vivo, hearts of frogs were injected with varying concentration of ethanol. In vitro, blood was extracted from frogs and a series of alcohol concentrations was added. Measurements for the in vivo and in vitro parts of the investigation were made by fluorescence cytophotometry. Frog livers were used to extract ATP by trichloroacetic acid and centrifugation. ATP synthesis was analyzed with a spectrofluorometer using a luciferin-luciferase system for bioluminescence.

The in vivo experiment showed that 0.1% alcohol did not increase cell membrane permeability of Hoechst when compared with the control (Ringer's solution). The in vitro experiment showed that 5% ethanol was the amount of alcohol that significantly changed cell membrane permeability of red blood cells to the Hoechst molecule, and that 40% ethanol induced disintegration of cell membranes. ATP synthesis decreased significantly in 0.05% and 0.10% blood alcohol as compared with Ringer's solution, probably due to the breakdown of the proton gradient in mitochondria. Results showed that specific concentration of alcohol might alter cell membrane permeability. However, the effect of alcohol on permeability appears to depend on the size of the molecule.

INVESTIGATING DNA REPLICATION ORIGINS IN *C. elegans*

Jessica Heck and Michelle Malott
Departments of Biology and Chemistry, MN State Univ., Moorhead, MN

In order to ensure that only one complete copy of the entire genome is accurately replicated before mitosis, DNA replication is initiated at multiple locations in the genome called replication origins. Replication origins are of great importance to the regulation of DNA replication and make it impressively efficient. These initiation sites must be uniformly distributed throughout the genome in order to replicate the entire genome within a limited time period. The human c-myc sequence may be involved in the regulation of replication initiation. The human c-myc gene is a proto-oncogene that has been extensively characterized with regard to promoter regulation and chromosomal structure. DNA replication has been demonstrated to initiate within a 2.4-kb region upstream of the c-myc gene in human cells growing in culture. In addition, these sequences, when put into a plasmid and transected into human cells, are able to direct the

replication of the plasmid once per cell cycle. The ability of these sequences of DNA to initiate the replication of a larger piece of DNA, such as a plasmid independently of a chromosome, is referred to as autonomously replicating ability. The c-myc sequences allow plasmids to replicate independently of the chromosome, and are thus said to be autonomously replicating sequences (ARS).

In order to more fully understand replication initiation in metazoan cells, we propose to examine the c-myc sequences that act as replication origins in the model organism *C. elegans*, a small nematode. *C. elegans* is an ideal organism for this type of study because its entire genome is sequenced and has been used extensively to study molecular processes and genetic activities. We intend to work to develop a method to study the initiation of DNA replication within a 2.4-kb fragment of the human c-myc gene using *C. elegans* as a model organism.

PYK2 IN THE INITIAL ATTACHMENT OF HUMAN PROSTATE CANCER CELLS TO PLASTIC

Jessica N. Heck,[1] M. Lee,[2] T. C. Yuan,[2] and M. F. Lin[2]
[1]*MN State Univ., Moorhead, MN,* [2]*Univ. of Nebraska Medical Center Department of Biochemistry and Eppley Institute*

Proline-rich kinase 2 (PYK2) is a non-receptor protein tyrosine kinase (PTK) structurally related to focal adhesion kinase (FAK). Although both kinases differ in tissue distribution, signal transduction, and intracellular localization, PYK2, like FAK, plays an important role in regulating cell adhesion and motility of normal and cancer cells.

We found that among several human prostate cancer cells, PYK2 is highly expressed in androgen-sensitive LNCaP cells. Thus, we used LNCaP cells as a model system initially to investigate the role of PYK2 in prostate cell adhesion. In the LNCaP cell model, C-81 cells (passage number between 81-120) exhibit anchorage-independent growth, while C-33 parental cells (passage number <33) have an anchorage-dependent manner. Furthermore, C-81 cells adhere to plastic flasks significantly faster than C-33 cells ($p<0.01$). Interestingly, PYK2 expression is elevated in C-81 cells approximately 3-fold over that in C-33 cells.

Utilizing phosphorylation site-specific antibodies, we found that the tyrosine phosphorylation level of PYK2 at Y402 (pY402) increases in C-81 cells, approximately 3-fold higher than that in C-33 cells. Similar results were observed in different passages of MDA PCa2b cells, another androgen-sensitive human prostate cancer cell. Higher passage MDA PCa2b cells exhibit higher levels of both PYK2 protein and pY402 and attach to plastic flasks faster than the lower passage cells. Furthermore, increased expression of PYK2 by cDNA transfection in C-33 LNCaP cells increased their adhesion, while the rapid adhesion ability of C-81 cells was decreased by the expression of a kinase-deficient mutant of PYK2 (PKM). Taken together, our data indicated that PYK2 plays a role in the initial adhesion process of LNCaP and MDA PCa2b human prostate cancer cells.

DNA RAPD ANALYSIS OF *Daphnia pulicaria* EPHIPPIAL EGG BANKS IN LONG LAKE, MINNESOTA

Leif Hembre, PhD and Graham Plumhoff
Hamline Univ., St. Paul, MN

Zooplankton egg banks are reservoirs of dormant embryos found in lake sediments that are analogous to the seed banks of terrestrial plants. They are important to the survival of populations because they contain biological diversity that can affect the genetic makeup of active populations.

In this project, we are exploring the potential impact that the hatching of resting eggs (ephippia) has on an active population of *Daphnia pulicaria* by determining the genetic diversity of the egg bank. To do this, we have extracted DNA from individual *Daphnia* hatched from ephippial eggs of different age (dating back to the 1920s) and subjected the DNA to a replication procedure called Randomly Amplified Polymorphic DNA (RAPD). The samples were then analyzed by gel electrophoresis to investigate genetic polymorphism among individuals. Indications suggest that samples more similar in age are more closely related than those of different age.

CAN SOME PREDATORS AVOID BEING CHEMICALLY LABELED BY THEIR PREY?

Jodi Hendrickson, Courtney Rud, Jeni Donner, and Brian Wisenden
Dept. of Biology, MN State Univ., Moorhead, MN

Minnows (and all Ostariophysi) respond with antipredator behavior to chemical alarm cues released from damaged minnow skin. Previous studies have shown that minnows can also detect alarm cues, or their metabolites, that leak out of the gut of predators. This chemical labeling of predators by their prey reduces predator hunting success. Therefore, there should be selection pressure on predators to mask or break down these signaling molecules to avoid alerting the prey of their presence.

We tested two predators: northern pike and largemouth bass. Pike are known to be chemically leaky in that the prey can detect alarm substance of ingested prey; however, bass have never been tested. Evolutionarily, bass (Acanthopterygii) are much more advanced than pike (Protacanthopterygii). Are bass as chemically leaky as pike, or have bass evolved a mechanism to block chemical labeling by minnow prey? We tested this idea using zebra danios as a representative minnow species. We prepared predator odor from bass or pike that had been maintained on a diet of either danios or swordtails (a non-minnow). We used blank water, and undigested skin extract swordtails, as negative controls. We used undigested skin extract of zebra danios as a positive control. We quantified antipredator behavior of

danios as a reduction in activity and movement toward the substrate. If zebra danios respond to the bass on a zebra danio diet, then we can conclude that bass give off a chemical label that can be detected by the prey. If the danios do not have a response to bass on a danio diet, then we can conclude that bass can block chemical labeling.

MICROSPECTROPHOTOMETRY FOR FORENSIC PURPOSES

Krina Hoy and Gary Mabbott
Dept. of *Chemistry, Univ. of St. Thomas, St. Paul, MN*

The goal of this work is to obtain spectra of an entire image through a microscope with the aid of a digital camera and computer software in order to analyze the dye of fibers. Spectroscopy can be used to find wavelength maxima absorbed and/or reflected by a fiber along with a specific wavelength profile at a particular point and has then been used to match the unknown dye of a fiber with a known dye in forensic labs. However, a conventional spectrometer cannot work with a single fiber.

In experiments thus far, a Variable Interference Filter has been mounted directly underneath the eyepiece of the microscope and any sample being viewed through the lens will first be viewed through the filter. This filter helps us to specify the wavelength that is viewed. Also, a waveguide (in the form of Plexiglas) was found to reduced glare and spread light more evenly along the fiber.

Future experiments will include reconstructing spectra from different objects using computer software (Igor Pro ®). We are in the process of characterizing wavelength dispersion (change in wavelength with respect to pixel number) for each position of our filter and are currently experimenting with different methods of internally calibrating the intensity of the light illuminating the fiber and investigating the use of internal wavelength calibration standards.

PRELIMINARY ANALYSIS OF LOCAL WATER SOURCES TO BUILD AN ENVIRONMENTAL SCIENCE CURRICULUM

Christine Isenberg
College of St. Catherine, St. Paul, MN

The objective for this research project was to use elementary-age test kits and Globe protocols to check the presence of testable chemical species in the Dew Drop Pond located on the College of St. Catherine campus. The Globe project is an international initiative to engage students in taking air, soil, water and land cover measurements as part of an elementary and secondary curriculum. These measurements are then made available and used by professionals in the scientific community. The Globe project protocols for hydrology include water temperature, dissolved oxygen, pH, nitrate, water transparency and alkalinity. For this project additional analysis included iron, hardness, phosphate, carbon dioxide, silica, chloride, ammonia nitrogen and sulfide levels. Four sites were selected and tested on three separate occasions. After analysis of the data, levels for pH, alkalinity, dissolved oxygen, chloride, carbon dioxide and hardness were all within normal ranges. The results reveal the chemical levels of the Dew Drop Pond to be conductive to a healthy aquatic environment. The work presented is an example of a successful interdisciplinary research project between the elementary education department and the chemistry department.

THE REACTION OF BORAZINE AND PHENYLISOCYANATE

Ben Johnson, Cory Nelson, and Gary Edvenson
Dept. of Chemistry, MN State Univ., Moorhead, MN

The reaction of borazine, $B_3N_3H_6$, with phenylisocyanate was carried out at room temperature in an attempt to make a boron-containing nucleoside derivative. The product was characterized by ^{1}H, ^{13}C, and ^{11}B NMR spectroscopy, as well as IR spectroscopy. The ^{1}H and ^{11}B NMR spectra are inconclusive. The IR spectrum indicates that a reaction has occurred, and the ^{13}C NMR spectrum shows the possibility of a product containing a carbonyl functional group. Further studies and attempts at characterization are in progress.

