# **ERGO**

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#### LETTER FROM THE EDITOR

"Research is creating new knowledge."

Neil Armstrong

I am happy to present this year's Ergo. I appreciate having a journal where the students can submit their scientific analysis of the world around us for publication; research prose is just as important as creative prose. In our fourth year of publication, Ergo is still working to get off the ground, but I hope in the years to come, it will become just as popular as Metaphor.

This year's submissions made it very difficult to choose what to include in the journal. I learned a lot from reading the papers, and I am impressed by the amount of work that went into them. I hope that the students whose papers were not selected will rewrite them for submission next year, as the research was done well if not presented properly.

I would like to thank Dr. Cavitt for the opportunity to help create this year's journal; Amy Douangdara for her tireless support; Valerie Frokjer for stepping in when needed; my staff for getting things done on time despite our few numbers. I would also like to thank the liaisons for their help in ensuring that all the submissions were reviewed. Special thanks go out to the reviewers; both faculty and student, for helping me decide which articles to include. I know it was a difficult job.

Kirsty Winkler *Editor-in-Chief* 

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FEATURED ARTICLES	

### JOHN B. GODDARD SCHOOL OF BUSINESS & ECONOMICS ECONOMICS

## Contributing Factors to Utah Regional Income

Jeremy Ward & Doris Geide-Stevenson

#### **ABSTRACT**

This paper explores economic factors that contributed to per capita income in Utah counties during the 2000 census. The study utilizes a combination of neoclassical, urban and regional economic theories to establish income determinants at county levels. An ordinary least squares regression indicates that (1) average education has a significant positive impact on income, (2) distance from large city has a negative impact on income, and (3) capital levels have a positive impact on income, but are found to have the smallest impact on income. Labor factors are found to have weak relationships with income. Furthermore, the study finds that higher education has the greatest impact on income.

#### INTRODUCTION

In 2000, the Census Bureau reported that the richest county in Utah, Summit County, had \$33,767 per person in income, while San Juan County, the poorest county, had only \$10,229 per person (U.S. Census Bureau 2009a). Residents in Summit County had 3.3 times more income than residents of San Juan County; a similar inequality was found between the U.S. and Mexico in 2008 in purchasing power parity terms (World Bank 2009). As both Summit and San Juan Counties are in the same state it can be assumed that they have similar cultures; laws and institutions are reasons often cited as causes of income inequality (Olson 1996). There must be other factors that vary within the state that cause income levels to be so dramatically different within Utah. This study will investigate what these possible factors could be by looking at county cross-sectional data for Utah from the 2000 Census.

Special focus of this study will be the impact of education on income at county levels. This variable merits special attention because while

attending school an individual is not as productive as they could be if they were working full-time and they often require state assistance to fund schooling. In Utah, the enrollment in public higher education institutions accounted for more than 5% of the population in 1999 (Economic Report to the Governor 2009). Higher education cost Utah taxpayers \$618,114,400 in 2003, equating to about \$4,500 per student (Utah Board of Regents 2009 & Economic Report to the Governor 2009). This study will also investigate what impact education had on county level per capita income in 2000.

Factors contributing to regional income variation can be explained using both neoclassical and regional economic growth theories. Neoclassical economic growth theory states that growth and income of an economy are functions of the quantity and quality of labor, and capital available within an economy (Dornbusch 2007). Research has found mixed results regarding the importance of labor and capital in regional models. Moomaw (2002) and Glaeser (1995) have found that they are irrelevant; while Spieza (2007) finds evidence that they are important and should be included.

Labor involves a quality factor referred to as human capital and human capital theory establishes that as individuals invest resources in activities that improve their productive capacity, they will increase their marginal productivity and their income (Becker 1962). Regional application of human capital theory suggests that regions with higher levels of education will enjoy higher levels of per capita income, and existing regional research supports such postulation. Findings also point out that average education level is more important than total education and that education has the largest impact on income at regional levels (Baldwin, 2008; Erickcek, 2006; Gleaser, 1995; Moomaw, 2002; Spieza, 1995; Vachal, 2005). While some evidence suggests that education has a negative impact on income, this result is likely caused by the population that was sampled; rural communities struggling with brain drain (Huang 2002) and elderly retired individuals no longer earning a significant income (Kim 2004).

Urban and regional economies are able to benefit from special types of economies of scale that occur because of the concentration of individuals and industries within a city or region. These economies of scale, called agglomeration economies, come in one of two forms: localization and urbanization economies. Localization economies occur when there is a high concentration of one particular industry in a city or

region allowing firms to take advantage of knowledge spillovers within the industry and an industry specialized labor force that facilitates innovation at lower costs (O'Sullivan 2000). Urbanization economies come about because there is a large urban population with varying skills that complement and enhance one another. Knowledge spillovers occurring among industries and supporting industry establishment encourage innovation and reduce costs to firms, enabling areas with larger populations to enjoy higher income levels (Jacobs 1970). Much research has been done to determine which agglomeration economy has a greater impact on income and whether they actually occur. Some research has shown that the impact on income is much greater from localization than from urbanization, meaning that greater benefits are found in industry specialization rather than in increased population size (Henderson 1988, Vachal 2005). But Glaeser (1992) and Jacobs (1970) have argued that when many industries are so close together, each individual industry will have little incentive to innovate because they can freeride another company's ideas; thus any company that innovates will be unable to capture all the benefits, suggesting that industry diversification is better than specialization.

A final theoretical approach deals with the location of the region in relation to the market. Firms prefer to locate closer to their buyers in order to reduce transportation costs and maximize profits. As a result, regions located closer to a market will attract more income generating firms (O'Sullivan 2000). Vachal (2005) found evidence that income significantly decreases as city location increases in distance from a large population center.

#### **METHODS**

#### Model Specification

The theoretical relationships outlined between income and the factors explored above provide a foundation for developing a model that captures the per capita income differences in Utah counties. Using these theories, income can be expressed as the following function:

Income = f (Labor, Capital, Human Capital, Industry Concentration, Urban Population, Market Distance) (1) Using a Cobb-Douglas production function that models imperfect substitutability among capital and labor inputs, and adding other variables to account for regional income factors results in equation 2 where *y* is per capita income, *h* is per capita human capital, *k* is per capita capital stock, *l* is labor force participation rate, *U* is unemployment rate, *DM* is distance to market, *IC* is industry concentration and *UP* is urban population (for model justification see Glaeser, 1995; Huang, 2002; Moomaw, 2000; Vachel, 2005). This model will be used to determine contributing factor to county income in Utah.

$$\ln y = \beta_1 \ln h + \beta_2 \ln k + \beta_3 \ln l + \beta_4 U + \beta_5 DM + \beta_6 \ln IC + \beta_7 UP$$
(2)

#### Data

The data includes variables gathered for each of the 29 counties in Utah. Population data gathered from the U.S Census Bureau's 2000 Census (2009a) was used to calculate the per capita variables to control for factor variation caused by different population sizes.

Income is measured as the average annual per capita income of each county as reported by the 2000 Census (2009a). This reflects the income that resides in a county, not the income that is earned inside the county (i.e. income for where someone lives rather than works).

Human capital is split into two separate variables to account for differing impacts. The first variable is the percent of the population age 25 years and older whose highest education is at least a high school level or some college but not a bachelor's degree (%ED12-15). The second measure is the percent of the population age 25 years and older that have at least a bachelor's degree education level (%ED16). This data was gathered from the 2000 Census (2009a) and was divided by the population of the county.

Businesses that produce goods and services categorized by the Economic Report to the Governor for the State of Utah (2009) as Business Investment Purchases (BIP) tend to be more capital intensive, and as a result counties with high sales categorized as BIP are expected to have higher capital levels. Data on the tax collected from such purchases was gathered from the Utah State Tax Commission (2009) and was used to account for capital stock levels.

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Labor as defined in this study includes the unemployment rate (UMEMP) and the labor force participation rate (LPR). The Census (2009a) provided data on the number of people considered to be of working age, the number of people in the labor force, and the number of people that were unemployed. This data was used to compute the UNEMP and LPR.

Distance to market (DISTMKT) was measured by the highway distance from the county-seat to the nearest city the size of Salt Lake City or larger (e.g. Las Vegas, Denver, etc) using an internet mapping system. The shortest distance is recorded as the distance to market variable.

Industry concentration was measured by developing a Herfindahl-Hirschman Index (HHI). This index gives a measure of the degree of industry concentration within the county; the higher the HHI the higher the industry concentration. Data collected for income by industry was gathered from the Bureau of Economic Analysis (2009), and also reflects where income resides and not where it is earned. The HHI will result in an imperfect but usable estimate of industry concentration because people may work in a county other than where they live. The BEA did not provide figures for all industries in every county so the missing figures were estimated, where the average missing income accounted for about 6% of the total county income.

Finally, urban population (UBPOP) is given in the 2000 Census (2009a), and is defined as the number of people that live in areas with at least 500 people per square mile (U.S. Census Bureau 2009b). A description of the data is found in Table 1. Means, standard deviations, maximums and minimums are given for each variable.

Table 1. Explanation of Data

Variable	Mean	Std Dev	Max (County)	Min (County)		
Income	\$15,893	\$4,166	\$33,767 (Summit)	\$10,229 (San Juan)		
%ED12-15	38.7%	5.3%	51.1% (Daggett)	27.9% (Utah)		
%ED16	11.2%	4.4%	28.1% (Summit)	6.3% (Juab)		
BIP	\$2,598	\$1,442	\$8,099 (Carbon)	\$563 (Kane)		
LPR	64.1%	5.7%	77.7% (Summit)	53.4% (San Juan)		
UNEMP	5.9%	2.5%	15.1% (San Juan)	2.2% (Beaver)		
DISTMET	127 miles	78 miles	290 miles (San Juan)	0 miles (Salt Lake)		
HHI	0.205	0.053	0.375 (Daggett)	0.136 (Sevier)		
UBPOP	67,693	177,004	887,916 (Salt Lake)	0*		
* These counties h	* These counties had no urban population: Beaver, Daggett, Emery, Garfield, Morgan, Piute, Rich and Wayne					

#### Regression Model and Hypotheses

An ordinary least squares regression was used to explain variation in county-level income levels. The model was set up as follows:

$$\ln PCI = \beta_0 + \beta_1 \ln \%ED12 + \beta_2 \ln \%ED16 + \beta_3 \ln BIP + \beta_4 \ln LPR + \beta_5 UNEMP + \beta_6 DISTMET + \beta_7 \ln HHI + \beta_8 UBPOP$$

The theories outlined above provide expected relationships between income and each of the independent variables, and provide the basis for a set of hypotheses (Table 2). All variables will be tested using an alpha of 0.05 ( $\alpha$ =0.05).

Table 2. Expected Relations

Variable	Expected Relation	Null Hypothesis (Ho)
%ED12-15	Positive	$\beta_1 \leq 0$
%ED16	Positive	$\beta_2 \leq 0$
BIP	Positive	$\beta_3 \leq 0$
LPR	Positive	$\beta_4 \leq 0$
UNEMP	Negative	$\beta_5 \ge 0$
DISTMET	Negative	$\beta_6 \ge 0$
HHI	Uncertain	$\beta_7 = 0$
UBPOP	Positive	$\beta_8 \leq 0$

#### **RESULTS**

Results from the regression analysis and goodness-of-fit measures are outlined in Table 3. The adjusted R2 indicates that the model explains 80% of the variation in per capita income. The estimated coefficients indicate that both capital and education have a positive impact on income. A negative relationship is found between income and distance to nearest metropolis, indicating that the farther away a county is from a large city the lower its expected income. The remaining variables were found to be statistically insignificant at the 95% confidence level.