ANALYSIS OF CELL-CYCLE REGULATORS DURING CENTROSOME AMPLIFICATION IN HUMAN BREAST CANCER CELL LINES

Heidi Jo Johnson, Robert Busby, Antonino B. D'Assoro, M.D., and Jeffery L. Salisbury
Dept. of Biology, MN State Univ., Moorhead, MN, and the Mayo Clinic, Rochester, MN

In malignant breast tumors, the deregulation of cell-cycle checkpoints may lead to centrosome amplification, chromosomal instability, and the generation of diverse cancer phenotypes. In this study, we looked at three cell lines: MCF-7, MCF-7 Dnp53, and MDA-MB 231, each one expressing a unique centrosome phenotype. MCF-7 has wt p53 and its cell cycle was blocked by the depletion of estrogen hormone. MDA-MB 231 has mutant p53, lacks the estrogen receptor, and is not responsive to estrogen depletion. We transfected MCF-7 with a plasmid containing a mutant p53 driven by the CMV promoter, allowing for a dominant negative effect on the cell line, masking the wild type p53. Previous studies in our lab suggest that in cancer, inactivation of the p53 pathway, Rb hyper-phosphorylation, and deregulation of the G1/S cell-cycle checkpoint represent key events in the genesis of centriole amplification. If any of these key events is disrupted, the results could lead to an accelerated progression of human breast tumors toward a more aggressive phenotype. We have analyzed key cell-cycle regulators by Western blotting techniques to study centrosome amplification in the three human breast cancer cell lines mentioned above.

Our data confirm that the DNA and centriole cycles are linked through the regulation of the G1/S checkpoint. This checkpoint is regulated by the p53

pathway, which is activated in response to DNA damage. FACS analysis and Western blotting show that MCF-7 and MCF-7 Dnp53, when treated with hydroxyurea (HU), which inhibits nucleotide synthesis and induces DNA damage, arrest in S-phase through activation by the p53 mediated checkpoint. The more aggressive cell line MDA-MB 231, following HU treatment, arrested its DNA cycle, whereas the centriole cycle continued to progress, resulting in a hyper-amplified centrosome phenotype.

Centrosome amplification drives chromosomal instability and results in tumor cell heterogeneity. With a better understanding of the key regulators in cell-cycle progression, this may provide a mechanistic link between DNA replication and centrosome duplication cycles.

EFFECTS OF OXIDATIVE STRESS ON *Saccharomyces cerevisiae* FKH1 TRANSCRIPTION FACTOR KNOCKOUT

Heidi Johnson, Michelle Malott and Ellen Brisch
Depts. of Biology and Chemistry, MN State Univ., Moorhead, MN

Forkhead proteins are known to play a role in regulating early development, cell differentiation, and cell cycle progression in many different eukaryotic cells. FOXO3a is a protein from the forkhead FOXO family of human transcription factors. Transcription factors play an important role in regulating cell cycle, cell death, and oxidative stress. They bind DNA through a winged-helix structure. Once translocated into the nucleus, they are able to induce the transcription of genes necessary for these specific functions in the cell. Saccharomyces cerevisiae is a model organism commonly used to study many of these fundamental processes. FKH1 and FKH2 are yeast forkhead transcription factor proteins in S. cerevisiae yeast cells. Although there have been limited studies on FKH1 and FKH2, the conserved DNA binding domain among transcription factors suggests that the yeast transcription factors will behave in a similar fashion to that of FOXO3a. Therefore, FKH1 and FKH2 could be used as a means to study the role of forkhead transcription factors in cellular response to oxidative stress. Bioinformatic analysis will be preformed to obtain the conservation between these three forkhead proteins. Preliminary data has suggested that oxidative stressors, such as hydrogen peroxide, cause DNA damage to cells. When the cell is exposed to hydrogen peroxide, FOXO3a initially inhibits the process of apoptosis, possibly allowing the cell to repair its damaged DNA. We hypothesize that FKH1 and FKH2 will behave in a similar manner. We will focus on the FKH1 protein while performing experiments with wild type strains along with mutated strains lacking the FKH1 gene.

PHOSPHOLIPASE D MEDIATES PHENYLEPHRINE-INDUCED CELL GROWTH AND MIGRATION FACTORS IN CCL39 CELLS

Jessica Johnson, Rachel Sang, Kit Mitchell, Mark Wallert, and Joseph Provost
Dept. of Biology, MN State Univ., Moorhead, MN

Stress fiber formation is an important event in regulating the cell growth and migration of cells. G protein-coupled receptors induce stress fiber formation through a variety of mechanisms. Several studies implicate Gq in the activation of stress fibers, but the mechanism is unknown. In endothelial cells, migration requires both ERK and phospholipase D (PLD) activity. We report here that the addition of the specific α_1-adrenergic agonist phenylephrine (PE) to CCL39 fibroblasts induced stress fiber formation similar to that found with cells treated with lysphosphatidic acid (LPA). PE-induced stress fibers were significantly inhibited in cells treated with the MEK inhibitor PD98059 or primary alcohols.

To investigate the signaling pathway mediating the adrenergic receptor, we examined the ability of PE to activate a number of potential signaling intermediates. Addition of PE induced a threefold increase in PLD activity and a large increase in ERK phosphorylation. Moreover, PE activation of ERK was blocked by the addition of 1-butanol but not 2-butanol. Finally, activation of ERK by PE was attenuated when cells expressed a dominant negative RhoA. These data suggest that PE-stimulated stress fiber formation is mediated by ERK activation and that this pathway is likely activated by action of PLD. Additional evidence for the role of α_1-adrenergic receptors in regulating cell growth is shown by assaying wound-healing rates in the presence or absence of 1- and 2-butanol. The effect of phenylephrine on wound healing is evident in the initial stages of the healing process. Taken together, these results indicate a novel role for PLD in activation of the ERK growth pathway to stimulate early cellular events induced by PE. This work was supported by a MSU Moorhead Faculty Grant, NSF-DUE 0088654 and MRI-DBI 0110537

LAKE-ENHANCED SNOWFALL DISTRIBUTION FOR DULUTH, MINNESOTA

Laura Johnson
Dept. of Earth Sciences, St. Cloud State Univ., St. Cloud, MN

Lake-effect snow seemed to be a prominent weather phenomenon in Duluth, Minnesota, and a forecasting guide to aid in forecasting snowfall amounts would be helpful. After researching the difference between lake-effect and lake-enhanced snow, it seems that pure lake-effect snow does not affect the Duluth area, and lake-enhanced show affects Duluth only a couple of times a year. This paper examines how lake-effect snow is formed, how lake-enhanced snow differs from lake-effect snow, how a guide for forecasting lake-enhanced snow in Duluth was created, how the guide is used, and

conclusions drawn about lake-induced precipitation in Duluth.

ANALYSIS OF HERBICIDES IN THE MINNESOTA RIVER BY SPME-ASSISTED GC-MS

Joseph Katzenmeyer
Gustavus Adolphus College, St. Peter, MN

Solid-phase microextraction (SPME) can be used to concentrate an analyte in order to analyze a sample with greater sensitivity. It also eliminates many of the steps needed to do a traditional extraction with organic solvents. This, in turn, eliminates organic solvent waste.

In this situation, a polyacrylate fiber was used to extract the organics from a sample of Minnesota River water. Various herbicides could be seen, including atrazine, cyanazine, alachlor, and metolachlor. Gas chromatography allows a complete separation of the compounds regardless of their structural similarities. With a preconcentration step using solid-phase extraction, the method allowed the detection of atrazine down to approximately 10 ppt.

A working curve was prepared. However, the results were erratic day to day. Difficulties, such as using a surrogate and possible humic acid blocking, will be discussed.

HEADSPACE ANALYSIS OF CHEMICAL TAGGANTS IN PLASTIC EXPLOSIVES

Joseph Katzenmeyer
Gustavus Adolphus College, St. Peter, MN

Volatile chemicals, called taggants, are required to be added to commercial explosives for the purpose of detection by dogs. New methods for the detection of these taggants are being studied. 2-nitrotoluene, 4-nitrotoluene, 2,4-dinitrotoluene (DNT) and 2,3-dimethyl-2,3-dinitrobutane(DMNB) were studied using a PDMS/DVB SPME fiber, and GC-MS was used to analyze. It was discovered that all of the chosen taggants could be detected in this manner.

In the case of DMNB, the partition coefficients were determined for the PDMS/DVB fiber. This was necessary in order to determine the amount of taggant present in the vial from the result on the fiber. The coefficient between the headspace and the solid was 2.9×10^{-9}. It was also discovered that the coefficient measured for the interface between the fiber and the air was greater than 1 (1.55 actually). This leads us to believe that equilibration is not reached in the timeframe of the experiment. The difficulties resulting from slow equilibration will be discussed.

AVIAN DIVERSITY AND REPRODUCTIVE SUCCESS IN RESTORED PRAIRIE ECOSYSTEMS

Sara Kelzer, Dr. Jeff Port, and Charlene Simpson
Bethel College, St. Paul, MN

As a result of the Conservation Reserve Program (CRP) authorized by the U.S. Congress in 1985, grassland acreage has increased in the United States. One requirement of the program is the planting of a cover crop (seed mixes of three to five grassland flora species) for the duration of the contract period. While research has clearly demonstrated CRP plots to be superior avian habitat compared with agricultural fields, little research has been done comparing CRP plots with restored prairies.

During the 2003 breeding season, we researched the effects of habitat diversity on avian abundance and reproductive success. Using three side-by-side habitat restorations provided by the St. Croix Watershed Research Station (unrestored old farm field, low-diversity fields [CRP], and high-diversity fields composed of 50-60 different species), we were able to compare adult songbird abundance and reproductive success between these three field types. The results suggest species abundance is correlated with habitat diversity. Of our 26 nests, 19 were found in the high-diversity plots. Clay-colored sparrows (*Spizella pallida*) nested and were observed almost exclusively using the high-diversity plots, while the sedge wrens (*Cistothorus platensis*) used the high-diversity plots for foraging only, and nested in the old field.

MOVING TOWARD AN INVESTIGATIVE LABORATORY CURRICULUM: AFFINITY PURIFICATION OF RECOMBINANT FUMARASE

Emily King and Jeff Dahlseid, Ph.D.
Depts. of Biology and Chemistry, Gustavus Adolphus College, St. Peter, MN

Science laboratory curricula are enriched when there are opportunities for personal connection through extended project continuity, project ownership, and creative investigation. To so enrich students' biochemistry laboratory experience, we have begun development of a curriculum with a primary focus upon purification and characterization of a protein enzyme from baker's yeast. Using baker's yeast as a model organism also allows reinforcement of students' molecular genetic literacy. A yeast strain expressing carboxy-terminal, histidine-tagged recombinant fumarase from a high-copy plasmid was used as a rich source of enzyme. In addition, media containing lactate, a non-fermentable carbon source, were used to promote high levels of fumarase expression and growth conditions were determined for optimal yield of wet cell mass. Cell lysis and extract storage conditions suitable for an undergraduate curriculum were determined. The separation of fumarase from total cellular protein was evaluated using three different metals (Ni^{2+}, Co^{2+}, and Cu^{2+}) in immobilized-metal-affinity chromatography. Together, these procedures result in an effective, one-step protocol for isolating and purifying histidine-tagged fumarase with Ni^{2+}-affinity chromatography. This segment of the curriculum establishes continuity over several laboratory periods while emphasizing student responsibility and will lead to opportunities for student-directed investigation.

STATUS OF NATIVE FLOWERING PLANT SPECIES ON GOAT PRAIRIES IN WINONA, MINNESOTA

Sarah Kraszewski (Advisor: Dr. Phil Cochran)
Saint Mary's Univ. of Minnesota, Winona, MN

Goat prairies are found on south- and southwest-facing slopes of bluffs in the Driftless Area of southeast Minnesota and receive more direct sunlight than other slopes. These prairies are inhabited by a variety of plants and animals that do not thrive elsewhere. Goat prairies are declining due to succession and to invasion by exotic species.

The objective of this study was to assess the status of native flowering plant species on goat prairies in the Winona region in order to establish a baseline against which to measure future changes. Another objective was to compare species richness to prairie size. After a pilot study in fall 2002, 11 goat prairies were surveyed five times from May through October 2003. During each survey, the presence of native plants was recorded. Prairie size was determined using aerial photographs and Arc Map. There was no significant correlation between species richness and prairie area. A total of 42 plant species were identified and the peak blooming season in terms of the number of plant species in bloom was during the July sample period. Some species were found on only one prairie, whereas several were present on all 11 prairies. An average of 24 species was found on each prairie, with a minimum of 18 and a maximum of 31 species. Thus no single prairie contained all species.

EXPRESSION OF NITRIC OXIDE SYNTHASE IN ASTROCYTES WHEN EXPOSED TO ASPARTAME

Kathy Kurtz (Advisor: Dr. Randy Krainock)
Saint Mary's Univ. of Minnesota, Winona, MN

Aspartame is an artificial sweetener found in diet drinks and sugar-free foods. This drug has been implicated as a factor causing or worsening the symptoms of multiple sclerosis (MS). Similarly, the actions of nitric oxide synthase (NOS) have been linked to the pathogenesis of MS. Nitric oxide synthase is a calcium dependent protein. Products of the aspartame metabolism have been shown to increase calcium concentrations.

To explore the possible interaction between aspartame and NOS, cultured astrocytes were exposed to aspartame for 2 and 6 hours. Protein electrophoresis was performed on cell lysates, and the resulting membranes were blotted for the presence of NOS protein.

DETECTION OF THE PEROXYACETYL NITRATE PHOTOPRODUCT, NO_3, USING CAVITY RING-DOWN SPECTROSCOPY

Sarah Kvasnicka
College of St. Catherine, St. Paul, MN

Peroxyacetyl nitrate (PAN, $CH_3C(O)OONO_2$) is a significant global transporter of nitrogen oxides (NOx = NO_2 + NO) in the atmosphere. Studying PAN, a major reservoir for these nitrogen oxides, is key to understanding both the oxidizing capability of the atmosphere and the origin of acidic precipitation. In cold regions and at high tropospheric altitudes, photolysis is the dominant pathway in the destruction of PAN and in the production of the nitrate radical.

We present a relatively new method of studying PAN photolysis that probes the photoproducts using a highly sensitive absorption technique called cavity ring-down spectroscopy (CRDS). We show that the photolysis of PAN at 289 nm is significant, and we demonstrate that CRDS is a reliable instrumental tool for the detection of NO_3.

SYNTHESIS AND MUTAGENIC TESTING OF 2-PHENYLMETHOXYETHANAL

Jennifer LaMourea
Dept. of Biological Sciences, Saint Cloud State Univ., St. Cloud, MN

Ethylene glycol ethers are used in a variety of industrial and household products including paints, cleaning agents, and cosmetics. Absorption of these compounds may be harmful to humans as a consequence of the toxic aldehyde metabolites.

Using a two-step sequence, 2-phenyl-methoxyethanal was synthesized from commercially available ethylene glycol. The potential mutagenic properties of the compound are being tested in two biological systems, *Drosophila melanogaster* (the common fruit fly) and bacteria. A skewed male-to-female ratio of *Drosophila* progeny will represent a mutagenic characteristic of 2-phenylmethoxyethanal. If the compound produces a higher frequency of mutational reversion in bacteria than in the control bacteria during the Ames test, it can be considered a mutagen.

MODULATION OF PHOTIC RESPONSIVENESS OF A CIRCADIAN PACEMAKER FOLLOWING LIGHT-DARK ENTRAINMENT

A. J. Langseth and Dr. D. E. Nelson
Univ. of St. Thomas, St. Paul, MN

In mammals, circadian rhythms of behavior and physiology are driven by an endogenous circadian pacemaker located within the hypothalamic suprachiasmatic nucleus. This pacemaker is entrained by environmental light signals that are associated with an animal's LD cycle. The sensitivity of the mammalian photic entrainment pathway (PEP) has been extensively studied in constant darkness (DD). Several pieces of recent evidence suggest that an animal's light-dark history may play an important role in establishing the sensitivity of the PEP to light input. Changes in the responsiveness to light and the period of the pacemaker with extended durations in DD have been observed in both mammals and *Drosophila*.

We have quantified changes in responsiveness of the mouse photic entrainment pathway over cycles 1-14 in

constant darkness following LD entrainment. Mice were maintained in LD12:12 (140μW, fluorescent light) and transferred to constant darkness. The circadian rhythm of running wheel activity was monitored by a data acquisition system (Stanford Software). Saturating light pulses (15min, 500lux; 330mW) were delivered to each mouse at circadian time 16 on cycles 1-14 of DD (9-12/group). Phase delays measured during cycles 1 and 2 of DD (29±9; 63±13min) were significantly smaller than delays of 95±6 to 110±7min induced during subsequent circadian cycles (N=71; P<0.05; ANOVA, Tukey). We also measured photic responsiveness after 25.5hLD (12.75h:12.75h). Following this "delaying" LD cycle, shifts measured on circadian cycles 1, 2, and 3 in DD (3±12; 18±17; 87±10 min) were significantly smaller than shifts of 145±10 to 150±9 measured during cycles 10-14 (N=55; P 0.05; Tukey). Large delays induced on cycles 10-14 were also significantly greater than delays measured after LD24h (N=81; P<0.05; Tukey).

These data demonstrate that entrainment to LD cycles reduces the responsiveness of the PEP to light during the initial circadian cycles in DD. Aftereffects of 25.5LD also include an increased responsiveness of the photic entrainment pathway by 5-7 cycles in DD that persists for at least 14 cycles. During entrainment to LD cycles, the responsiveness of the photic entrainment pathway appears to be modulated directly, through the effects of light on a photoreceptor, or indirectly, by altering the pacemaker itself. These findings also provide an important link between experiments that measure light-induced clock resetting after long durations in constant conditions and the synchronization of circadian clocks during continuous LD cycles experienced by most mammals, including humans. (Supported by MH060122-02.)

COLLABORATIVE SOFTWARE SUPPORT FOR SIX SIGMA METHODOLOGY

Daniel T. Larkin
QMCS Dept., Univ. of St. Thomas, St. Paul, MN

Six Sigma is a process improvement methodology for eliminating defects in any process. The method began as a quality management tool in production, but has since been adopted by the software development industry. Many tools have been developed to assist the Six Sigma methodology, most of which provide statistical analyses, a key element of the methodology. My study examines a different type of software that provides support at earlier steps where team collaboration is crucial. The experiment in this study involves feasibility analysis, an important step in software development, using Six Sigma methodology in concert with collaborative software tools that are part of the Group Systems suite. The experiment consists of two teams--with equal experience and skill levels--analyzing the requirements and feasibility issues of a software development case study. Team A meets as a group without the support of team collaboration software, orally discussing the problem in order to find probable solutions. Team B uses the collaborative research tool to do the analysis. The hypothesis is that the team using the collaborative software will (1) be more efficient, (2) have higher participation, (3) comment more freely on suggestions by teammates, and (4) produce a more complete feasibility analysis as a result of the anonymity of the environment and the ability to track and react to all contributions of team members.

USING SNOW-WATER-EQUIVALENT ALGORITHMS TO DETERMINE THE WATER CONTENT OF A FORESTED SNOW PACK IN COLORADO

David Lerach
St. Cloud State Univ., St. Cloud, MN

From February through March 2003, the University of Michigan's Microwave Geophysics Group took 19- and 37-GHz frequency microwave radiometer brightness measurements of a snow pack at Fraser Experimental Forest in Fraser, Colorado. The group is currently developing a model that will convert these radiobrightness measurements into moisture contents for different terrains.

My project consists of converting these measurements to Snow-Water-Equivalent (SWE) values using four different algorithms derived by Environment Canada for varying terrains: prairie, sparse forest, deciduous forest, and coniferous forest. Once values of SWE are calculated and quality-controlled, they will be compared with actual SWE measurements to discover which algorithm performs the most accurate conversion. Hypotheses based on the forest type and snow depth reports indicate that the sparse forest algorithm should perform the best; however, initial calculations reveal only that the algorithms contain both diurnal and snow melt errors. Attempts to filter these errors will be made, percent errors for each algorithm will be calculated, and results will be presented.

CELL-CYCLE GENES AND THEIR EFFECTS ON MITOCHONDRIAL INHERITANCE AND DYNAMICS

Tammy Lien, Autumn Dinnel, Bree Hamann, and Ellen Brisch
Dept. of Biology, MN State Univ., Moorhead, MN

Mitochondria are responsible for cellular respiration. As they are essential for life, it is critical that they be transported from mother to daughter cell during division. Previous research has shown that mitochondria are transported along the cytoskeleton; they move, fuse, and separate while anchored to the cytoskeleton (Boldogh, 2001). Thus, the cytoskeleton plays an important role in the morphology and distribution of mitochondria throughout the cell cycle. Under normal conditions, mitochondria appear as reticular tube-like structures located at the cortex of the cell. Under abnormal conditions, morphology takes on different

forms, and mitochondria can begin to aggregate, or clump, abnormally (McCoy, Johnson, Risan, 2003). These mutations are usually associated with some loss of function.

The work we propose is based on previous research where proteins closely associated to the mitochondria were mutated (membrane-associated proteins involved with fusion and fission of membranes) and morphological as well as distribution changes were observed (Brisch, 2001). However, the changes were never associated with specific phases of the cell cycle. We plan to mutate three cell-cycle genes, cdc28, swe1, and bub1. We will use a TS Degron/PCR approach to generate temperature-sensitive cell-cycle-specific mutants. After successful transformation, we will examine mitochondrial morphology and distribution regarding our mutations via microscopy and fluorescent staining.