Because natural logs were used on many of the variables, including the dependent variable, the interpretation of the coefficients is different from a simple regression model. The coefficients of the explanatory variables that are logged indicate the percent change in income for a one percent change in the explanatory variable. Note that a one percent increase does not mean a one percentage point increase in the given percent measure (e.g. 20% to 21%), rather it means a one percent increase in the percent measure (e.g. 20% to 20.2%, where 0.2/20 = 0.01 or 1%). For example, the coefficient of at least 16 years of

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education is estimated to be 0.484, so a 1% increase in the percent of the population with at least 16 years of education will increase the expected per capita income by 0.484%.

Table 3. Regression Results (Dependent variable = natural log of annual per capita income, n=29)

Variable	Coefficient	t statistic	1 Std Dev Increase Impact	Ho			
Constant	10.5603	24.57					
%ED12-15†	0.4663	2.86***	\$ 1,011	Reject			
%ED16†	0.48408	5.65***	\$ 2,858	Reject			
BIP†	0.12069	2.47**	\$ 894	Reject			
LPR†	0.1407	0.33		Fail to Reject			
UNEMP	-0.448	0.41		Fail to Reject			
DISTMET‡	10032	-2.70***	- \$ 1, 222	Reject			
нні†	0.02002	-0.55		Fail to Reject			
UBPOP‡	-0.001758	-1.38*	- \$ 500	Fail to Reject			
$R^2 = 85.7\%$ Adjusted $R^2 = 80.0\%$ F-stat (p-value) = 14.99 (0.000)							
Mater CinniCarage I	$N_{eff}$ Circles and leads are indicated as $k = 10$ , $k \neq 10$ , $k \neq 10$						

Notes. Significance levels are indicated as \*p = .10; \*\* p = .05; \*
† Indicates the natural log of the variable was used

<sup>‡</sup> These variable were scaled to make coefficients easy to interpret; the coefficient are interpreted as the change is income as: DISTMET changes by 100 and UBPOP changes by 10,000

The meaning of the coefficients of the variables that were not logged is interpreted as the percent increase in income caused by an absolute increase in the explanatory variable. The estimated coefficient of the distance to metropolis is -0.10032, meaning when the distance to nearest metropolis increases by 100 miles the expected per capita income falls by 0.10032%. To investigate which variable had the greatest impact on income all variables are set equal to their mean values and an expected per capita income is obtained. Then one variable is increased by one standard deviation and the change in the expected per capita income is recorded. Table 3 shows the results from this method. This method found that of the significant variables, the percent of the population with at least 16 years of education had the greatest impact on per capita income and business investment purchases had the smallest impact. This result shows that higher education levels were the most important determinant of county income in this sample.

#### CONCLUSION

This paper has explored possible economic factors that explain the differing incomes in Utah counties. The data set and regression analysis found that of all of the variables included in this model higher education has the greatest impact on the income of the county. Also, the location of the county in relation to the nearest metropolis was found to be another important factor in income determination. Other

notable factors found were the positive impacts on income of general education level and capital stock level. Industry concentration as measured by the HHI was found to be insignificant, likely due to the measurement of income. Labor was not found to explain income variation at county levels, probably because of very high mobility at a county level (Glaeser, 1995).

Further research could investigate the relationship between changes in county income and changes in education over time in Utah. Future research should also take into consideration the income measurement being used. Similar studies could use income figures that represent where income is created or earned rather than where it resides (a county's GDP); doing this would capture industry concentration better than this study has.

Due to the cross sectional data set used in this study the findings cannot say much with regard to how a county can increase its income; such conclusions would require different research. The study has found what determined the income of Utah counties at the time of the 2000 Census and can be used to help future research indentify factors that should be included in modeling county level income.

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### JOHN B. GODDARD SCHOOL OF BUSINESS & ECONOMICS ECONOMICS

## Knowledge is the Power to Go Green in Ogden

Kimberly Robinson, Jeremy Ward, Chance Kendall, David Olson & Clifford Nowell

#### **ABSTRACT**

Analyzing data from sixty local residents, we took a look at the influences behind the accuracy of recycling in Ogden, UT. We examined the influence of expectations from neighbors, family, and friends, as well as personal attitudes toward recycling and their level of knowledge regarding recycling. We also looked at whether or not homeownership impacts recycling behavior, and questioned whether or not residents who have invested in Ogden through homeownership recycle more accurately than other residents. Through statistical analysis, we discovered the only significant factor in determining the percent to which residents recycled accurately is the level of knowledge they have regarding proper recycling. We discovered that on average 83% of a resident's recycling bin contains recyclable materials. We conclude that Ogden residents lack the necessary information to recycle accurately and could improve their accuracy through an educational program. Furthermore, because accuracy rates did not appear to differ among income, homeowners, renters, or location, remedial measures should not be aimed at any particular area or any particular demographic group.

#### INTRODUCTION

The Utah Department of Environmental Quality (DEQ) estimates over half of all solid waste in Utah is generated in just Salt Lake, Utah, and Weber counties alone. However, between 2006 and 2007 the Utah DEQ observed a 2.5 million ton decrease in non-hazardous solid waste, which DEQ attributes to recycling (Utah Report on The Environment, 2008). Ogden City is combating the rising tide of refuse with a free curbside-recycling program initiated in May 2002. In just the last ten

months, Weber County Transfer Station has collected approximately 210,160 tons of garbage, extracting and recycling approximately 70 tons per week (K. Cragun, personal communication, November 18, 2009).

While other studies focus on the behavioral determinants of recycling participation, this study focuses on the accuracy with which participants recycle; using determinants observed to be significant in previous participation research to model the accuracy of recycling in Ogden city. To do this, we randomly selected sixty Ogden residents from geographically distinct locations across Ogden city, photographed the contents of the residents' recycling canisters and surveyed the residents.

Heckler (1994) described the behavioral processes which motivated people to recycle. She found that people who live in neighborhoods plagued by high crime rates and lower levels of income tended to recycle less. Heckler noted that this could be due to the fact that these people are more concerned with survival and safety than their impact on the environment.

A key aspect of this study explains that people are best reminded about recycling by direct information that can be found in conspicuous locations. The simpler the message is, the more likely people are to understand and remember the information, and the better they will recycle. For example, a picture which identifies things that can be recycled on a refrigerator magnet or simple flyer is more effective than a lengthy message that explains why recycling is important for the environment. Also, messages that explain the direct benefits of recycling for the individual have a greater impact than messages concerned with the overall importance of recycling. Derksen, et al. (1993) similarly found that the level of concern for the environment had little to no impact on recycling behavior. The study found that the majority of people claimed to care about the state of the environment, which led researchers to believe that the level of concern for the environment was independent of personal characteristics such as race, age, income, etc. Further, they found that although the level of concern for the environment was higher in rural populations than urban populations, this did not positively impact their recycling behavior. The findings prove that the level of concern for the environment was not the determining factor in an individual's recycling behavior; instead, the availability of recycling facilities and curbside pickup were the strongest determining factors.

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Valle, et al. (2009) also found an inconclusive relationship between an individual's attitude toward the environment and their recycling attitude. Socio-demographic attributes like gender, age, or health also had no relationship with recycling participation. Valle's findings coincided with Derksen's results in that the significant influence was found between recycling behavior and the perceived convenience and performance of the recycling program as a whole.

Recycling programs that optimize the physical proximity of containers and are less demanding have been shown to have a positive impact on participation levels. Educating consumers on the benefits of recycling and the recycling procedure have also been found to positively impact participation levels.

Finally, Hornick, et al. (1995) studied the effects of two different categorical aspects of recycling: incentives and facilitators. Incentives were broken down into internal and external incentives. Internal incentives included personal attitudes towards recycling, while external incentives included laws and rewards. The study found that external incentives, such as monetary rewards for recycling, did impact recycling behavior, but were not the only option for the sustainability of a recycling program. Promoting internal incentives could also sustain the program. If individuals felt satisfied or committed to the program, then the programs would survive. This provides an optimistic outlook for governments, because it implies that recycling programs can be successful without offering a monetary incentive.

In addition, Hornick, et al. (1995) found that facilitators also impacted recycling behaviors. This category was broken down into internal and external aspects as well. Internal facilitators included knowledge of how to recycle, while external facilitators included aspects like the frequency of pickups. Of internal facilitators, knowledge and peer pressure led the way as the most influential factors on behavior. On the external facilitator side, the availability of recycling services proved to be the key determinant.

#### **METHODS**

Drawing on previous research, we focused our research on a few significant determinants: (1) neighborhood expectations or social norms; (2) personal attitude towards recycling; (3) invested interest

in the community; (4) knowledge; (5) income; and (6) home location within the city. We also looked at the reasons why a person might choose to not recycle. These results were used to view the correlation between the dependant variable, recycling accuracy, and each possible determinant.

Accuracy was calculated by photographing the contents of each resident's 90-gallon blue recycling bin and analyzing it for the percentage of correct items in the bin. Each observation was given a score between zero and one. A zero would be given for a can which contained no recyclable items and a one was given to those cans which contained only recyclable items. We determined whether or not items could be recycled by referring to the list of acceptable recyclable items on the Ogden City Public Services website (see appendix A). In order to obtain a measure of income levels, we used the assessed market value of each home sampled as a proxy. The market value was obtained from Weber County Assessors website. Location was obtained by segregating the sample evenly across the four municipal voting wards. Fifteen residences were randomly selected from each ward. This gave us a total sample of sixty homes.

Observations on the other determinants were obtained through a survey. It was administered in person between mid-October and early November. The survey contained five questions. The first concerned social expectations; each individual surveyed was asked to indicate the number of instances in which expectations from neighbors, friends, or family was a motivator in their recycling behavior, using a Likert-item scale (frequently, often, sometimes, seldom, never). The second question assessed whether or not the resident felt a strong personal obligation to recycle, using a Likert-item scale (strongly agree, agree, neutral, disagree, strongly disagree). The third question gathered information on reasons why people chose not to recycle. The participant was asked to rank the five most common reasons previous research cited as explanations for non participation. The participant ranked these five items from one to five, where one was the most relevant reason they choose not to recycle and five was the least relevant reason they did not recycle. However, a very large percentage of participants did not respond to this portion of the survey properly, and we decided not to include the data from this particular part. The fourth question was directed at the participant's living situation; residents were asked to indicate whether they rented, owned, or were in the process of buying their home. We were interested in investigating whether or

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not having a vested interest in Ogden City through home ownership would correlate with a greater level of concern about the future of Ogden City and better recycling accuracy. We later aggregated the categories of home ownership and in the process of buying into a single category. This eliminated any misunderstanding about the meaning of owning a home. The final question was designed to ascertain how knowledgeable the participant felt about the proper use of the recycling bin. Participants were asked to indicate the extent to which the statement, "I do not know how to use my recycling canister or where to find information about Ogden recycling," reflected their feelings, using a Likert-item scale (strongly agree, agree, neutral, disagree, strongly disagree). Strongly agree meant that the participant felt they did not know how to use the bin and strongly disagree meant the participant felt they had a perfect knowledge of how to use the recycling bin. Based on previous studies we summarized the expectations for each variable on accuracy (Table 1).

Table 1.

Independent Variable	Expectation Correlation
Income	Positive
Location	Unknown
Personal Obligation	Positive
Neighborhood Expectations (Peer Pressure)	Positive
Knowledge	Positive
Owning	Positive
Renting	Negative

After compiling the data, we decided to use dummy variables for each response to survey questions about personal obligation, neighborhood expectations, living situation and knowledge. We also used dummy variables for each category (i.e. municipal ward 1, 2, 3, 4) of location. We established benchmarks for each response or category as a means of comparing how each response differed from the average value of the benchmark response (Table 2). Without establishing benchmarks, we would have had a case of perfect collinearity. Then, we ran a multiple linear regression with recycling accuracy as the dependant variable specified as a function of income, location, personal obligation, peer pressure, knowledge, and living situation.

Table 2. Benchmark category or response

Qualitative Variable	Benchmark
Location	Ward 4
Personal Obligation	Frequently
Neighborhood Expectations (Peer Pressure)	Strongly Agree
Knowledge	Strongly Disagree
Living Situation	Own

#### **RESULTS**

From our survey we found on average 82.79% of the items in a recycling container could actually be recycled, while 17.21% could not be recycled. We also found 33% of the residents recycled 100% accurately.

The results from performing the initial regression on all of the variables included in the survey are reported in Table 3. Most of the factors were not statistically significant. The only two that may have some ability to predict recycling accuracy are the ownership status of the individual (whether they rent or own their residence) and how well an individual understood how to properly use the recycling container.

Table 3. Results from original regression (dependent variable: percent recycled accurately)

Variable	Coefficient	SE	t statistic	p-value
Constant	0.8354	0.1900	4.40	0.000
Assessed MKT Value	0.00000051	0.00000050	1.03	0.311
Rent	-0.14051	0.08430	-1.67	0.105
Location				
Ward 1	0.0611	0.1403	0.44	0.666
Ward 2	0.0533	0.1302	0.41	0.685
Ward 3	0.1501	0.1285	1.17	0.251
Social Pressure				
Often	0.0657	0.1003	0.65	0.517
Sometimes	0.0268	0.1246	0.22	0.831
Seldom	-0.1696	0.1056	-1.61	0.118
Never	-0.06328	0.07633	-0.83	0.413
Personal Obligation				
Agree	-0.07802	0.06382	-1.22	0.230
Neutral	0.1014	0.1048	0.97	0.340
Disagree	0.0644	0.1288	0.50	0.620
Strongly Disagree	-0.1546	0.1622	-0.95	0.347
Lack of Understanding				
Agree	-0.2283	0.1002	-2.28	0.029
Neutral	-0.2549	0.1464	-1.74	0.091
Disagree	-0.04966	0.06506	-0.76	0.451
$R^2 = 44$ .	6%	Adj. $R^2 = 18.5\%$	F-statistic (p-	value)= 1.71 (0.093)

To test for the robustness of some variables, we also ran a trimmed model to see if the significance on understanding remained the same. In particular, we ran several models with understanding and rent, including a model with understanding, rent, and social pressure. We omitted all the variables except understanding because after various t-tests and F-tests of incremental contribution the other variables were found to be statistically insignificant. This does not mean understanding is the only possible explanation for recycling accuracy, but that of the factors we investigated understanding is the only significant one. The results from our final model are summarized in Table 4.

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Table 4. Final Regression model results	(dependent variable:	percent recycled accurately)

Variable	Coefficient	SE	t statistic	p-value			
Constant	0.92571	0.03825	24.20	0.000			
Lack of Understan	Lack of Understanding						
Agree	-0.29571	0.08722	-3.39	0.001			
Neutral	-0.3257	0.1082	-3.01	0.004			
Disagree	-0.11441	0.05290	-2.16	0.036			
$\mathbb{R}^2$	= 27.5%	Adj. $R^2 = 22.9\%$	F-statistic (p-v	value) = 6.06 (0.001)			

The final model explains more of the variation in recycling accuracy than the original model. The adjusted R2 increased by 4.4% and the F-statistic is significant at the .01 level. The improvement in the adjusted R2 and F-statistic indicates this model is more robust than the original model. The estimated coefficients are all found to be significant at the .05 level; thus all the variables included in this final model have a significant impact on recycling accuracy.

The results indicate that the more a person understands how to use their recycling container the more accurately they will recycle. This finding is not surprising, and along with the fact that all of the other variables were insignificant, it shows that improving recycling accuracy is merely a matter of educating participants about the recycling program.

Table 5 shows the expected percent of correct items in an individual's container that would be recyclable for each level of understanding. The decrease in the percent correct that occurs as understanding increases from poor to fair is likely the result of a small sample size. We would expect this discrepancy to be resolved with an increased sample size. Table 5 also shows the expected amount of garbage in tons that would be inaccurately recycled per week (based on a 44 week average of 70 tons a week) for each level of understanding.

Table 5. Expected accuracy of recycling for each understanding level.

	Understanding Level			
	Poor Fair Good Great			
Expected Correct Percentage	63%	60%	81%	93%
Incorrect tons per week	25.9	28	13.3	4.9

These results clearly show that as a participant's level of understanding increases, the amount of refuse incorrectly placed in recycling bins will decrease significantly. This is good news, because if more is done to educate people about what can and cannot be recycled the amount of waste that is incorrectly recycled can decrease by as much as 21 tons, or the weight of three male African elephants.

#### DISCUSSION

Interest in recycling as a management strategy for solid waste is increasing globally. Ogden City's recycling program incorporates many of the significant factors other researchers have found to positively impact recycling behavior. This is reflected in the 83% average accuracy recycling rate in Ogden. However, with only 33% of residents recycling 100% accurately, we definitively show that there is room for improvement.

Improper recycling can result in negative consequences for the resident (i.e., removal of the individual's recycling canister) and for the entire city as well. At the lowest level of understanding, the cumulative effect of hundreds of residents inaccurately recycling 37% of their canister could be very costly for the city. The results clearly show that even after six years of recycling in Ogden, Weber County Waste Management cannot assume residents have a basic understanding about recycling procedures such as rinsing, lids, or when to put the canister out for pick up. These issues should be clearly spelled out for residents.

A major conclusion derived from the regression analysis is that Ogden residents lack an understanding of proper recycling. The study indicates that if more is done to help Ogden residents understand how to use their recycling containers the percent of items correctly recycled can increase up to as much as 93%, a 10% percentage point increase from the current average. This increase in proper recycling would decrease the amount of non-recyclables waste Ogden residents place in containers from 12 tons to nearly 5 tons per week—a significant cost savings for the Ogden City recycling program. This is very promising for Ogden; educating residents is a factor easier to control than other factors like personal attitudes.

Another major conclusion is that Weber County Waste Management will have to look at a broad based educational program. Our inability to discover a significant relationship between accurate recycling and homeownership, income, or location indicates the program should not target renters, low income neighborhoods or any other specific region of Ogden City. We also show that there is no need to try to influence residents' personal attitudes toward recycling. Positive attitudes toward recycling did not translate into accurate recycling or vice versa. The program would benefit most from providing simple and direct information placed in conspicuous locations.

#### CONCLUSION

Additional research should include other variables not specified in our research. Future research might find variables in our research significant if the response bias from using a personal interviewer survey approach is controlled. We feel perhaps the participant's responses were influenced by a psychological need

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to respond positively toward recycling in the surveyor's presence. Additional research into the differences between the recycling accuracy between Salt Lake, Utah, and Weber Counties would provide insight into whether or not some variables are specific to just the Ogden area. This could enable public officials to respond efficiently to improve the recycling program.

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### JOHN B. GODDARD SCHOOL OF BUSINESS & ECONOMICS ECONOMICS

## Maximizing the Value of Thoroughbred Mares: *A Hedonic Approach to Foal Value*

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Ralph Nye Undergraduate Research Scholarship

#### **ABSTRACT**

The purpose of this research is to identify whether or not a link exists between the racing career and value of the foals of a female thoroughbred racehorse. Such a link clearly exists for male thoroughbred racehorses. This paper utilizes the hedonic pricing model in order to determine if the dam's performance as a racehorse impacts the selling price of her foals. It is discovered that there is no statistically significant relationship between said performance and the selling price of the foal.

#### INTRODUCTION

Thoroughbred horseracing has been recorded in America as far back as 1665, although the traditional beginning of the sport is usually attributed to Governor Samuel Ogle of Maryland, who introduced the first organized races in 1745. The sport has continued to grow and now encompasses thousands of horses and a racing industry valued at over \$1 billion annually (The Jockey Club, 2009). With such a lucrative offering, thoroughbred racing has caused many auxiliaries, such as thoroughbred breeding, to require closer examination.

Within the thoroughbred industry there are two key sources of revenues, the racetrack and the breeding program. For owners, especially of female horses, it is imperative to identify the time when a horse should be taken off the track and placed in the breeding program. In order to maximize revenues, the owner must pinpoint the optimal time for the switch. This timing is very difficult, and requires a vast amount of information in order to make the best decision.

In order to make this decision more accurate, we must question whether or not a horse's performance on the track improves the selling price of future foals. For sires, the answer is undoubtedly yes; but that certainty is lacking for dams. This paper will attempt to answer that question indirectly for dams. In order to do so, a hedonic pricing function will be developed to examine the influence of a dam's racing performance on the selling price of her foals.

#### Literature Review

Demand theory is the theoretical basis underlying the model to be used. Frank (2008) defines the determinants of demand to include income, tastes, expectations, population, and the prices of substitutes and complements. These determinants are best used to describe the demand for homogenous goods. Racehorses are not homogeneous goods, but rather heterogeneous goods. All racehorses have certain base characteristics that help them to accomplish their purpose, but every horse is also unique. This is what marks them as a heterogeneous good. With a heterogeneous good, such as racehorses, the individual characteristic of each foal has an impact on selling prices. Therefore, the determinants of demand for a heterogeneous good include all of those listed for the homogeneous good as well as the characteristics of the foal.

In the equine industry, there are many key factors that play a role in the demand for a given animal. Ray (1991) attempts to identify these factors by analyzing the stud fees associated with the top 20 performance quarter horses for the year 1987. She explains that the value of these fees is very dynamic and depend on the performance of the offspring of the sire, which means that the owner of the sire wants the sire's offspring to perform extremely well, thereby increasing the demand and price of the sire's stud fee. Ironically, by producing the top performing offspring, the sire is essentially creating substitutes for his genes, which then lowers the demand, and hence price, of the stud fee.

Ray goes on to estimate the effect of this relationship, as well as many others, in determining the value of the stud fee for racing quarter horses. In her model she used the stallion's lifetime wins/starts ratio, previous year's earnings by the stallion's offspring, number of the stallion's offspring who won a race the previous year, and the number of wins for said offspring. The most significant variable found was the earnings of the offspring from the previous year. Interestingly, the stallion's

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personal wins/starts ratio turned out to be statistically insignificant. This suggests that the value of the stud fee was more strongly influenced by the stallion's other offspring's performances, rather than his own. From this study, it appears that data collected for horses on the same generational level are more accurate predictors than data for horses on different generational levels.

Following Chezum et al. (2000), another important factor that influences the selling price of thoroughbred racehorses is where the horse came from. Chezum et al. (2000) finds that the market exhibits adverse selection depending on who bred the horse. Chezum et al (2000) classified breeders into one of three categories: (1.) breeders who sell all of their foals; (2.) breeders who race all of their foals; and (3.) breeders who both race and sell their foals. Those breeders that fall under the third category have foals that sell at lower values because buyers are assuming the owners are retaining their best stock for private racing and selling the lower quality stock. It is assumed that breeders have asymmetric information and are not revealing the true reasons for selling the foal.

#### **METHODS**

#### Model

The hedonic pricing model is an analytical tool commonly used to identify the significance and impact of characteristics of a good or service on price. Essentially, this pricing model is used to place a value on the heterogeneous aspects of a good. The hedonic model attempts to show the existence and the impact of a relationship between the variable aspects of the good and its selling price.

Rosen (1974) provided the basic theoretical framework for the hedonic pricing model. In his research he defines hedonic prices as the "implicit prices of attributes and are revealed to economic agents from observed prices of differentiated products and the specific amounts of characteristics associated with them" (Page 34). More simply, the price of a good is influenced by the characteristics of the good and the hedonic pricing model attempts to capture that relationship. This is accomplished through regression analysis where we attempt to identify the impact of an independent variable (one of the product's characteristics) on a dependent variable (price). The most common functional form of this analysis is a linear regression.