COMPARISON OF GROWTH RATES AND SURVIVAL OF PAINTED TURTLES (*Chrysemys picta*) IN CLAY COUNTY, MINNESOTA

Karen R. Lubenow, Lateesha M. Hektner, Joanna M. Schmit, Natasha W. Gruber, Donna M Bruns Stockrahm, and Jerome W. Stockrahm
Dept. of Biology, MN State Univ. Moorhead, MN

Painted turtles (*Chrysemys picta*) were live-trapped during the summer and early fall of 2001, 2002, and 2003 in Clay County, Minnesota, to study growth rates, recapture rates between years, population characteristics, and movements. In 2001, two sloughs (< 2 km apart) were trapped, 2.7 ha and 6.2 ha, respectively. For 2002 only, a third slough (<1 ha) positioned between the first two sloughs was added to the study. For each captured turtle, outer scutes were notched for individual identification. Turtles were weighed, sexed and measured for length and width of carapace, then released. For 2001, data for 250 turtles were analyzed. In 2002, a total of 118 turtles were trapped where 75 were new animals (37 males, 30 females, and 8 juveniles) and 43 (34 males and 9 females) were recaptured turtles from 2001. In 2003, a total of 133 turtles were trapped where 42 were new animals (20 males, 18 females, and 4 juveniles) and 91 (59 males, 29 females, and 3 juveniles) were recaptured turtles. In spite of intense trapping effort, trapping success between 2001, 2002, and 2003 varied greatly. Possible reasons for these differences, including mortality factors, will be investigated. Growth rates and survival rates will be discussed.

A MORPHOLOGICAL AND MOLECULAR ANALYSIS OF THE VARIOUS COLOR MORPHOLOGIES OF THE SEA STAR, *Oreaster reticulatus*

Erin Lynch and Craig Tepper
Cornell College, Mt. Vernon, IA

Oreaster reticulatus is a large, thick sea star commonly found in the tropical western Atlantic Ocean. Adults range in diameter from 16 to 20 cm and have been reported to have numerous, distinct morphologies with respect to background and spine color patterns (Buchsbaum et al., 1987). Humann (1992) reported that different developmental stages may have distinct colors. Since *O. reticulatus* reproduce by synchronously shedding their gametes into the water (Buchsbaum et al., 1987), the distinct color morphs could be the result of having (1) a single polymorphic species with background color correlated with developmental stage as currently reported in the literature, (2) two or more distinct species, or (3) interspecific hybridization.

This study focuses on individuals collected from Graham's Harbor on the island of San Salvador, Bahamas. Each collected individual was measured across the disk, and the colors of the background and spines were recorded. In addition, a few tube feet were removed, DNA was isolated, and Amplified Fragment Length Polymorphism (AFLP) analysis was conducted to determine if monomorphic markers existed between the different color morphs.

Preliminary results reveal a statistically significant size difference in individuals that have a green background color. The green color morph was smaller than all other background color morphs. There was no significant size difference between individuals that had red, orange, or yellow backgrounds. Preliminary AFLP results show that all color morphs are highly polymorphic. We are currently using numerous primers as we attempt to determine whether color-specific monomorphic DNA markers exist.

APPROACHES TOWARD THE SYNTHESIS OF KASUMIGAMIDE USING SOLID-PHASE SYNTHESIS

Rita S. Majerle, Amanda J. DeGraw, and Kiah Smith
Hamline Univ., St Paul, MN

Peptide-based compounds have exhibited a range of biological activities having served as hypertensive agents to biocides. The isolation of the anti-algal compound kasumigamide from the cyanobacterium *Microcystis aeruginosa* was recently reported. Kasumigamide and its analogs are attractive alternatives to many of the current algacides that are available because of their specificity and very low toxicity. This tetra-amino acid compound, which contains a novel amino acid, has not been synthesized to date. Modeling studies of the peptide would provide site interaction information that could be useful in producing more potent biocides. We report here to date our solid-phase synthesis of kasumigamide and our initial modeling studies.

GENETIC DIVERSITY INFLUENCING SURVIVAL AMONG DECLINING POPULATIONS OF BLACK-TAILED PRAIRIE DOGS

Tracy Mastel, Alisha Pagel, Michelle Malott, and Donna Bruns Stockrahm
Dept. of Biology, MN State Univ., Moorhead, MN

The populations of black-tailed prairie dogs, *Cynomys ludovicianus,* are steadily declining due to a variety of human-impact factors such as habitat alteration, recreational shooting, and agricultural control. A study of genetic diversity could provide valuable insight on the potential survival of black-tailed prairie dogs in future generations. We are using PCR to examine microsatellite markers in order to compare the level of variation of DNA from black-tailed prairie dogs between and among prairie dog towns from Theodore Roosevelt National Park in North Dakota. Microsatellites are regions of the genome that are highly variable between individuals and are being increasingly used as markers of genetic variability in studies of population genetics and conservation. We will present our results and conclusions on our development of a method for analyzing microsatellites from this species of prairie dog. Ultimately, our goal is to determine whether or not the genetic variability of TRNP prairie dogs has been impacted by the lowered numbers of prairie dogs in that population.

DISCOVERING PATHOGEN GENES BY USING YEAST AND PLASMIDS

Kara McArdell
Dept. of Biological Sciences, Saint Cloud State Univ., St. Cloud, MN

Toxoplasma gondii is a single-celled pathogen of animals and humans. It is the reason why you don't take out the kitty litter when you are pregnant. It is one of the many reasons why you should not eat undercooked meat.

Our experimental objective is to identify the DNA sequences of this human pathogen which are responsible for cell division. Since the culture of human pathogens is difficult and dangerous, we employ bakers' yeast as a surrogate experimental system. In brief, we put DNA from the pathogen into bakers' yeast and identify changes in the yeast over time. Our method involves the fundamental genetic principles of mutation, transformation, and complementation. First, a conditional mutant of bakers' yeast is identified. This conditional mutant is then transformed with a gene from the pathogen, and the yeast that no longer have the mutation are identified. The basic assumption is that if a gene can rescue a bakers' yeast mutant, and that mutant has been described in detail, then we can conclude that the gene has the role of the mutation in yeast. We then extrapolate that job of the gene to the pathogen. Thus, we are able to make a conclusion regarding the basic cell biology in a pathogen without endangering students with experimentation using the pathogen directly.

Further studies will allow us to investigate the isolation of the *Toxoplasma* gene or genes that allowed the yeast to grow. The results of these experiments will lead to the development of drugs to inhibit the gene or genes form working, thereby inhibiting growth of the pathogen, and possibly curing any diseases it may cause.

ELECTROPHYSIOLOGICAL CHARACTERIZATION OF ETHOLOGICALLY RELEVANT NEURAL RESPONSE PROPERTIES IN THE TORUS SEMICIRCULARIS OF *Rana pipiens*

Kimberly McArthur and Dr. Michael Ferragamo
Gustavus Adolphus College, St. Peter, MN

Frogs use acoustic signals to facilitate reproduction. For acoustic communication to be effective, females must be able to extract and localize the calls of conspecific males against a background of environmental noise and competing heterospecific signals. The frog's central auditory system accomplishes these tasks at the level of the midbrain via selective neural response properties such as frequency tuning, adaptation, and envelope detection. These properties are the results of both cellular and circuitry mechanisms.

In our lab, we use electrophysiological extracellular recordings in the frog auditory midbrain (torus semicircularis) to study neural responses to acoustic stimuli. We correlate neural response patterns with properties of the acoustic stimulus to discover how these properties are being encoded by the central auditory system.

TREE COMMUNITY DYNAMICS ON SHOVEL POINT, MINNESOTA

Vanessa McClellan
Hamline Univ., St. Paul, MN

Shovel Point is located approximately 80 km northeast of Duluth on the North Shore of Lake Superior and is visited by over 200,000 people each year. The purpose of this study was to determine the demographic (size) structure of the main conifer populations in order to predict the future forest community. Using 10- X 10-m plots, we censused all tree stems in an area 220 m along the cliff edge and 30 m away from the cliff edge. Stems were classified by size as seedlings, saplings, or adults. White spruce make up the majority of the adult conifer community, but less than 10% of the seedling community. White spruce adults are equally abundant on the cliff edge and in the forest interior; seedlings are most rare at the cliff edge. Balsam fir seedlings, saplings, and adults all increase in frequency from the cliff edge to interior. While the overstory near the cliff edge contained a number of large white pine, no regeneration was observed for this species. In the future, white pine will be replaced by spruce and fir. Along the cliff edge, no conifer species was regenerating, suggesting a gradual retreat of the treeline away from the cliff edge.

A COMPETITION MODEL FOR MOSQUITO POPULATIONS

Nicholas F. McClure
St. John's Univ., Collegeville, MN

A new strategy for controlling malaria would be to genetically engineer a new mosquito whose numbers could be more easily controlled by conventional means or a mosquito that is less efficient at transmitting the malarial parasite. This modified mosquito would need to outcompete the natural mosquito. The model reported here examines different features of mosquito life history to determine which might be most significant in enabling the modified population to win out in competition. The model consists of differential equations for competing mosquito populations that incorporate logistic growth, the mosquito life-cycle stages, and seasonal influences. A parameter sensitivity analysis suggests that modified birth, survival, and death rates would best enable an engineered population to win out in competition with another population.

VISUALIZING MITOCHONDRIAL DYNAMICS DURING THE CELL CYCLE IN YEAST

Austin McCoy and Ellen Brisch
Depts. of Biology and Chemistry, MN State Univ., Moorhead, MN

Mitochondria function to provide cells with energy for all metabolic processes. Throughout the cell cycle, mitochondria are highly dynamic. They continuously move about and change shape depending on which stage of the cell cycle they are in. This process is termed mitochondrial dynamics. In *Saccharomyces cerevisiae*, the inheritance of mitochondria from mother cell to daughter bud during cell division is an essential feature of yeast cell growth.

The analysis of mutants defective in mitochondrial morphology and inheritance has led to the identification of some of the proteins that control mitochondrial dynamics. Classically, temperature-sensitive yeast mutants were used to identify cell cycle regulatory proteins. The analysis of mutants defective in events such as bud formation, DNA synthesis, spindle pole body duplication, and cytokinesis led to the identification of proteins that control each of these integral steps in cell division.

It is our hypothesis that molecules that control cell division and cell cycle regulation play a key role in mitochondrial dynamics. We have tested our hypothesis by examining the following cell cycle mutants for defects in mitochondrial dynamics: *cdc13, cdc14*--meiosis and sporulation, *cdc 5, cdc7, cdc15*--kinases, *cdc2, cdc6, cdc9*--DNA replication. We are visualizing mitochondria using specific dyes and fluorescence microscopy. Our results show that cell cycle mutants do not have normal mitochondria; however, the wild-type strain also shows some defects in mitochondria, suggesting that the heavy mutagenesis may have generated background mutations that affect mitochondria. We are currently examining specific temperature-sensitive cell cycle mutants generated by others in the lab. By identifying cell cycle mutants with mitochondrial defects, we can build a model for how mitochondrial dynamics are coordinated during the cell cycle.

ANTIBIOTIC RESISTANCE OF *Escherichia coli* ISOLATED FROM CONVENTIONALLY VERSUS ORGANICALLY RAISED BEEF CATTLE

Rachel McPherson (Advisor: Dr. Jeanne Minnerath)
Saint Mary's Univ. of Minnesota, Winona, MN

The increase in antibiotic resistance among microorganisms is one of the most pressing health concerns today. Antibiotics are present in the feed of livestock. These antibiotics are listed as "energy supplements" and are believed to increase the livestock's growth rate by a number of mechanisms.

The purpose of this study was to compare the antibiotic resistance of the enteric bacterium, *Escherichia coli,* isolated from conventionally raised beef cattle, versus *E. coli* isolated from organically raised beef cattle. Bacterial samples were gathered from the feces of five cows at each of two organic farms and from five cows at each of two conventional farms. *E. coli* was isolated from the feces, and the minimum inhibitory concentration (MIC) assay was conducted to determine the lowest concentration of antibiotic needed to inhibit the growth of the *E. coli.* The antibiotic tested in this study was from the same family of antibiotics as that present in livestock feed. Results from this study will indicate whether bacteria isolated from conventionally raised cattle are more resistant to the antibiotic tested than bacteria isolated from organically raised cattle.

THE ROLE OF PROTEIN KINASE C IN THE REGULATION OF STRESS FIBER FORMATION IN CHINESE HAMSTER LUNG CELLS

Alison L. Metcalf, Tabitha M. Burnside, Matthew P. Duval, Mark A. Wallert, and Joseph J. Provost
Depts. of Biology and Chemistry, MN State Univ., Moorhead, MN

The organization of actin monomers to form stress fibers requires activation of both the small G-protein RhoA and the action of the sodium-hydrogen exchanger (NHE) in the plasma membrane. In previous experiments we have demonstrated that the α_1-adrenergic agonist phenylephrine (PE) stimulates the formation of stress fibers in Chinese hamster lung fibroblasts (CCL39). We have found that PE stimulates the translocation of RhoA from the cytoplasm to the plasma membrane.

Knowing that traditional α_1-adrenergic stimulation acts through Protein Kinase C (PKC), we went on to investigate the role of PKC in RhoA activation and stress fiber formation. To do this, we tested the effects of three PKC inhibitors: bisindolylmaleimidine I (BIM), Go6976, and Ro-31-8220. BIM is a derivative of the general PKC inhibitor straurosporine that acts as a competitive inhibitor for the ATP-binding site of PKC. It

is highly specific for PKCα, βI, βII, γ, δ, and ε isozymes. Go6976 is an indolocarbazole that specifically inhibits PKCα by blocking Ca^{++} binding. Finally, Ro-31-8220 is a staurosporine analogue that inhibits active membrane-bound PKC 12.5 times better than cytosolic PKC.

To investigate the role of PKC in PE stimulation of stress fibers, cells were stimulated with PE in the presence of PKC inhibitor. Under each of the conditions, treating the cells with PKC inhibitor prior to the addition of 100 μM PE completely blocked stress fiber formation. The role of RhoA in stress fiber formation in CCL39 cells was confirmed when the inhibitor Y27632 blocked stress fiber formation. Y27632 inhibits the RhoA associated kinase (ROCK) a known downstream effecter of RhoA involved in stress fiber formation. To determine the ability of PE to activate RhoA, EGFP-tagged RhoA was used to observe translocation. Unstimulated control cells display RhoA dispersed throughout the cytoplasm while PE stimulated cells show RhoA predominantly associated with the plasma membrane. To measure the role of PKC in RhoA stimulation, translocation experiments were repeated in the presence of the PKC inhibitors.

HOW DO MAPK/ERK KINASES REGULATE MICROTUBULE SPINDLE FORMATION?

Sumeda Nandadasa, Ava-Gaye Simms, Akila Weerasekara, Lisa Magstadt, and Ellen Brisch
Dept. of Biology, MN State Univ., Moorhead, MN

The mitotic spindle formation is the key process that allows the segregation of the newly replicated chromosomes into two poles. Microtubules (MT) are the key components of which the mitotic spindle is formed. Understanding Microtubule assembly is important to understanding spindle fiber regulation. In our study, we are trying to understand how microtubule assembly is regulated, what key proteins are involved, and what gives the signal for microtubule sub-particles to assemble and disassemble

In earlier experiments, we have found that two key proteins are involved and they are sizes 44 and 48KD. By antibody tests, we predict that these proteins are in the Mitogen Activated Protein Kinase (MAPK) family and Extracellular Receptor Kinase (ERK) family by using specific antibodies. By using a collaboration of protein assay techniques together with Western blot techniques, we plan to further analyze these two proteins and to identify them using protein microsequencing.

DEVELOPING AN *IN VITRO* ASSAY TO CHARACTERIZE THE FUNCTION OF THE MAMMALIAN CIRCADIAN PACEMAKER

Kyle Nelson
Univ. of St. Thomas, St. Paul, MN

Circadian rhythms in mammals are driven by a neuronal pacemaker within the hypothalamic suprachiasmatic nucleus (SCN). Molecular-feedback loops involving circadian genes produce circadian periods in SCN neurons. Ultimately, these molecular cycles produce changes in firing rates in SCN neurons over the course of the circadian cycle. SCN activity is relatively high during the subjective day and low during the subjective night in mammals. Circadian modulation of neuronal activity in the SCN ultimately synchronizes rhythms in peripheral tissues.

Our lab has focused its research on outputs from the mouse circadian pacemaker—at the level of circadian behavior. To develop a comparable assay of circadian functions *in vitro,* we are attempting to record from SCN neurons from the hypothalamic slice. In this initial project, we are attempting to establish the necessary techniques in our lab for constructing electrodes and recording neuronal activity from single SCN neurons. Coronal slices of the mouse hypothalamus were collected using a tissue chopper (approx. 500 μm). Slices containing the SCN were isolated visually and placed into a perfusion/recording chamber. We used Earle's Balanced Salt Solution (EBSS) supplemented with glucose, bicarbonate to buffer the pH, and bubbled with 95% O_2-5% CO_2. The perfusion chamber consisted of an outer section filled with EBSS and an inner section that allowed EBSS to superfuse up around the tissue slice, which was suspended in the EBSS on a plastic mesh screen. The flow rate of EBSS was 1 ml/min and the temperature was held at 35°C using a circulating bath. We constructed microglass electrodes from thin-walled pipette glass (Stoelting Co.) using a micropipette puller (Sutter Instruments, P-30), filled with a 3M NaCl solution and measured to have a resistance of 4-5 MOhms.

We have initially recorded single unit activity from SCN slices harvested at two times of day from mice that had been maintained on a 14:10 LD cycle. During the subjective day (ZT 4–6:30), neuronal firing rates were 10.3 (+/- 0.6) Hz (mean +/- SEM; n=8). During the subjective night (ZT 18:30–19:15), neuronal firing rates were significantly lower, averaging 3.2 (+/- 0.3) Hz (n=5; Student's t-test; $P<0.001$). This day/night difference in single unit frequency is very similar to that reported by other groups for mice. In the future, we hope to record multi-unit activity from SCN slices in an effort to better "automate" the recording and tracking SCN activity (pacemaker period and phase) over several circadian cycles *in vitro.* Supported by MH-060122-02.

A HISTORICAL AND CHEMICAL ANALYSIS OF MAPLE SAP AND SUGAR AT ST. JOHN'S UNIVERSITY, COLLEGEVILLE, MINNESOTA

Erika Nunnink
College of St. Benedict/St. John's Univ., Collegeville, MN

The maple syrup operation at St. John's was started in 1942 and is still in operation today. In the spring, monks, students, staff, and others gather in the sugar bush to tap sugar maple trees (*Acer saccharum*) and to collect sap to prepare pure maple syrup.

One purpose of this project was to analyze historical documents and to identify long-term trends in the maple sap and syrup production. In addition, we

attempted to see if there was a correlation between the sugar concentration of sugar maple sap and the location of the tree in the sugar bush. We hypothesized that trees near wet areas would yield a higher sugar concentration. We obtained sap from over 500 trees, measured the sugar concentrations with a refractometer, and marked the trees using Global Positioning System (GPS). These data were plotted on a GIS base map and analyzed. The results of this work will be discussed.

TECHNIQUES FOR VISUALIZING LARGE DIRECTED ACYCLIC GRAPH ONTOLOGIES

Getiria Onsongo
Macalester College, St. Paul, MN

The Gene Ontology (GO) database is a multi-species data resource containing gene descriptions and categorizations, which are represented as very large directed acyclic graphs (5000-7000 nodes). When displaying this information, existing tools distort the structure of the GO graphs by displaying them as trees. Explicit information about the children with multiple parents is lost when using a tree to represent these graphs. Moreover, the size of each DAG requires the use of collapsable trees in these tools, thus hiding many relationships between terms. Visualizing the ontologies as graph structures will enhance biologists' abilities to understand and explore these ontologies.

We implemented two techniques for displaying large graphs for the GO database. The first approach introduces the use of multiple views. One view presents the context view, a global view of the entire graph, while the other views are used to explore the graph in detail. The second technique uses fisheye views, a term commonly used to describe the use of distortion techniques to simultaneously display both context and detail in one view. While these techniques have been used in the past for data visualization, they have not been used in combination on graphs of this size. This new visualization tool will enable both biology researchers and biology students to more easily use the GO database for scientific inquiry and learning.

MAGMATIC RESURFACING ON VENUS

Chris Orth and Chris Reese
Univ. of Minnesota, Morris, MN

The focus of this project is the unique magmatic evolution of Venus. NASA's Magellan mission to Venus revealed a global resurfacing event which took place ~500 million years ago. According to analysis of the impact crater record, the age of the surface of Venus is between 300 and 1000 million years. Various theoretical models have been proposed to explain this global resurfacing event. One possibility is that the resurfacing was caused by widespread volcanism due to a late onset of mantle convection beneath an immobile surface. This hypothesis is explored using physical models with constraints from observational data to calculate thermal evolution scenarios for Venus.

EXPRESSION OF MITOCHONDRIAL GENES IN TRITICEAE

Karunadharma Pabalu,[1] Hossain Khwaja, Maan Sidambaran, and Kianian Shahryar[2]
[1]*Dept. of Biology, MN State Univ., Moorhead; MN,*
[2]*Wheat Germplasm Enhancement Program, Department of Plant Sciences, North Dakota State Univ.*

Nuclear and organelle genomes in plants play and important role in expression of productivity traits. The subcellular genomes—mitochondria and chloroplast—code for only a small number of genes but they are unique and irreplaceable for the regulation of the cellular processes in plants. Mitochondrion is the center of energy synthesis and serves essential functions in the development of the plant. Mutations of mitochondrial genes lead to changes in development of the plant. Mutations of mitochondrial genes lead to changes in development such as cytoplasmic male sterility (CMS) which is observed in as many as 150 plant species. Also there are a number of nuclear encoded sequences that affect mitochondrial and ultimately organismal functions. Wild species are the usual reservoir of genes for improvement of pest resistance, grain quality, and agronomic fitness of any cultivated species. The genes affecting nuclear-cytoplasmic (NC) interactions are believed to influence the success of gene transfer from wild to cultivated species.

Analysis of gene expression in mitochondria in euplasmic and alloplasmic lines of wheat will provide valuable information in the understanding of these NC interactions in the cell. The purpose of this study is to identify mitochondrial genes, isolate them, and compare them with other mitochondrial genomes from grass species such as rice and maize. The ultimate goal of this project is to design and develop a microarray system for characterization of mitochondrial gene expression in various alloplasmic and euplasmic plants and their organs.