#### Data

All the data from this experiment was taken from the Keeneland Auction website for their November Breeding Stock Sale that took place from November 10 through November 16 of 2009. The sample took 110 weanlings from the first and second sessions that had a final agreed price. Observations were omitted when the foal was part of a total dispersion, meaning the owners were forced to sell all foals, regardless of how unhappy they were with the final price.

The main reason for limiting the sample to weanlings at the Keeneland Auction from the November sale was to control for exogenous variables such as economic conditions, implicit costs for care, racing viability, and data presentation. As such, the model can be used explain the impact of characteristics endogenous to the breeding, instead of allowing exogenous variables to control the selling price of the foal. An explanation of each variable gathered and its expected impact on the dependent variable, selling price, follows in Table 1. *See next page*.

There are a few variables that would be an improvement to the model; however, due to time, cost, and unavailability constraints they were unobtainable. These variables include Dam Wins/Starts, Dam Other Foal Wins/Starts, Confirmation, and Breeder Type (as a measure of adverse selection as explained in the *Literature* section). The main reason the data cannot be collected regarding the dam is due to the fact that it has not been compiled. There currently is no one in the industry that finds the information pertinent enough to track it. This means that in order to obtain the data an individual would have to find the results for all races the dam and/or her offspring competed in and compile it. This is simply not feasible for this project.

While having this data would likely provide information, these variables are not presented to the buyer at the time of the sale. The only way for the buyer to obtain this information is by personal collection PRIOR to the sale.

#### Method

Several model specifications were explored to ensure robustness, including a full model of all available explanatory variables. After analyzing these models, a restricted model containing fewer variables was selected.

Table 1. Variable Descriptions and A Priori Expectations on price

Variable	Description	Expected Relationship	Theory
Selling Price	Dependant Variable	N/A	N/A
Foal Gender	Dummy Variable 1=Male and 0=Female	Positive	Male horses tend to produce more offspring and race more competitively
Sire Career Earnings	Measure of sire's performance	Positive	Athletic horses should produce other athletes
Sire Other Foal Wins/Starts Ratio	Measure of sire's ability to produce athletic offspring	Positive	Generational Data
Sire Other Foal Career Earnings	Measure of sire's ability to produce athletic offspring	Positive	Generational Data
Dam Career Earnings	Measure of the dam's performance	Positive	Athletic horses should produce other athletes
Dam Other Foal Career Earnings	Measure of dam's ability to produce athletic offspring	Positive	Generational Data
Sire First Foal Unraced	Dummy Variable 1=Sire has no foals old enough to race	Positive	Compensate for unproven sires
Sire First Foal Raced	Dummy Variable 1=Sire with few foals that have raced, but still noted as new	Unknown	Has potential to be either positive or negative
Dam First Foal	Dummy Variable 1=Dam has no foals old enough to race	Unknown	Captures impact of unproven dam, could be either ±.
Dam Unraced	Dummy Variable 1= Dam did not race	Negative	Dam has not proven her ability as a competitor
Sire Total Get of Racing Age	Count of total offspring of racing age a sire has	Positive	Measure of how proven the sire is as a father
Site Career Earnings <sup>2</sup>	Previously described variable squared	Negative	Earnings exhibit diminishing returns
Sire Other Foal Career Earnings <sup>2</sup>	Previously described variable squared	Negative	Earnings exhibit diminishing returns
Dam Career Earnings <sup>2</sup>	Previously described variable squared	Negative	Earnings exhibit diminishing returns
Dam Other Foal Career Earnings <sup>2</sup>	Previously described variable squared	Negative	Earnings exhibit diminishing returns
Sire Total Get of Racing Age <sup>2</sup>	Previously described variable squared	Negative	Possible substitution of sire by his other offspring

(T. 1)

Within this restricted model, all seven Sire and Sire Other variables were included. The correlation among these variables was very high. As such, a dummy variable was created to limit multicollinearity issues. The proposed variable is a ranking for the 2009 Top Weanling Producing Sires. This variable is broken down into two categories: Top 10 Sires and Top 11-20 Sires. With the inclusion of this variable, we can exclude the seven Sire and Sire Other Foal variables, which improves the model's degrees of freedom and avoids multicollinearity issues. Equation 1 represents the model used to obtain addressable results.

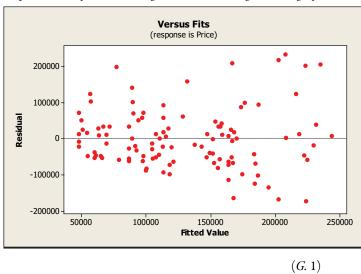
Equation 1. Regression Equation

 $SellingPrice = \beta_0 + \beta_1 * FoalGender + \beta_2 * DamCareerEarnings + \\ \beta_3 * DamOtherFoalCareerEarnings + \beta_4 * DamCareerEarnings^2 + \beta_5 * \\ 2009TopTenWeanlingProducingSire + \beta_6 * 2009Top11-20WeanlingProducingSire \\ (E. 1)$ 

#### **RESULTS**

When Equation 1 is regressed, scatterplot G.1 is produced. Although it is not obvious, this graph reveals the potential for heteroscedasticity.

Graph G.1. Scatterplot of residuals against fitted values of regression using Equation 1.



Heteroscedasticity basically means that the variances throughout the regression are not constant, but rather dependent upon the magnitude of a variable in the model. The graph reveals this by plotting the points in a funnel fashion, rather than a randomized plot. In this case, the variances are fairly close together for the lower levels of the Fitted Value and then tend to become more spread out as the Fitted Value increases. Using Park/Glesjer tests, heteroscedasticity is proven to exist, and therefore must be corrected.

In order to correct for heteroscedasticity, each explanatory variable must be divided by the square root of the fitted value. Also, the regression must now include a variable of the reciprocal of the square root of the fitted value, and the intercept removed. The final regression model, which has been corrected for heteroscedasticity, yields the results found in R.1.

$$\frac{Selling \Pr ice}{\sqrt{Fits}} = 59,267* \frac{1}{\sqrt{Fits}} + 30,311* \frac{FoalGender}{\sqrt{Fits}} + 30,829* \frac{DamCareerEarnings}{\sqrt{Fits}} \\ +18,412* \frac{DamOtherFoalCareerEarnings}{\sqrt{Fits}} - 0.0249* \frac{DamCareerEarnings^2}{\sqrt{Fits}} \\ +108,796* \frac{2009Top10WeanlingProducingSire}{\sqrt{Fits}} \\ +57,746* \frac{2009Top11-20WeanlingProducingSire}{\sqrt{Fits}}$$

$$(R.~1)$$

Table T.R.1. Regression R.1 analysis

Variable	T Statistic	P Value	Significant (Given 95% Confidence)
1/RT Fits	4.73	0.000	Yes
Foal Gender	2.06	0.042	Yes
Dam Career Earnings	0.44	0.662	No
Dam Career Earnings^2	-0.54	0.593	No
Dam Other Foal Career Earnings	1.38	0.170	No, but significant at the 90% confidence.
2009 Top 10 Weanling Sire	5.18	0.000	Yes
2009 Top 11-20 Weanling Sire	3.27	0.001	Yes

(T.R. 1)

The T-Tests for each individual variable conclude that Foal Gender, 2009 Top 10 Weanling Sire, and 2009 Top 11-20 Weanling Sire are all statistically significant at the 95% confidence level. This is due to the fact that the null hypotheses of

$$H_0: \beta 1 \le 0$$
  
 $H_0: \beta 5 \le 0$   
 $H_0: \beta 6 \le 0$ 

are all rejected because the P-Values for each variable in question are less than 0.05.

The calculated F-statistic that tests for the significance of the model as a whole was 39.09 with a corresponding P-Value of zero. From these results we can conclude that the model is statistically significant at the

95% confidence level. The results of the F-Test lead me to reject the null hypothesis of:

$$H_0: \beta_0 = \beta_1 = \beta_2 = \beta_3 = \beta_4 = \beta_5 = \beta_6 = 0$$

With the viability of the method described, it is important to interpret the meaning behind the values within the model. The coefficients for both Dam Career Earnings and for Dam Career Earnings2 were determined to be statistically insignificant, and are therefore not interpretable.

The coefficient for Dam Other Foal Career Earnings was proven to be statistically significant at the 90% confidence level. The value of this coefficient, 18,412, implies that the value of the foal increases by almost \$20,000 for every million dollars earned by the dam's other foals. The coefficient of foal gender means that male horses are worth approximately \$30,000 more than their female counterparts. The impact of a sire being ranked among the 2009 Top 20 Weanlings Sires creates a large impact. Horses ranked in the Top 11-20 have foals that sell for almost \$58,000 more than their unranked peers. This value is almost doubled for horses within the 2009 Top 10 Weanling Sires, whose foals sell for about \$109,000 more than an unranked sire's offspring and \$51,000 more than sires that fall in the Top 11-20 range.

#### CONCLUSION

The hedonic model results lead to the conclusion that the dam's racing performance does not statistically influence the selling price of her future foals. Although this is not what was expected, it is the result of the model. These results do not answer the question of how to maximize the profits for a given female racehorse, but they do lead to a more informed insight. Essentially, when dealing with the quality of animals represented by the sample, the potential impact of the female horse's racing performance will not improve her standing as a broodmare.

Although the dam's racing performance may not impact the selling price of future foals directly, there is the possibility that the racing career may impact future breeding by earning the dam the right to be bred to top

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stallions. It is very common for stallion owners to provide incentives for breedings with "approved mares," meaning that mares which have certain characteristics are likely to receive discounts and potentially free breedings. This is in the best interest of both parties because a proven mare is more likely to produce a winning foal than an unproven mare. The foal can then go on to improve the popularity of the sire and to prove the stallion as a superior sire.

#### **ACKNOWLEDGEMENTS**

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#### APPENDIX A: THOROUGHBRED TERMINOLOGY

The following glossary of Thoroughbred Terminology was taken from The Jockey Club website:

#### Age of a Horse

Age of the Thoroughbred: For the purpose of determining age, the date of birth for all Thoroughbreds is deemed to be January 1 of the year of foaling.

Foal: A young horse of either sex in its first year of life.

Weanling: A foal of any sex in its first year of life after being separated from its dam.

*Yearling*: A colt, filly or gelding in its second calendar year of life (beginning January 1 of the year following its birth).

#### Breeding Terminology

Breeder: The breeder of a foal is the owner of the dam at the time of foaling, unless the dam was under a lease or foal-sharing agreement at the time of foaling. In that case, the person(s) specified by the terms of the agreement is (are) the breeder(s) of the foal.

Stallion: A male horse that is used to produce foals.

Sire: A male horse that has produced, or is producing, foals.

*Broodmare*: A filly or mare that has been bred (mated) and is used to produce foals.

Dam: A female horse that has produced, or is producing, foals.

#### Gender of a Horse

Colt: An entire male horse four years old or younger.

*Horse*: When reference is made to gender, a "horse" is an entire male five years old or older.

*Gelding*: A male horse of any age that is unsexed (had both testicles removed).

Filly: A female horse four years old or younger.

Mare: A female horse five years old or older.

#### Kendall & Geide-Stevenson · 33

#### In addition to The Jockey Club's Glossary it is necessary to define the following words:

*Get*: The offspring of a stallion.

*Produce*: The offspring of a dam.

Confirmation: The physical structure of a horse's body. Good conformation means that the horse has a desirable physical build. Bad conformation means a horse has a physical structure that may predispose him to injury, or be uncomfortable to ride.