IDENTIFICATION OF PROTEINS INTERACTING WITH ACTIN CAPPING PROTEIN ALPHA SUBUNIT ISOFORMS IN MURINE MYOCARDIUM

Ryan Peck and M.C. Hart
Dept. of Biological Sciences , MN State Univ., Mankato, MN

Actin, an essential component of all living cells, contributes to cell shape, cell motility, and the integrity of specific structures such as the sarcomere of striated muscle. Actin molecules are rod-like in structure, and are composed of monomers that attach and disassociate in a dynamic process. Capping protein (CP) anchors actin filaments, regulates the length of actin molecules, and maintains a cell's ability to contract and relax. CP is a heterodimer composed of an alpha and beta subunit. There are three forms of the alpha subunit (α1, α2, and α3) and three forms of the beta subunit (β1, β2, and β3). Previous studies indicate that β1 and β2 have unique

localizations and functions within murine myocardium. Recent studies have shown that α1 and α2 co-localize in myocardium but have distinct localizations in skeletal muscle cells. This early evidence suggests that α1 and α2 may also have novel functions within muscle cells and may interact with novel proteins.

The goal of this research is to identify cellular proteins that interact with α1 and α2 utilizing coimmunoprecipitation. This was accomplished using previously generated polyclonal rabbit anti-α1 and chicken anti-α2 antibodies. The antibodies were purified and chemically attached to an immobilized matrix. The matrix is currently being used to identify novel proteins that interact with α1 and α2. The results will be analyzed by Sodium Dodecyl Sulfate Polyacrylamide Gel Electrophoresis, native gel electrophoresis, and Western Blot analysis.

MOSQUITOES AS POLLINATORS? A PILOT STUDY

Heather Rickerl, Jen Hatton, Luke Ouradnik, and Alison Wallace
Dept. of Biology, MN State Univ., Moorhead, MN

What good are mosquitoes? When people hear the word "mosquito," they tend to think of blood-sucking insects. In reality, the main nutritional source for mosquitoes is nectar. An Internet search revealed site after site claiming mosquitoes are important, even vital, pollinators for plants. An exhaustive search of the primary literature produced one published paper on *Habenaria obtusata*, a bog orchid species which relies on mosquitoes for pollination. L. T. Nielsen, a retired Utah professor, found pollen from over a dozen plant families on mosquitoes he trapped for his research on mosquitoes as disease vectors.

We decided to investigate the possibility that mosquitoes might be pollinators of plants in our region. To do this, we used a CDC Miniature Light Trap from July through September 2003. Our basic goal was to trap mosquitoes weekly throughout the summer and look for evidence of pollen on their bodies. We experimented with two trap sites (city backyard and mesic prairie) to investigate whether or not this type of trap would work for our purposes, and varied our methods of killing mosquitoes to see which method yielded mosquitoes whose bodies were largely intact after death. Out of 1337 mosquitoes trapped over the course of the summer, pollen grains were found on one male mosquito of the species *Aedes vexans*, making mosquito pollination a very elusive, yet possible, occurrence in nature. Future plans include implementing more rigid protocols and trapping more mosquitoes in a variety of sites to look for evidence of mosquito contact with pollen.

EFFECTS OF EXERCISE AND HYDRATION STATUS ON URINARY CAFFEINE CONCENTRATIONS

Kateri M. Roessler (Advisor: Amy Olson)
Dept. of Nutrition, College of St. Benedict, St. Joseph, MN

Caffeine is an effective ergogenic (performance-enhancing) aid at doses of 3-5 mg caffeine per kilogram body weight (roughly the amount in 2-3 cups of coffee). However, the NCAA imposes a legal limit for urinary caffeine concentration at 15 mg/mL. The amount of oral caffeine that results in exceeding this limit and the effects of exercise and hydration on caffeine excretion are unclear. Current work was primarily performed on male subjects in controlled laboratory settings. Recently world-class athletes, including runners Letitia Vreisde and Inger Miller, had gold medals taken away after drug tests showed urinary caffeine levels exceeding the legal limit.

This study focused on healthy female college cross-country runners and examined hydration status in addition to exercise. Urinary caffeine concentrations were assessed after ingesting 5 mg caffeine per kilogram body weight both with exercise (and subsequent mild hydration change) and without exercise. In both trials, subjects ingested 1 L of caffeinated sports drink. Drinks were prepared so each subject received 5 mg caffeine per kilogram body weight. In the exercise trial, subjects ran outside under normal training conditions for 60 minutes, one hour after ingestion. Urinary caffeine concentrations were assessed using Liquid Chromatography Mass Spectroscopy.

NA^+–H^+ EXCHANGERS ARE ACTIVATED BY PHENYLEPHRINE VIA PROTEIN KINASE C ISOFORMS THAT DIVERGE INTO BIFURCATING PATHWAYS INVOLVING RHOA AND ERK

David S Ronderos, Anusha Mishra, Joseph Provost, and Mark Wallert
Dept. of Biology, MN State Univ., Moorhead, MN

In Chinese hamster lung fibroblasts (CCL39), phenylephrine (PE) activates both ERK and the Na^+–H^+ exchanger (NHE) to regulate stress fiber formation. PE activation of $\alpha_1$adrenergic receptors activates conventional isoforms of protein kinase C (PKC).

Previous research from our laboratory indicates that PE addition leads to activation of RhoA in CCL39 cells. Additionally, general PKC inhibitors such as staurosporine and BIM have been shown to block both RhoA and ERK activity in cells treated with PE. Multiple PKC isoforms are differentially regulated by a variety of cell membrane receptors to control diverse cellular functions.

The focus of this study was to determine which PKC isoform(s) are involved in the PE activation of RhoA and ERK. Using enhanced green fluorescent protein tagged-PKC isoforms, we investigated the ability of PE to stimulate PKC translocation using the conventional PKC isoforms α, β_1, β_2 and γ. Our

experiments show that PE activates multiple PKC isoforms. This finding allows for the possibility that distinct PKC isoforms are responsible for the independent activation of ERK and RhoA. Dominant/negative PKC constructs and specific PKC inhibitors are also used to examine the potential role for different PKC isoforms in the regulation of the RhoA-ROCK pathway and the ERK pathway. Our research has also shown that activation of ERK, RhoA, and NHE are all required for stress fiber formation by PE in CCL39 cells. Defining a role for multiple PKC isoforms in the regulation of stress fiber formation would dramatically improve our understanding of this process. This work was supported by a grant from the NSF MCB-0080243, DUE 0088654, and MRI-DBI 0110537, and MSU Moorhead Faculty Grant.

POINT MUTATIONS WITHIN WATERMELON GLYCOXISOMAL MALATE DEHYDROGENASE ACTIVE SITE AND THEIR EFFECTS ON SUBSTRATE-BINDING EFFICACY AND SPECIFICITY

Castel Santana, Andrew Thompson, James Denker, and Joseph Provost
Depts. of Biology and Chemistry, MN State Univ., Moorhead, MN

Malate Dehydrogenase (MDH) is an enzyme that is involved in the pathways of the Krebs Cycle, carbohydrate, fatty acid, and amino acid metabolism. The role of MDH is to catalyze the reduction of oxaloacetate (OAA) to malate via oxidation of NADH to NAD+. Lactate Dehydrogenase (LDH) is a glycolitic pathway enzyme, which catalyzes the conversion of pyruvate to lactate. Upon examination of the amino acid sequence of a watermelon MDH isoform, we found that its active site amino acid residues 102 and 171 are conserved. When the sequences of this isoform is aligned with that of *Bacillus stearothermophilus* LDH (BsLDH), differences were found at or near these key sites (102:MDH-arginine, LDH-glutamine; 170: MDH-valine, LDH-alanine; 172: MDH-alanine, LDH-phenylalanine).

The goal of this project is to shift the substrate specificity of a watermelon MDH isoform through saturation mutations, which should result in nearly every possible amino acid substitution at each of these key sites. A shift in substrate specificity from OAA to pyruvate will, in essence, innovate the function of MDH into LDH. Mutants of the watermelon MDH will be constructed using the Stratagene Quickchange mutagenesis kit employing degenerate oligos with a highly efficient, long-range polymerase to create site-directed mutants for the watermelon glycoxisomal isosyme. The resulting mutants will be assayed for specific enzyme/substrate interactions (MDH functioning vs. LDH functioning). We will develop a nitrocellulose filter assay system or, alternatively, we will create a stop-time, spectrophotometeric enzymatic assay to measure the catalytic rates of the reactions. Once mutation has been obtained, a Sanger-dideoxy DNA sequencing reaction will be performed in order to confirm the amino acid changes made to the resulting mutants, and the specific kinetic changes in the mutants will be measured.

USING GENOMICS TO DEVELOP A FISH EXPERIMENTAL MODEL OF TUBERCULOSIS

Erica Savage
St. Olaf College, Northfield, MN

Currently one-third of the world's population has latent tuberculosis, and this stage of the life cycle is of extreme importance to epidemiologists. Microarray work by our collaborators has identified the "Dormancy Regulon," a group of 50 bacterial genes upregulated in response to low oxygen conditions suspected to play a role in latency.

Here we describe our work establishing an *in vivo* experimental model of tuberculosis using zebrafish and the mycobacterium pathogen *Mycobacterium marinum.* As an initial step, we have established an infection protocol and have visualized *M. marinum* infection of zebrafish in real time using the fluorescent G-13 bacterial strain. To test the role of the Dormancy Regulon in our fish model, we datamined the incomplete *M. marinum* genome project and identified homologs for most (38 of 50) of the "Dormancy Regulon" genes. We then isolated promoters for the homologs of the dormancy genes Rv2031, Rv2626, Rv2627, and Rv3130 using PCR. Cloning of these fragments into a GFP (green fluorescent protein) expression construct is currently underway.

THE NUCLEAR EXCHANGE JUNCTION OF *Tetrahymena thermophila:* 1. THE DEVELOP-MENT OF AN ISOLATION PROTOCOL FOR FUTURE MOLECULAR AND MORPHO-LOGICAL CHARACTERIZATION 2. "FENESTRIN" CHARACTERIZATION AND THE EXCHANGE JUNCTION OF bcd MATING CELL LINES

Johanna Savage
Dept. of Biology, St. Olaf College, Northfield, MN

The understanding of membrane fusions represents one of the great biological enigmas of our time. Membranes are defined by their definitive function to isolate one structure or organism from another. Hence, the occurrence of membrane fusions presents a unique and mystifying paradox.

The present study focuses on the membrane fusion event of conjugation (sexual reproduction) in *Tetrahymena thermophila.* Two separate, but related, studies focused on the isolation of the conjugation junction (nuclear exchange junction) and its subsequent characterization in *bcd* mutant cells. With regard to the first study, isolation of the nuclear exchange junction was successful using a 0.12% Triton X-100/50% Ethanol fixation protocol followed by ultrasonication. Second, using "fenestrin" as an analytical tool, our observations suggest that there are no significant developmental differences between the exchange junctions of wild type

and *bcd* mating cells. Both studies present preliminary conclusions for the development of future studies.