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### DR. EZEKIEL R. DUMKE COLLEGE OF HEALTH PROFESSIONS CLINICAL LABORATORY SCIENCES

## Blood Culture Processing and the Effects on Time to Positivity

Minh C.Vu, Andrew Baggs, Joshua Hastings & Scott Wright

#### **ABSTRACT**

This research will determine if there is a significant difference between the current processes of "collection then incubation" vs. "collection, transportation, then incubation" of patient blood cultures collected from three IHC clinical laboratories. Utah Valley Regional Medical Center in Orem, Utah, collects their blood cultures and immediately incubates them on site. They transport any positive cultures to the Intermountain Medical Center's central laboratory in Murray, Utah for further workup. McKay Dee Hospital in Ogden, Utah, collects the blood culture bottles and transports them to the Intermountain Medical Center's central laboratory for incubation and further work-up if needed. The average distance between these two locations to the central laboratory is thirty-seven miles with a travel time of roughly forty-five minutes. Six months of positive blood culture data from IHC was collected and analyzed using chi-squared method with an alpha level of 0.05 to determine if there was a statistically significant difference between the two facilities. The chi-squared calculation gave a critical value of 3.841 with p = 0.05. This indicates a significant difference between the methods of collection to setup from the two facilities. There was not a significant difference in the overall collection process to final result.

#### **ABBREVIATIONS**

IHC = Intermountain Healthcare
MT = Medical Technologist
CNS = coagulase negative *Staphylococcus*STSP = *Streptococcus* species
CV = coefficient of variation.

#### INTRODUCTION

The whole blood culture process begins the moment the patient's blood has been drawn directly into a blood culture bottle. Time stamps are a way of keeping track of the time and location for a specimen's progress in the laboratory. The phlebotomist that collects the blood places their initials, date, and time on the bottles; thus creating the first time stamp in the blood culture process. These collected specimens are then brought to the on-site laboratory to be received by specimen processing. Specimen processing accesses the patient's name and the ordered test on computer; this creates another time stamp. Depending on the laboratory's location the process changes at this point.

McKay Dee Hospital located in Ogden, Utah, collects a batch of patient blood culture bottles and sends them by courier to the Intermountain Medical Center's central laboratory in Murray, Utah. When the blood culture specimens at this location are placed into a batch, another time stamp is created. As the courier arrives at the central laboratory, the specimen processors receiving the blood cultures create a new received time stamp. The batch is then taken to the microbiology department to be placed into the blood culture incubator known as the BACTEC 9240, creating another time stamp (Nolte et al., 1993). Any blood culture bottle with pathogens present causes the BACTEC to sound an alarm. An MT will take out that bottle and perform a Gram stain looking for any pathogen to verify a true positive. If no pathogens are seen, the bottle will be placed back into the BACTEC for further incubation. If a pathogen was seen, the MT will enter the result into the computer, which creates another time stamp. At this point the blood culture is identified as positive for bacteria.

The process of blood culture processing at Utah Valley Regional Medical Center located in Provo, Utah, differs from McKay Dee Hospital. At the Utah Valley Regional Medical Center, the collected blood culture bottles are immediately placed into the BACTEC on-site, creating a time stamp. This method of processing blood cultures omits the time delay on incubation of collected blood culture bottles. Any blood culture bottles with pathogens present will cause the BACTEC to sound an alarm. The MT will take these bottles out and perform a Gram stain looking for any pathogen to verify a true positive. If a

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pathogen is seen, the MT enters the result into the computer where a time stamp is created. The MT sends these positive blood culture bottles by courier to the central laboratory for identification of the pathogen growing in that blood culture bottle. A time stamp is recorded when these blood culture bottles are sent from Utah Valley Regional Medical Center as well as when they are received at the central laboratory.

The distance between McKay Dee Hospital and Utah Valley Regional Medical Center are roughly equal in distance from the Intermountain Medical Center's central laboratory. The average distance between these two locations to the central laboratory is thirty-seven miles with a travel time of approximately forty-five minutes. Once the blood cultures arrive at the central laboratory from either location the process of identification is identical. An MT places a small sample of the blood onto culture plates which is designed to grow out all pathogens. This media is allowed to incubate overnight, after which a preliminary identification of the pathogen is entered into the computer system which creates the final time stamp.

The purpose of this research project is to find the most time efficient process for the identification of positive blood cultures by comparing the various time stamps previously recorded from the two methods.

#### **METHODS**

#### Data Analysis

Six months (August 2008 to January 2009) of data including collection time, time to positivity, and preliminary identification time was provided from the central lab on previously collected patient blood cultures. Only positive blood cultures with the following organisms present were used in the statistical analysis: CNS, *Staphylococcus aureus*, *Escherichia coli* and *Streptococcus* species. Utah Valley Regional Medical Center and McKay Dee Hospital were compared using the central laboratory as standard. The chi-squared formula

$$(\chi 2 = \frac{(Observed - Expected)^2}{Expected})$$

and an alpha level of 0.05 were used to determine any level of statistical significance.

#### Simulated Blood Cultures

Blood culture samples were simulated according to the following method. Bacterial suspensions were diluted in physiological saline to attain the desired number of bacteria for blood culture bottle inoculation. The bacterial concentration was determined by a quantitative plate count method: 0.1 ml of six 10-fold serial dilutions using a 0.1 MacFarland standard was spread on blood agar (Haimi-Cohen et al., 2002; Schwetz, 2007). Following overnight incubation at 37°C the plates contained 10 to 25 colonies. The bacterial suspension with an approximate density of 1 to 100 CFU/ml was prepared from the serial dilution and served as the inoculum. Approximately 0.4 ml of the suspension was injected aseptically into thirty-four BACTEC aerobic bottles (Karen-Mae & Brecher, 1999). Two inoculated blood culture bottles were immediately placed in the BACTEC 9240 instrument. The remaining thirty-two inoculated bottles remained at room temperature (20 to 25°C) and were inserted in the BACTEC 9240 in pairs every thirty minutes for a period of eight hours. As the bottles turned positive, the time to positivity was recorded. The results were analyzed using the Gaussian distribution method.

#### **RESULTS**

A total of 2375 positive blood cultures were analyzed. The average time from collection to setup, collection to positivity, and collection to final results of positive blood cultures with the following organisms present were used in the statistical analysis: CNS, *S. aureus*, *E. coli* and *Streptococcus* species (Table 1). Using the values from Table 1 the chi-squared formula gave a critical value of 3.841 with p = 0.05 (Table 2). The simulated blood culture had an average incubation time of 16.7 hours with a CV of 6.3%. *See tables on next page*.

#### DISCUSSION

#### Simulated Blood Cultures

The CV is the absolute expression of precision. As the CV decreases, the precision increases. The CV obtained during the duration of this experiment was 6.3%. The blood cultures that remained at room temperature (20 to 25°C) before being incubated in the BACTEC were unaffected and still turned positive at an average time of 16.7 hours after the initial room temperature delay was subtracted out.

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**Table 1.** Average turnaround times, in hours, of blood culture processing for four most common organisms isolated from McKay Dee Hospital and Utah Valley Regional Medical Center and Intermountain Medical Center's Central Laboratory

	CNS	S.aureus	E.coli	STSP
Collection	0.871	0.838	0.550	0.789
Collection to Setup	1.999	2.134	1.980	1.960
Collection to Positivity	27.348	24.014	19.439	25.976
Collection to Final Result	77.368	63.575	71.219	51.701
Total Count	215	214	70	263
McKay Dee Hospital				
	CNS	S.aureus	E.coli	STSP
Collection	0.582	1.211	0.382	0.696
Collection to Setup	6.228	6.396	5.724	6.023
Collection to Positivity	37.133	30.297	22.868	35.511
Collection to Final Result	82.950	62.486	66.909	53.737
Total Count	147	132	68	175
Intermountain Medical Center's Co	entral Laboratory			
	CNS	S.aureus	E.coli	STSP
Collection	0.530	0.412	0.491	0.506
Collection to Setup	2.190	1.805	2.664	2.027
Collection to Positivity	31.490	25.357	22.478	19.423
Collection to Final Result	76.710	60.117	68.792	47.331
Total Count	250	383	102	356

Table 2. Comparison times between Utah Valley Regional Medical Center and McKay Dee Hospital to the Intermountain Health Center's Central Laboratory

$\chi 2$				
	CNS	S.aureus	E.coli	STSP
Collection	0.224	1.990	0.031	0.230
Collection to Setup	7.462	11.737	3.690	7.880
Collection to Positivity	1.556	1.034	0.418	0.418
Collection to Final Result	0.513	0.292	0.137	1.270

#### Data Analysis

The chi-squared formula gave the following Ho for collection times of the following organisms: CNS = 0.224, *S. aureus* = 1.990, *E. coli* = 0.031, and STSP = 0.230. The collection times for McKay Dee Hospital and Utah Valley Regional Medical Center fell below the chi-squared critical value of 3.841. There was not a significant difference between the collection methods of the two facilities.

The chi-squared formula gave the following Ho for the collect to setup times of the following organisms: CNS = 7.462, *S. aureus* = 11.737, *E. coli* = 3.690, and STSP = 7.880. The collect to setup times for

McKay Dee Hospital and Utah Valley Regional Medical Center were above the chi-squared critical value of 3.841. This indicates that there is a significant difference between the setup times of the two facilities. *E. coli* had a H<sup>O</sup> below the critical value that may be due to the low sample numbers obtained.

The chi-squared formula gave the following Ho for collection to positivity times of the following organisms: CNS = 1.556, *S. aureus* = 1.034, *E. coli* = 0.418, and STSP = 0.418. The collection to positivity times for McKay Dee Hospital and Utah Valley Regional Medical Center fell below the chi-squared critical value of 3.841. There was not a significant difference between the time it took the blood culture bottles to become positive.

The chi-squared formula gave the following  $H^{\circ}$  for collection to final result times of the following organisms: CNS = 0.513, S. aureus = 0.292, E. coli = 0.137, and STSP = 1.270. The collection to final result times for McKay Dee Hospital and Utah Valley Regional Medical Center fell below the chi-squared critical value of 3.841. There was not a significant difference between the current processes of "collection then incubation" vs. "collection, transportation, then incubation" of patient blood cultures collected from the two facilities.

McKay Dee Hospital's process of collect, transport and incubate is approximately four hours longer than Utah Valley Regional Medical Center's current method. Once the blood cultures arrive at the central laboratory from either location the process of identification is identical. This delay in McKay Dee Hospital's processing is subtracted out during the overnight identification process. This can explain why there is not a statistical difference between the two facilities.

#### CONCLUSION

There was not a significant difference between the current processes of "collection then incubation" vs. "collection, transportation, then incubation" of patient blood cultures collected from the two facilities.

Although processing should be done in a timely manner, any reasonable delays in processing should not interfere with the overall outcome of the identification of pathogens in blood cultures.

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#### **ACKNOWLEDGEMENTS**

Blood culture bottles, any needed materials, and use of the BACTEC 9240 were provided by IHC through George Hinde.

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### DR. EZEKIEL R. DUMKE COLLEGE OF HEALTH PROFESSIONS CLINICAL LABORATORY SCIENCES

## The Effects of High Ionic Strength Solution on Platelet Storage

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Eccles Undergraduate Research Scholarship

#### **ABSTRACT**

Platelet components are stored at room temperature on an agitating device to maintain a state of continuous motion which keeps the platelets from aggregating until ready for transfusion. For in-vitro testing of red blood cells, a 0.03 Molar low ionic strength solution is used to decrease the zeta potential (net negative charge) surrounding red blood cells. This decreases the distance between adjacent membranes and enhances testing sensitivity. In a like manner, we propose that by suspending donor platelets in a high ionic strength solution [0.9 - 1.0 M], the net zeta potential will increase or at least maintain the distance between adjacent platelet membranes. We postulate that saturation of the platelets with HISS will inhibit platelet aggregation for the duration of storage, and allow the storage of donor platelet components without agitation. Eliminating agitators will save hospitals money, space, and time. To quantify the findings, platelet recovery counts were run on the Coulter Max M. Stationary treated platelets yielded a 77.8% recovery, which conforms to the FDA's minimal requirements of 66%.

#### INTRODUCTION

Currently, therapeutic platelet components are stored at room temperature (20-24°C) on an agitator to maintain a state of continuous motion for a maximum of five days (Gregory, 2002). Transfused units must be utilized within these five days; this motion is intended to keep the platelets from aggregating. Platelet components left stationary for 24 hrs are considered expired and no longer suitable for transfusion.

In blood bank testing, a 0.03 Molar (M) Low Ionic Strength Solution (LISS) is used as an *in-vitro* testing reagent to decrease the zeta potential (net negative charge) surrounding red blood cells (RBCs) (Swarbrick, 2006). LISS allows the red blood cell membranes to come closer together, thus enhancing antibody mediated agglutination reactions of RBCs (Blodget, Hains, & Wren, 2008). Previous research has identified that zeta potentials are found on PLT membranes (Klinger, 1997). This current research project proposes that by suspending donor platelets in a High Ionic Strength Solution (HISS) [0.9 - 1.0 M], the net zeta potential will either maintain the typical separation of platelets within the unit or increase the distance between each platelet, reducing the potential for aggregation and thereby maintaining platelet viability (Figure 1).

The hypothesis of the study is that saturation of the platelets with HISS will inhibit platelet aggregation for the duration of storage time (five days), and allow for the storage of donor platelet components without agitation.

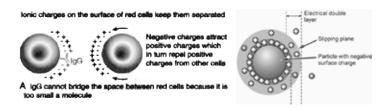


Figure 1.

#### **METHODS**

Test tubes containing 0.25 mL of platelets were inoculated with four different concentrations of HISS: 0.90, 0.94, 0.98, 1.0M. Each concentration contained three different HISS-to-platelet ratios: 1:1, 1:2, and 1:4. The samples were run in triplicate providing thirty-six preliminary test samples. Each tube was left stationary to simulate HISS inoculated units to identify the effective concentration levels. The reagents used to make the preliminary HISS are summarized in Table 1 on the next page.

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Table 1. Preliminary HISS test concentrations and rati
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Solvent	Solutes			
9 mL dH20	1.0 M NaCl	.98 M NaCl	.94 M NaCl	.90M NaCl
y 1112 GF 120	0.5259 g	0.5156 g	0.4944 g	0.4736 g
5 mL dH2O (Buffer Sol. 0.15 M)	.0888 g Monobasic Dihydrate Na Phosphate .0253 g Dibasic Anhydrous Na Phosphate			
200 mL dH2O	5.8815 g NaGlycine (0.3M)			
4*50 mL total HISS	41 mL of Buffer NaGlycine solution/9 mL NaCl			

Recovery of platelets was assessed upon gently reconstituting the platelets of the preliminary study after storage. Following standard operating procedure, each technician inspected trial tubes for visual agglutination. Each concentration gave similar results: no visible agglutination. Therefore, the middle grade concentration of 0.98 M at a 1:2 ratio was chosen for the primary testing to utilize a minimal amount of NaCl and small volume of HISS in order to obtain the desired effect.

For the primary testing, 275 mL 0.98 M HISS was made. To make the HISS, a 5.5 mL buffer solution, 49.5 mL NaCl solution, and a 220 mL 0.3M NaGlycine were mixed together. The buffer solution consisted of 0.097 g Monobasic Sodium Phosphate and 0.028 g Dibasic Sodium Phosphate. The NaCl solution was made by adding 5.670 g NaCl to 99 mL distilled water. The 0.3 M NaGlycine solution consisted of 7.3518 g NaGlycine dissolved in 250 mL distilled water. Finally, 2.5 N HCl was added to adjust the pH to 6.7 (Table 2).

Table 2. HISS Solution Recipe for primary studies 0.98 Molar NaCl

Solvent	Solutes
49.5 mL dH20	5.670 g NaCl
5.5 mL dH2O (Buffer Sol. 0.15 M)	0.0.097 g Monobasic Dihydrate Na Phosphate 0.0028 g Dibasic Anhydrous Na Phosphate
220 mL dH2O	7.3518 g NaGlycine (0.3M)
275 mL total HISS	41 mL of Buffer NaGlycine solution/9 mL NaCl
	2.5 N HCl was added to adjust the pH to 6.7

A platelet count of 1798 PLTS/mL, volume 1119 mL, with a PLT count of  $20.0 \times 103$  was measured to establish a baseline. Sixty 150 mL pediatric bags containing 16-18 mL of platelets were received. Three of the units' volumes were adjusted to minimize variation; bags were grouped into four groups: non-HISS agitated and non-HISS stationary to serve as a control group, and HISS agitated and HISS stationary to

act as a study group. Using aseptic technique each unit was inoculated with the appropriate volume of HISS to equal 1:2 ratio of 0.98 M. These units were maintained at room temperature (20-24°C) for the duration of the study, and agitated platelets were rotated at 120-135 cycles/min.

After the storage period, a visual inspection was done and platelets were counted on Weber State University's Coulter Max M to compare the recovery values against the baseline. Samples were plated on Sheep Blood Ager (SBA) to check for possible bacterial contamination.

#### **RESULTS**

After the preliminary test, no visible agglutination or degradation differences were found in any of the thirty-six HISS tubes. A combination of 0.98 M and 1:2 ratio of HISS solution were selected for the full study.

After the agitation/stationary phase of the primary study, macroscopic inspection of the pediatric units showed precipitants in 25 of the 30 agitated units. These agitated results contrasted sharply to the result from the stationary units where only 6 of the 30 units had visible particulates. A platelet count was measured for each sample on the WSU Coulter Max M instrument at a 1:10 dilution. The results showed that the stationary platelet aliquots treated with the HISS solution contained 77.8% of the original sample platelet count. The treated and agitated samples contained 60.5% of the original sample platelet count, untreated and agitated contained 67.9% of the sample, and those untreated and stationary contained 64.0% of the original sample. The results of each bag were compared to an average platelet count of 1798 x 103/ $\mu$ L taken at day one (Figure 2). See next page.

Five samples of the treated and agitated group and one sample of the untreated stationary group had bacterial contaminants.

#### DISCUSSION

Numerous chemicals are under study to enhance and prolong platelet storage (Kaufman, 2006). Since the average cost of platelets is approximately \$500 per unit, un-transfused units are a significant

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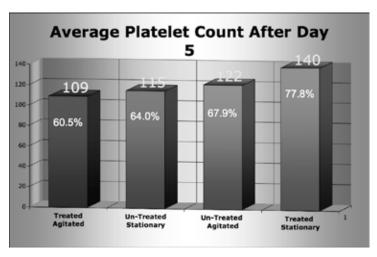


Figure 2.

financial loss. There is a need for more cost effective methods of storage. Platelet agitators/rotators are effective, but these machines are expensive, require maintenance, and take up lab space. Eliminating the agitators/rotators will save money, time and space.

According to the data collected in this study, the stationary HISS treated platelet units yielded promising results having the highest PLT recovery of 77.8%. The current validated storage method which is untreated and agitated PLTs produced 67.9% recovery; these results support the Food Drug Administration's (FDA) standard recovery requirement of 66% (Kaufman, 2006).

Several factors could have influenced the macroscopic precipitants and poor results of the agitated units. First, the rate of agitation may have been too vigorous (70 rotations/min common rate), producing an aggregation cascade. Second, samples treated with HISS may have contained too high of a salt concentration and may have caused proteins in component bags to precipitate. Third, the heat produced by the agitator may have increased the temperature outside of the viable range in the units. More precise temperature regulation and agitating methods should be implemented in future studies.

Bacterial contamination is consistent with penetration of component units. With the contamination of samples randomly dispersed and not uniform throughout treated units, it suggests that contamination came

from some source other than the HISS inoculates. All the samples were inoculated from a reservoir solution. Platelet unit bags would need to be pre-inoculated to avoid contamination.

#### CONCLUSION

Treated stationary units contained the highest retention average of platelet recovery (77.8%). The FDA regulations for storage require that a candidate platelet product must demonstrate at least a 66% of both the recovery and survival of fresh platelets (Kaufman, 2006). According to the results, after further testing with adequate sample size, pre-inoculated bags with HISS, and with FDA validation, HISS could successfully replace the need for mechanical agitators.

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### DR. EZEKIEL R. DUMKE COLLEGE OF HEALTH PROFESSIONS CLINICAL LABORATORY SCIENCES

#### Prolonging Reconstituted Coagulation Control Stability with the Addition of Trebalose

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Eccles Undergraduate Research Scholarship

#### **ABSTRACT**

The objectives of this research were to prolong the expiration of reconstituted coagulation controls used by medical laboratories by adding a unique sugar molecule called trehalose and to determine if the shelf life of lyophilized controls could be extended. Trehalose was chosen because it has previously been shown to protect proteins from denaturation, thus making it appropriate to use with protein-based coagulation controls. Prothrombin (PT) and Activated Partial Thromboplastin Time (aPTT) tests were performed on various levels of coagulation controls at different time intervals over a period of 72 hours. Additionally, controls with a stated expiration date from more than three years ago were also analyzed. This study illustrated that trehalose provided no benefit in prolonging the stability of reconstituted controls. It is important to note that regardless of the addition of trehalose, all controls stayed within +/-2 standard deviations for at least 72 hours, surpassing the manufacturers' recommendation of only eight hours. Furthermore, the expired controls still yielded valid results suggesting the stability of the lyophilized controls could be extended years beyond the stated expiration date. These findings could lead to less waste of coagulation controls and significant monetary savings for medical laboratories.

#### INTRODUCTION

The purpose of this research was to determine if the addition of trehalose to reconstituted coagulation controls would interact with the proteins, and owing to the stabilizing properties of this sugar, prolong 50 · Buckner, et al. · 51

the control stability. This was done by adding trehalose to Level 1 and Level 3 coagulation controls during reconstitution and then performing PT and aPTT testing on these controls at specified time periods. This was the original hypothesis, but after a preliminary trial run, unexpected results obtained led to the formation of a second hypothesis proposing that expired lyophilized controls are stable much longer than their stated expiration date.

Controls are biological samples that are similar in composition to the patient samples being tested. In the case of coagulation testing, sodium citrate plasma is used. A reference range is established for the controls and used to validate that the test is producing accurate results, after which patient samples can be reported. Without these controls, common coagulation tests such as PT and aPTT cannot be validated and reported to clinicians. These coagulation tests are often performed to help a clinician diagnose coagulation disorders or to monitor a patient on anticoagulation therapy.

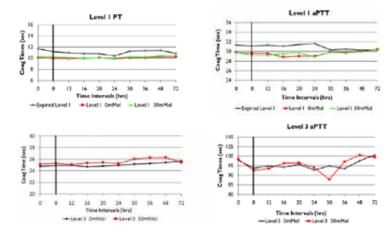
Trehalose is a nonreducing disaccharide sugar molecule containing two glucose units. This sugar is present in some bacteria, fungi, yeast, and invertebrates, but is not found in mammalian cells. Trehalose has been shown to protect cells from environmental stresses such as desiccation, heat, cold, and anoxic situations (Elbein, 2003). Trehalose has also been shown to stabilize proteins and lipids (Holovati, 2007). Research indicates that trehalose has this stabilizing effect due to its structure and chemical makeup which aids in preventing dissociation of proteins. To date, a wide range of uses for trehalose have been discovered and utilized, such as in cosmetics, in foods as a sweetener and preservative, during organ transplants to help keep the organs viable before transplantation, and in desiccated donor platelet units for long term storage, as well as many others (Higashiyama, 2002; Wolkers, 2006). Trehalose has been used in many lyophilized products during the lyophilization process and is shown to help stabilize these once reconstituted (Allison, 1999).