PRIMARY ATTACHMENT AND METASTASIS 24-48 HOURS POST INJECTION USING EGFP IN B16 MELANOMA

Matthew D. Schwartz (Advisor: Dr. William Heidcamp)
Gustavus Adolphus College, St. Peter, MN

Previous studies have shown it is possible to tag B16 mouse melanoma cells with vital stains to study the attachment, growth, and metastasis of tumors. These studies used fluorescent cytoplasmic or membrane-bound tags and were useful for only the first few hours post injection before the signal became too dim to detect, with cell division. Other studies on metastasis use colony formation after one to two weeks, too late for detection of the primary attachment of melanoma cells to the endothelial lining of lungs.

A transfected GFP B16 cell line was created in our labs. Preliminary testing of the transfected cell line allowed FACS sorting, but the intensity of the fluorescence was too low for epifluorescence. Subsequent retransfection with EGFP produced a clone with enough intensity for use with epifluorescence. This newly created metastatic clone is used to study primary attachment of B16 tumor cells in vivo 24-48 hours after tail vein injection, a time during which the components of the extracellular matrix (ECM) are most important.

INVESTIGATION OF MDH FUNCTION USING SITE-DIRECTED MUTAGENESIS

Sarah Y. Skolness, Austin J. McCoy, Jessica N. Heck, and Joseph Provost
Dept. of Biology, MN State Univ., Moorhead, MN

Malate dehydrogenase (MDH) plays an important role in the citric acid cycle where it catalyzes the reversible oxidation of malate to oxaloacetate. MDH is found in all eukaryotic organisms as it is important for the production of ATP, and thus, mutations in the amino acid sequence of MDH can alter its performance and function in the cell.

The purpose of this study was to create several point mutations in various MDH isoforms. These constructs will be used to investigate the importance of particular amino acids in the overall function of MDH. Three different MDH clones were used: yeast mitochondrial MDH and MDH isolated from *E. coli*. Cultures of each MDH clones were grown and plasmid DNA was isolated. Mutagenesis was performed using PCR and the Stratagene Quickchange kit. The final result was the production of three different MDH clone mutants: pYmMDH-168I, pYmMDH-168E, and pE_MDH-R81I. These mutants will be used for further studies with MDH function.

AN INITIAL BIOCHEMICAL ANALYSIS OF AUTISM

Jill M. Skolte, Moses K. Wananu, Joseph J. Provost, and Mark A. Wallert
Dept. of Biology, MN State Univ., Moorhead, MN

Autism is a pervasive developmental disorder with a collection of behavioral symptoms including dysfunction in social interaction and communication in affected children. Autism is associated with sensory disturbances, obsessive-compulsive-like behavior, lack of bonding to caregivers, and motor disturbances.

We have obtained four lines of lymphocyte cells, two from children with autism and two from children without autism. We are going to test them for several proteins which may be altered in children with autism. While there is little understanding of the biochemical basis for the cause of autism, one phenomenon of this disorder is the formation and development of neural synapses. A significant percentage of people with autism display chromosomal alterations in chromosomes 9 or 15. The genes associated with these abnormalities code for two proteins, called hamartin and tubarin. These proteins are very closely related and when functioning normally, regulate the small G-protein RhoA. RhoA is an important signaling molecule which regulates cytoskeletal structure, important for cell growth and development. RhoA also activates the sodium-hydrogen exchanger (NHE), and NHE may act as an anchor for cytoskeletal proteins. Thus alteration in either RhoA or NHE would significantly impact the development of neural cells as they mature. We intend to test for RhoA activation levels in our cells and for NHE activity. With this research, we hope to gain an understanding of one potential cause of autism.

VENTILATORY PHENOTYPES BETWEEN TWO STRAINS OF NEONATAL RATS

Gregory Solheid
Augsburg College, Minneapolis, MN

Phenotypic strain differences establish a foundation for physiological genomic studies aimed at elucidating the genetics of chemoreception and control of breathing. This study aimed to determine if there were different ventilatory phenotypes between two inbred strains of neonatal rats: Brown Norway and Dahl salt sensitive. Ventilatory responses to eupneic breathing and hypercapnia using 6-7% CO_2 were measured.

The research done on these two strains of neonatal rats shows that there are no differences between ventilatory phenotypes in the Brown Norway and the Dahl salt sensitive rats. Neither the Brown Norway rats nor the Dahl salt sensitive rats had the presence of a critical window. However, both strains did have an increasing ventilatory response to CO_2 as they aged, and the ventilation in both strains began to steady by day 21. The early postnatal exposure to high CO_2 concentrations did not have any carryover effect in ventilation on days 30 and 40 in either strain. However, the ventilation in both strains at day 40 was significantly different from the

normal ventilation for the adult rats, as previously determined.

SOCIALIZATION CONFLICTS IN GIFTED WOMEN AND THE IMPACT OF THE COURSE "PSYCHOLOGY OF GIFTED WOMEN" ON HONORS STUDENTS

Alicia Stachowski, Khulan Altangerel, and Dr. Zoa Rockenstein
St. Cloud State Univ., St. Cloud, MN

Women continue to be seriously underrepresented in leadership positions and occupations requiring high intellectual ability, despite the fact that they compose 50% of the intellectually gifted population. The social construction of gender fails to take intellectual ability into account, socializing gifted women for jobs and positions that result in their underachievement. This considerable lost-opportunity cost to the individual and society is a serious problem that needs to be addressed in higher education.

The following two questions guided our research: (1) Is there a difference between Honors and non-Honors women's attitudes concerning (a) academic achievement, (b) future perceptions of potential family/career conflict, and (c) gender equity? (2) Can a one-semester Honors class that addresses these issues make a significant difference in the attitudes of Honors women toward (a) awareness, and fullfillment, of their intellectual ability, (b) the potential for future career/family conflict, and (c) gender equity?

The questionnaire Students' Perceptions of Academic Ability and Family Career Socialization was created, based on a review of longitudinal studies on gifted women. This questionnaire was administered to Honors and non-Honors women in a pre/post control group design between the years 2000 and 2003. Results suggest that (1) some significant differences exist between non-Honors and Honors women; (2) while a one-semester class does make a significant difference in awareness of these issues for Honors women, more time and education may be needed to change attitudes formed through two decades of gender socialization.

SUPERANTIGEN PRODUCTION BY *Staphylococcus aureus*

Kristi Strandberg and Patrick M. Schlievert
Univ. of Minnesota Medical School, Minneapolis, MN

Staphylococcus aureus strains may make one or more of a family of superantigens; includes staphylococcal enterotoxins (SEs) and toxic shock syndrome toxin-1 (TSST-1). These toxins bind to class II major histocompatibility complex molecules and the variable parts of the beta chains of T cell receptors. The consequence is massive cytokine release from both T cells and macrophages with immune dysregulation; TSS is the severest manifestation.

This study was undertaken to evaluate 39 *S. aureus* strains for the presence of 16 superantigen genes through polymerase chain reaction (PCR) amplification after preparation of chromosomal DNA. All of the strains were positive for at least one superantigen. Unexpectedly, all strains contained the genes for SEH and J. Eleven of the strains were verified to contain the SEH gene by nucleotide sequencing. The gene for TSST-1 and SEs B and C, the causes of nearly all cases of TSS, were present in 2/3 of the strains, indicating strains capable of causing TSS are common. Two strains contained the TSST-1 and SEB genes, combinations not seen before this study. Collectively, the data indicated that all *S. aureus* strains contain superantigen genes, and the distribution determines the type of human illness that can be produced.

IS MITOCHONDRIAL INHERITANCE TISSUE-SPECIFIC? A NEW LOOK AT THE mtDNA DOGMA FROM A CELL BIOLOGY PERSPECTIVE

Lisa Streitz and Ellen Brisch
Dept. of Biology, MN State Univ., Moorhead, MN

Mitochondria play critical roles in the generation of metabolic energy (ATP) in eukaryotic cells. ATP is essential in driving many of the reactions that take place in the body. The role of a mitochondrion is to maximize and control the production of ATP. Furthermore, these cytoplasmic organelles make their own circular DNA, which is referred to as mitochondrial DNA (mtDNA). It is important to note that there is a distinction between nuclear DNA and mtDNA. While nuclear DNA encodes most of the proteins that drive mitochondrial processes, some critical ATP-producing enzymes are encoded in the mitochondrial genome. Mitochondria are extremely important to study because almost any mutation in mtDNA leaves an organism somewhat debilitated, by causing mitochondrial myopathy. Mitochondria have been thought to be maternally inherited for over 20 years. Results from previous experiments show that a child's mtDNA will be identical to that of the mother. Does this mean that there is no paternally inherited DNA? Perhaps not— however, researchers have mainly focused on testing mtDNA in blood samples.

To examine if inheritance patterns differ between tissues, Heidi Jo Johnson, Austin McCoy, and Jen Risan began planning an experimental approach and protocol development to test our hypothesis. Our hypothesis is that blood and muscle tissue will inherit mitochondria from different parents. The approach I am using to test my hypothesis is to sequence the mtDNA taken from two different strains of mice. Next, I plan to cross the parent mice and sequence the mtDNA of their offspring. I will be sequencing mtDNA from the blood as well as from the muscle tissue to see if mtDNA inheritance is, indeed, tissue-specific. Currently, I am working on mitochondrial isolation from different tissues and mtDNA extraction. Verifying the specificity of mitochondria is an important step for figuring out what cellular mechanisms are required to direct the mitochondria into different tissues.

This may open a whole new way of looking at mitochondrial inheritance and ultimately show us how this system is regulated.

THE AMES SALMONELLA ASSAY VERSUS A T4 BACTERIOPHAGE ASSAY FOR MUTAGENICITY

Emily Strom (Advisor: Dr. Richard Kowles)
Saint Mary's Univ. of Minnesota, Winona, MN

The Ames Salmonella test, set forth by Bruce N. Ames, is a widely accepted technique for the testing of substances for mutagenicity. Testing has been done, however, toward the development of a different technique for determining the mutagenicity of substances. The use of the bacteriophage T4 is more convenient than the Ames Salmonella test, which can be expensive and laborious. An assay based on reverse mutations of T4 virus has been successful in determining the mutagenicity of the known mutagens, sodium azide and an acridine substance.

Testing conducted with the Ames Salmonella assay for mutagenicity of sodium azide showed a reverse mutation rate of 8.6×10^{-6}, while the T4 virus assay provided comparable results with a reverse mutation rate of 1.45×10^{-6}. The acridine mutagen, on the other hand, while showing a reverse mutation rate of 2.1×10^{-4} in the Ames Salmonella test, showed a reverse mutation rate of only 1.33×10^{-6} using the T4 virus assay. This reduced rate of mutation may be explained by the fact that acridine causes a frameshift mutation, which is compatible with the Salmonella TA98 strain, while the T4 virus seems to be more sensitive to base substitution mutation. The T4 virus assay accomplished a reversion rate greater than that of a negative control in both cases. The spontaneous reversion rate was 6.15×10^{-7}. Testing of the T4 bacteriophage assay is presently being conducted on other putative mutagens.