The IL ACL 100 coagulation analyzer and rotors were used to run PT and aPTT tests on the control samples. The following items used were from Pacific Hemostasis: Level 1 and Level 3 coagulation controls, Thromboplastin-D, APTT-XL, CaCl2 0.02M, Calibration Plasma, and Reference Emulsion. Expired Ci-Trol Level 1 controls from Dade Behring were also used.

#### **METHODS**

A preliminary run was done in order to determine an appropriate concentration of trehalose to use with a larger sample volume. Trehalose was used in four different concentrations of expired Level 1 controls from Dade Behring and in date Level 3 controls from Pacific Hemostasis along with a set of controls without trehalose added. The four concentrations used were 10mMol/L, 30mMol/L, 50mMol/L and 70mMol/L. Samples were run in duplicate at nine different time intervals for a 72 hour period. The results demonstrated no statistical difference, thus the median 30mMol/L concentration was used in the actual run with a more extensive sample size.

For the actual run, PT and aPTT tests were performed on 182 samples of Level 1 and Level 3 with and without trehalose, and expired Level 1 controls without trehalose were analyzed at 10 different time intervals over a 72 hour period. Twenty samples of each concentration were used in the baseline run to establish a reference range for each level. Then, for all the other runs, 18 samples of each concentration and control were used. From the data obtained, a reference range was established, the results graphed and analyzed (Figure 1).



**Figure 1.** Results from the larger sample volume analysis. Level 1 PT and aPTT and Level 3 PT and aPTT demonstrating consistent and reliable results beyond eight hours within +/- 2 standard deviations up to 72 hours of analysis.

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#### **RESULTS**

The results from the preliminary run using the four different concentrations of trehalose did not yield any significant difference between each concentration. However, it did indicate two things: that the expired Level 1 controls were still giving precise values and that the controls for both the expired Level 1 and the in date Level 3 controls stayed within a  $\pm$ 2 standard deviation of the established control range for at least 72 hours.

Analysis of the results from the more extensive sample size demonstrated that reconstituted coagulation controls are stable for at least up to 72 hours, which is much longer than the manufacturer's stated eight hours. Results also concluded that the shelf life of lyophilized controls still yield accurate results years after their stated expiration date.

#### DISCUSSION

The primary hypothesis of this study was to see if by adding trehalose to lyophilized controls during reconstitution the reconstituted shelf life could be extended. At the designated time for the preliminary run the Level 1 controls from Pacific Hemostasis had not yet arrived, so expired Level 1 controls from Dade Behring were used to establish an appropriate concentration for a larger sample volume. The data from the preliminary run yielded unexpected results. No statistically significant difference was seen between the four concentrations of trehalose, so for the actual run the median concentration of 30mMol/L was used. The unexpected occurrence was that the Level 1 controls, which had been expired for more than three years, were still giving very reliable results. This finding led to the formation of a second hypothesis of whether or not the shelf life of lyophilized coagulation controls could be extended much further beyond the stated expiration date of the manufacturer. This hypothesis was put to the test with a more extensive sample size.

Both the controls with and without trehalose stayed in range for the analyzed time period, demonstrating that the addition of trehalose did not provide any additional benefit up to 72 hours. Results from the larger sample analysis indicated that reconstituted coagulation controls are stable within the established range of a  $\pm$ 2 standard deviation for

at least 72 hours, which is nine times longer than the manufacturer's stated eight hours (Pacific Hemostasis, 2005). In addition, the expired Level 1 controls from Dade Behring also demonstrated that reliable and accurate results could be obtained within a +/- 2 standard deviation years after their stated expiration date (Figure 1). This was significant considering they had been expired for more than three years. Both of these findings lead to the hope of less control waste and more monetary savings for the hospitals and clinics that perform coagulation testing, because of the demonstrated stability of both the lyophilized and reconstituted controls.

The potential monetary savings from these finding are substantial. One bottle of control from Pacific Hemostasis averages \$2.20. If coagulation testing in a laboratory is performed 365 days a year and two levels of control need to be reconstituted every eight hours, which is three bottles per level of control each day, it would cost \$4,818 per year. This cost is only for controls and does not take into account reagents, rotors or shipping costs. If controls needed to be reconstituted only once per day rather than three times, this would only cost \$1,606 per year, which would be a savings of \$3,212. Because the cost of controls vary from company to company, the savings are even more significant if a laboratory uses controls from Beckman Coulter. Using the same scenario as above, the cost to reconstitute two levels of controls 365 days a year with an average cost of \$6.25 per bottle would be \$13,687 per year. However, if these same controls were reconstituted only once per day, this would cost \$4,563, resulting in a savings of \$9,124 per year. The above examples demonstrate the potential savings if controls were reconstituted every 24 hours. If controls only needed to be reconstituted every 72 hours, the potential savings would be three times these amounts.

Trehalose could be tested in future studies to see if it aids in preventing denaturation of proteins in coagulation controls using a different approach. If trehalose was added to coagulation controls during the lyophilization process during manufacturing, this could possibly be more effective in preventing denaturation of the proteins as the controls are being desiccated. The researchers for this project did not have the means nor resources to do this, but believe this would produce more effective results than adding trehalose to deionized water during reconstitution.

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#### CONCLUSION

The two objectives of this research were to determine if adding trehalose to coagulation controls would extend the reconstituted shelf life and also determine if controls alone, both reconstituted and lyophilized, were stable longer than stated by the manufacturer. Trehalose was chosen because coagulation controls are protein based, and trehalose has unique chemical properties that prevent protein denaturation. This study illustrated that the addition of trehalose to the reconstituted coagulation controls demonstrated no statistical significance in prolonging the coagulation control values. However, the data did show that reconstituted coagulation controls are stable at least nine times longer than the manufacturer's recommended shelf life and stay within a +/- 2 standard deviation after the suggested eight hour outdate. Results also indicated that the lyophilized coagulation controls still produce reliable results and stay within a +/- 2 standard deviation, and that the shelf life of controls can be extended years beyond the stated expiration date. These findings can lead to a significant increase in monetary savings and a decrease in control waste for laboratories.

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#### DR. EZEKIEL R. DUMKE COLLEGE OF HEALTH PROFESSIONS **HEALTH ADMINISTRATIVE SERVICES**

#### Weber State University Students' Knowledge, Attitudes, and Intended Behaviors Associated with the H1N1 Flu Virus

Shanae Teuscher & Michael Olpin

#### **ABSTRACT**

A recent worldwide outbreak of the H1N1 Flu virus has resulted in a great amount of information being passed around about prevention methods, symptoms, and at risk populations. With excessive amounts of information available through several forms of media it's important to know what the public is learning, understanding, and doing in reference to this virus. A voluntary survey was administered to 121 Weber State students focusing on their knowledge, attitudes, and intended behaviors associated with the H1N1 flu virus. Results indicate that the majority of the surveyed students are aware of preventative measures that can decrease the likelihood of spreading this virus. A good portion of the respondents are also aware of which populations are at an increased risk of contracting the H1N1 virus. Of those surveyed, 41% feel that the H1N1 flu virus does not pose a serious threat to their health. Of the H1N1 flu vaccine, 66% of students feel it is effective in preventing the virus; however, 62% aren't sure if it is safe. Of the students, 53% stated that it isn't very likely or at all likely that they'll get the H1N1 flu vaccine. When students feel flu symptoms in the future, 77% of them will most likely stay home from school and work; whereas in the past only 69% would usually stay home. These findings suggest that WSU students are mostly aware of preventative behaviors and at risk populations for contracting the H1N1 virus. It's also evident that the majority of students aren't planning on getting the H1N1 flu vaccine because they feel the H1N1 flu virus isn't a threat to their health and they aren't sure if the vaccine is safe, although since the outbreak of the H1N1 flu virus more students are apt to stay home when feeling flu symptoms, reducing additional spread of the virus.

#### INTRODUCTION

Populations worldwide are affected by the recent outbreak of the H1N1 flu virus. Some populations are affected more than others, but overall it seems that this virus has become a topic of discussion on news channels and amongst the general public.

Attitudes and knowledge are likely to influence health ideas and behaviors in any given population. With this in mind, it's important to recognize the overall attitudes and knowledge of given populations in order to perceive the likely effects the H1N1 flu virus will have within those specific populations.

Here at Weber State University, the spread of the H1N1 flu virus and the possible effects it could have on the education of the students and the performance of the instructors has become a concern. "What if" scenarios are being discussed and action plans tentatively set. However, it may be beneficial and even essential that Weber State University faculty and staff are aware of the current student body's familiarity, opinions, and action plans in reference to the H1N1 flu virus. This information may indicate how well the students have been educated in regards to the H1N1 flu and the likelihood of a campus-wide H1N1 flu virus epidemic.

The intent of this research is to depict Weber State University students' knowledge about the H1N1 flu and its recommended preventative measures. This research will also illustrate WSU students' attitudes regarding the H1N1 flu and the threat it may or may not present, and their behaviors in reference to preventing the contraction of the H1N1 flu virus.

#### Literature Review

This literature review includes studies relating to the H1N1 flu virus. In an effort to ensure that the research was relevant to the most recent outbreak of the virus, current and recently released literature are the focuses of this review. The studies cited researched the publics' knowledge, attitudes, and behaviors associated with the H1N1 flu virus and recommended precautionary H1N1 flu virus measures.

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Recent studies indicate that individuals are more likely to take precautionary action associated with recommended preventative measures for the H1N1 flu virus if they perceive the following: that the H1N1 flu virus is severe, that the risk of contracting the H1N1 flu virus is high, that the outbreak will continue for a long period of time, that people can control their risk of contracting the H1N1 flu, and that specific behaviors are effective in reducing the risk. Those who are uncertain about the H1N1 flu outbreak and who feel that the outbreak has been exaggerated associated with a decreased likelihood of preventative behavior changes (Rubin et al., 2009; Seale et al., 2009).

It has been recommended that in addition to informing the public about the H1N1 flu virus, the efficacy of recommended preventative measures be emphasized. These measures may include hand washing, coughing into a handkerchief or arm, avoiding those with the virus, getting the vaccine, and taking additional special measures if at higher risk for contracting the virus (Clark et al., 2009; Seale et al., 2009; Luby, 2005).

From this review, it is evident that individuals are more likely to adopt behaviors that may reduce their risk of contracting the H1N1 flu virus if they feel that the virus may affect their health and that getting the virus is something they can possibly prevent.

#### **METHODS**

The data needed to determine WSU students' knowledge, attitude, and behavior regarding the H1N1 flu was gathered by means of a written questionnaire. Questions on the survey specifically probed for students' knowledge, attitude, and their past, present, and future intended behavior in relation to the H1N1 and seasonal flu viruses. Student names and identification numbers were in no way indicated on the questionnaire.

Prior to being distributed, the survey was submitted to a WSU Institutional Review Board committee for approval. Following this process, the survey was administered to a sample of convenience consisting of 121 students located in various introductory courses on Weber State's main campus. These courses consisted of Dr. Michael

Olpin's Stress Management class, Dr. Brian Chung's Human Biology course, and Hanalee Hawkin's First Year Experience class. The surveys were filled out on a voluntary basis.

After the surveys were completed the data was compiled in an Excel spreadsheet. The results and conclusions noted in this document were taken from the numbers and trends indicated on the spreadsheet.

#### **RESULTS**

In total, 121 students were surveyed; 52 male and 69 female. The marital status' included 98 single, 22 married, and one divorced. Two of the surveyed students had three children, five had one child, and the rest of the students had zero children. The class breakdown of the surveyed students is shown in figure 1. Declared and intended majors of the surveyed students are shown in figure 2; with the remaining students focusing their studies in other colleges and majors at WSU.