THE BIOLOGY OF BEHAVIOR: EFFECTS OF THE *mPer2* GENE ON CIRCADIAN RHYTHMS IN MICE

Lisa N. Tracy and Dwight E. Nelson
Dept. of Biology, Univ. of St. Thomas, St. Paul, MN

Several "clock" genes interact to form the mammalian circadian pacemaker, located in the neurons of the hypothalamic suprachiasmatic nucleus. The pacemaker is entrained to environmental light through a photic entrainment pathway. The interactions of the genes in the pacemaker (*Clock, BMAL1, Cryptochrome,* and *Period* genes) form a negative feedback loop that oscillates with a period of approximately 24 h. Outputs of the hypothalamic pacemaker are thought to drive or synchronize all other 24-h oscillations of the body, including body temperature, metabolism, and wake/sleep cycles. There are three mammalian period genes: *mPer1, mPer2,* and *mPer3*.

We are investigating the functional importance of the *mPer2* gene in the mammalian photic entrainment pathway by examining circadian phase resetting in *mPer2* mutant mice. To do this, we established a colony of mice from a pair of *mPer2* "knockout" mice from the laboratory that produced the mutation. These mice have no ability to make functional *mPer2* proteins. We back-crossed several generations with c57Bl/J mice. In order to maintain the colony, we developed a genotyping assay for the *mPer2* knockout gene. Using the rhythm of locomotor activity and the functional assay of the photic entrainment pathway (brief light pulses to induce phase shifts) we were able to examine the functional output of this pathway.

Experiments by other labs suggest that the *mPer2* gene is necessary for normal circadian rhythmicity and is essential for normal phase delays (resetting) of the circadian pacemaker during entrainment or synchronization of circadian oscillator to environmental light cycles. Our data show different results. A preliminary analysis of our data shows that in constant darkness, *mPer2* mutant mice displayed shorter circadian period that wild-type littermate controls. Our results also show that although some *mPer2* mutant mice display arrhthmicity, many remain rhythmic. From the rhythmic mutants, we found that the ability to phase delays is maintained and the magnitude of the light-induced phase shift may be even larger than in the wild-type controls. While *mPer2* mutant mice have a much shorter circadian period, neither the ability to maintain rhythmicity nor the ability to phase delay is abolished.

DENDROCHRONOLOGICAL ANALYSIS OF THREE YOUNG *Pinus* Species AT ST. OLAF COLLEGE

AmberBeth VanNingen and Kathleen Shea
St. Olaf College, Northfield, MN

Dendrochronology, the study of tree rings, is a useful tool in recreating climate histories or looking at the growth patterns of trees. Few studies have focused on the growth of young trees.

The objective of this study was to use the ring patterns in tree cores to examine the early growth patterns in three species of pines, *Pinus banksiana, P. resinosa,* and *P. strobus*. The trees were planted in 1993 as part of a conifer restoration progam at St. Olaf College, Northfield, MN. No significant relationship existed between precipitation and tree-ring width. This result is likely because the trees were still very young and there have not been extremes in temperature or precipitation in s. Minnesota over the last 10 years. The rapid growth of the trees is expected to slow down as they reach reproductive age and become large enough to compete with neighbors. This study showed that dendrochronological methods used for older trees can be applied to younger trees, with certain precautions.

HAND-RAISING IN AN ELECTRONIC LEARNING ENVIRONMENT: ENHANCING ONLINE INTERACTIONS THROUGH THE IMPLEMENTATION OF A MODERATED SYNCHRONOUS DISCUSSION SYSTEM

George Veletsianos
Macalester College, St. Paul, MN

Networked technologies such as the World Wide Web have had a profound impact upon teaching and learning. This study investigates the use of "chat" software in education as a tool to support, enhance, and advance learning. Even though instructors use such software to facilitate class discussions on an equal basis, encourage group interaction, and build learning communities, the use of generic computer-mediated communication software is not a panacea. Educational chat rooms that do not impose a hierarchy of power may suffer because of interactional anomalies and aggressive students who dominate the discussions.

To cater for these imperfections, we used Java to implement a moderated online learning environment that closely resembles classroom interactions. Subsequently, we evaluated our software in one Economics and one Computer Science class and received feedback from students as to the efficacy of our implementation. The importance of this project lies on two facts: (a) The existing literature recognizes the problems of teaching and learning in a synchronous online environment but does not evaluate the use of instructor-led chat rooms as an alternative to solve the problems, and, (b) contrary to existing solutions, this software will be made freely available to educators and other interested individuals via the web.

THE PLANT ENZYME, PYRUVATE, ORTHOPHOSPHATE DIKINASE (PPDK), CURIOUSLY FUNCTIONS BOTH AS A PHOTOSYNTHETIC PATHWAY ENZYME AS WELL AS A SEED METABOLISM ENZYME

Dylan G. Voge, Thomas A. Colquhoun, and Chris J. Chastain
MN State Univ., Moorhead, MN

Historically speaking, the plant enzyme pyruvate,orthophosphate dikinase (PPDK) was initially discovered as a cardinal enzyme in the C_4-photosynthetic pathway. Its role in C_4 photosynthesis is well elucidated, including its light/dark- regulation via site-specific reversible phosphorylation. In contrast, the presence of PPDK in C_3 plants, where it is of non-photosynthetic function and usually of low abundance, was not established until much later. Further, research into the function and regulation of PPDK in C_3 plants has received only scant attention and remains for the most part unexplored.

Recently, we documented that PPDK in developing seeds of rice, a C_3 plant, is highly abundant and undergoes light/dark-induced changes in phosphorylation state in a manner similar to C_4 leaf dikinase. For this presentation, we examined mature seeds of several species of C_3 and C_4 plants to see if PPDK was (1) similarly abundant and (2) phosphorylated as in rice seeds. Our preliminary results indicate that PPDK is abundant in cereal seeds such as corn, wheat, and oats, but is in low abundance in non-cereal seeds such as spinach and cotton. Phosphorylation of PPDK was shown to occur only for rice seeds.

THE EFFECTS OF EGG INCUBATION TEMPERATURE ON SWIMMING SPEED OF WOOD FROG TADPOLES

Tim Watkins and Julia Vraspir
Dept. of Biology, Macalester College, St. Paul, MN

Female wood frogs (*Rana sylvatica*) lay their eggs in the same location within a pond and thereby create clumps of eggs that may contain hundreds of egg masses. The middle of a clump is warmer than the periphery, by as much as 7°C. Differences in egg incubation temperature of this magnitude are known to affect several physiological and behavioral traits of reptiles, including locomotor performance. Little is known about such effects in anurans, but the one published study of frogs (Parichy and Kaplan 1995) indicates that cooler egg incubation results in faster hatchlings.

We sought to determine whether egg incubation temperature affects swimming speed in wood frog tadpoles, and if the temperatures that tadpoles subsequently experience during development modify such effects. We reared wood frog eggs at 15 and 21°C until they hatched, and then reared tadpoles from both egg treatment groups at 15, 18, and 21°C until they reached a late larval stage. Burst swimming speed over a range of test temperatures was determined from video records. Tadpoles that both incubated and developed at 15°C were larger and faster than those that spent their whole lives at 21°C. Among tadpoles that developed (post-hatching) at 18°C, there was no difference in speed between those that had incubated as eggs at 15°C vs. 21°C. There was a significant effect of incubation temperature on body size, however.

SYNTHESIS OF ENANTIOMERICALLY PURE TRISPHENOL FOR ENANTIOSELECTIVE METAL CATALYSTS

Charles J. Weiss and Gretchen E. Hofmeister
Carleton College, Northfield, MN

Our group has been prepg. and studying titanium and aluminum complexes of trisphenols, and evaluating their selectivity as catalysts for org. transformations. We have previously prepd. a chiral trisphenol and detd. that the stereocenter controls the conformation adopted by the trisphenolate ligand, when it is coordinated to titanium. Our current research concerns the prepn. of enantiomerically pure trisphenol, by resolving a chiral bisphenol intermediate in the synthetic sequence. Thus far, we have prepd. primarily one enantiomer (ratio) of

the precursor bisphenol. Once we have the enantiomerically pure bisphenol in hand, the final coupling reaction to produce enantiomerically pure trisphenol should enable us to prep. enantioselective metal catalysts.

METHOD DEVELOPMENT FOR THE QUANTIFICATION OF 2-PHENYLMETHO-XYETHANALAND METABOLITES FOR PHARMICOKINETIC STUDIES WITH ALDH

Emily Wessel
St. Cloud State Univ., St. Cloud, MN

In cellular systems, the enzymatic conversion of aldehydes to acids by aldehyde dehydrogenase (ALDH) may be accompanied by a myriad of other metabolic pathways, resulting in a complex set of metabolites for a given aldehyde. Analytical methods are needed for the characterization and quantitation of these metabolites.

In this study, 2-phenylmethoxyethanal was chosen as a model aldehyde. Preliminary studies have been conducted for the quantitation of the aldehyde using high-performance liquid chromatography (HPLC), and gas chromatography–mass spectrometry (GC-MS) combined with solid-phase microextraction (SPME). HPLC offers the advantage of compatibility with a wide range of potential metabolites of varying polarity and molecular weight; however, the ultraviolet absorbance detector on the HPLC is not sensitive enough for low concentrations. SPME/GC-MS is much more sensitive; however, it is limited to the analysis of relatively non-polar, low-molecular-weight metabolites. Both of these techniques have been demonstrated to be amenable to the analysis of the aldehyde itself. Liquid chromatography–mass spectrometry (LC-MS) appears to hold the greatest promise as the most sensitive and universal method for the characterization of 2-phenylmethoxyethanal metabolites.

ANALYSIS OF MOLECULAR VARIANCE (AMOVA) OF Y CHROMOSOMAL SHORT TANDEM REPEATS (Y-STRS) TYPED IN FORENSIC SCIENCE INVESTIGATIONS

Heather Wheeler
Dept. of Biology, Hamline Univ., St. Paul, MN

Y chromosomal short tandem repeats (Y-STRs) have recently been employed in forensic investigations when crime scene samples contain minimal amounts of male suspect DNA mixed with much greater amounts of female victim DNA. If variation due to race or region in Y-STR data is observed, separate racial or regional databases will allow conservative frequency estimates when results are reported in court.

Analysis of molecular variance (AMOVA) was used to estimate the variance contributions between regions (Connecticut and Minnesota), among races within regions, and within races (African American, Caucasian, and Hispanic). Most of the genetic variation (94.0%) was individual variation within races. A smaller (8.5%), yet still significant, amount of variation was among races, necessitating separate databases divided by race to report the most conservative estimates for forensic applications. Because no regional variance was observed, the Minnesota and Connecticut data can be combined within racial groups.