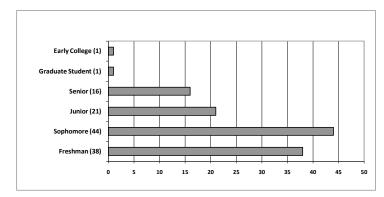


Figure 1. Class Breakdown

The students surveyed were asked to indicate whether they had any asthma or other respiratory health issues. Twelve indicated that they did and the rest said they did not.

A question was included to find out if students believed that they only needed to stay home from school or work if lab tests indicated that they positively had the H1N1 flu virus. The percentage of students that

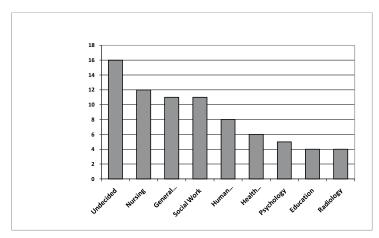


Figure 2. Intended Majors

indicated that people should stay home only if lab results were positive for the actual virus was 42%, while 58% felt that positive lab results weren't necessary to justify staying home.

Students were asked to list three things that could be done to help prevent or decrease the likelihood of getting the H1N1 flu. Of the answers given, 89% of those surveyed listed hand washing, 30% listed avoiding contact with sick people, 24% staying home when feeling sick, 22% covering mouth when coughing and sneezing, 15% suggested vaccination, 13% avoid face touching, 12% sanitizer use, 9% suggested healthy eating, and 9% wrote down staying home. Numerous other answers were also given; however, those with percentages below 9% are not listed here.

In two separate questions students were asked whether they felt that the seasonal flu vaccine and the H1N1 flu vaccine helps prevent people from getting the H1N1 flu virus. Twenty percent felt that the seasonal flu vaccine helps prevent the H1N1 flu virus while eighty percent felt that it doesn't help prevent it. Sixty-six percent felt that the H1N1 vaccine helps prevent the H1N1 flu virus while thirty-three percent felt that it doesn't. One student did not mark whether they felt that the H1N1 flu vaccine does or doesn't help prevent the H1N1 flu virus.

When asked whether students felt that the H1N1 flu virus poses a serious threat to their health, 6% strongly disagreed, 35% disagreed, 31% weren't sure, 22% agreed, and 6% strongly agreed.

Ten different populations were listed after the following question: "Populations at risk for contracting the H1N1 flu virus include which of the following:." This question was answered as indicated in table 1 below.

**Table 1.** Populations At An Increased Risk for H1N1 Contraction vs. Populations Perceived To Be At An Increased Risk

Populations at an increased risk for contracting the H1N1 flu virus	Correct Answers (CDC, 2009)	# of Students Who Marked This Answer	% of Students Who Marked This Answer
Adults 65+	X	90	75%
Adults between 25 and 55		66	55%
Asthma and other respiratory patients	X	93	77%
Children	X	110	91%
Infants	X	108	90%
Individuals who are obese	X	50	42%
Individuals with blood, kidney, liver disorders	X	61	51%
Individuals with cancer	X	64	53%
Individuals with weakened immune systems	X	113	94%
Pregnant women	X	101	84%

Students were asked to indicate the likelihood that they would get the H1N1 flu vaccine. Nineteen percent said not at all likely, 34% not very likely, 35% somewhat likely, 7% very likely, and 5% had already received it. When asked whether they felt that the H1N1 vaccine is safe students answered as follows: 37% said yes, 12% said no, and 50% said they weren't sure.

In the past, when the surveyed students have felt flu symptoms, 33% indicated they have always stayed home from school and/or work, 36% have usually stayed home from school and/or work, 17% have occasionally stayed home, 4% rarely stayed home, 2% never stayed home, and 7% have never had the flu.

The final question students responded to inquired about their future plans to stay home from school and work when they feel flu symptoms. The students answered as follows: 31% responded definitely, 46% probably, 18% probably not, and 4% definitely not.

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#### DISCUSSION

Several conclusions can be made based on the results of this study. Consequently, the following addresses each of the topics inquired about and the assumptions that have been drawn from the data's trends.

Just over half of the surveyed students marked that lab results positively indicating that one actually has the H1N1 flu virus aren't necessary to rationalize staying home when not feeling well. In the past, nearly seventy percent of these students have usually or always stayed home when they've had the flu. However, in the future seventy-seven percent of the students stated that they probably or definitely will stay home from school and work when they have flu-like symptoms.

This data shows that the majority of these students plan to stay home when they feel flu symptoms, regardless of whether or not they are formally diagnosed with the H1N1 flu virus. Additionally, it's evident that the outbreak of the H1N1 flu virus has encouraged a greater percentage of the students to consider staying home when they feel flulike symptoms.

Although students acknowledged that there are numerous populations at an increased risk for contracting the H1N1 flu virus, some of these populations are more known than others. A large portion of the students know that infants, children, and people with weakened immune systems are at an increased risk and a good portion are aware of the increased risk of adults aged sixty-five or older, individuals with asthma or other respiratory issues, and pregnant women. Of the listed populations for the respondents to choose from, only one option was incorrect according to the CDC. Adults between the ages of twenty-five and fifty-five are not at an increased risk for contracting the H1N1 flu virus but fifty-five percent of students marked this as an at-risk population.

This data points out that students are fairly aware of who is at a greater risk for this virus but since there was only one incorrect option this could also show that students may have just marked all the options that sounded like viable answers. Although more than half of the students marked the incorrect population, it may not be a bad thing for them to think that adults under the age of fifty-five are at risk; since this may lead to them taking and encouraging others to take precautionary measures in order to avoid the flu.

Survey respondents listed things that can be done to help prevent or decrease the likelihood of getting the flu. Ninety-six percent listed at least one accurate preventative measure; with the majority listing more than one. Many students are aware that hand-washing, covering their mouth when coughing or sneezing, and avoiding those with the virus will reduce their chances of getting sick. This data leads me to believe that students have been well educated about preventing the spread of the H1N1 flu virus (in addition to other viruses and illnesses).

When asked whether they felt that the H1N1 flu virus poses a serious threat to their health, the majority of WSU students specified that they didn't feel that it poses a serious threat and nearly a third indicated that they weren't sure whether or not it does. Since it's evident that personal attitudes and beliefs influence the individual's behavior (Rubin et al, 2009; Seale et al, 2009), it might be said that although students are aware of measures that may assist in preventing the contraction of this virus, many aren't actively adopting these preventative behavior changes. Depending on the local prevalence of the H1N1 flu virus this could potentially lead to an outbreak of flu cases.

A fifth of the students are under the impression that the seasonal flu vaccine helps prevent people from getting the H1N1 flu virus while two-thirds feel that the H1N1 vaccine helps prevent the virus. Although the majority feel the H1N1 vaccine can help prevent the virus, over half of the students feel that the H1N1 flu vaccine either isn't safe or they're unsure if it's safe. Over half of the students stated that it isn't at all likely or very likely that they'll get the H1N1 vaccine. As a result of this data, it's likely that even though students feel the H1N1 vaccine may prevent the H1N1 virus they won't get the vaccine due to the belief that it is unsafe and also due to believing that the virus does not pose a great enough threat to their health.

#### CONCLUSION

These findings suggest that WSU students are mostly aware of preventative behaviors and at risk populations for contracting the H1N1 virus. It's also evident that the majority of students aren't planning on getting the H1N1 flu vaccine because they feel the H1N1 flu virus isn't a threat to their health and they aren't sure if the vaccine is

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safe. Finally, since the outbreak of the H1N1 flu virus more students are apt to stay home when feeling flu symptoms, reducing additional spread of the virus.

### **AUTHORS NOTE**

It is the hope of the author of this research for other researchers to use this acquired data for their own scholarly purposes. In addition to many other purposes, this data may be used to estimate the possible influence the H1N1 flu will have on WSU students, their education, and their personal lives. It may also be used to ensure that future campus health campaigns will be effective in educating and inspiring healthy behaviors among WSU students.

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## COLLEGE OF SCIENCE MICROBIOLOGY

## Antibiotic Susceptibility in Halophiles Isolated from the Great Salt Lake

Brandon Cassel, Michele D. Zwolinski & Craig J. Oberg

### **ABSTRACT**

Halophile isolates from the South Arm of the Great Salt Lake (GSL) were analyzed for susceptibility to common antibiotics. Twenty one strains were tested against tetracycline, ampicillin, vancomycin, doxycycline, sulphamethazole/trimethoprim, and oxacillin using the disc assay procedure. Results showed that within the isolate collection were organisms with resistance to all of the antibiotics tested. All of the isolates were resistant to tetracycline, vancomycin, oxacillin, and doxycycline. More study is needed to determine the resistance mechanisms and the risk for transfer of resistance to human and animal enteric pathogens. Most of the Idiomarina-like isolates (S3, S6, and S11) were sensitive to ampicillin, as were two Halomonas-like isolates (S15 and S31). The antibiotic sensitivity profiles may be useful for resolving strain differences between organisms of the same species.

### INTRODUCTION

The Great Salt Lake (GSL) is the fourth largest terminal lake in the world, with an average salt concentration of about 12%, making it four times more salty than seawater. This provides a perfect environment for halophilic bacteria and serves as habitat for the brine shrimp that are food for many migratory birds (US Geological Survey, 2009). Since there are no outlets for the lake, everything that flows in remains, including sewage waste and residues from water/sewage treatment facilities. The high salt concentration inhibits the growth of most microbial pathogens. However, studies have shown that enteric bacteria such as *Enterococcus* can survive in the lake water (Kagie et al., 2008). These organisms enter the lake through sewage effluent into Farmington Bay which then drains into the South Arm. The role of migratory

birds and brine shrimp in the spread of pathogens is suspected, but not known. Birds may be a source of pathogens in the lake, but they are also at risk of infection. Brine shrimp infected with enteric pathogens could impact the aquiculture that depends on the brine shrimp industry (Kagie et al., 2008).

Although most halophilic organisms are not considered pathogens, they could transfer antibiotic resistance genes to enteric bacteria or other opportunistic pathogens present in the GSL. Antibiotic resistance to most therapeutic antibiotics has been demonstrated in microorganisms naturally found in soil, fresh water, and marine habitats (D'Costa et al., 2006; Martinez, 2008; Riesenfeld et al., 2004). This resistance is a natural result of interactions among organisms; some microorganisms even consume antibiotics as a source of carbon and energy (Dantas, et al., 2008). Naturally antibiotic resistant halophiles in the GSL could be a reservoir of resistance genes that could be transferred to pathogenic organisms.

Identifying antibiotic sensitivity is important for describing and distinguishing between newly isolated bacterial strains. Several studies have been done on antibiotic resistance of halophiles, but the majority were from marine environments containing only 3% NaCl; far less than the GSL. Hence, few of these organisms were true halophiles. Pathogenic marine vibrios from seafood, for example, have shown sensitivity to oxolinic acid, trimethoprim-sulphamethoxazole, doxycycline, flumequine, cefotaxime, nalidixic acid, ciprofloxacin, and several beta-lactam antibiotics (Ottaviani et al., 2000). In organisms isolated from the Java Sea, an environment more similar to the GSL than marine systems, Joseph et al. (1978) found halophilic vibrios resistant to ampicillin but susceptible to chloramphenicol and tetracycline. Lobova et al. (2000) found ampicillin resistant halotolerant bacteria from the saline environment of Shira Lake. Salinibacter, a true halophile, showed some inhibition by chloramphenicol, erythromycin, and deoxycholate (Bardavid & Oren, 2008).

The purpose of this research project was to describe the antibiotic sensitivity profiles of halophiles isolated from the GSL and to determine if potential for lateral gene transfer of antibiotic resistance genes between halophiles and enteric bacteria exists.

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### **METHODS**

### Culture Isolation

Samples were collected along the north shore of Bridger bay on Antelope Island, which is located on the South Arm of the GSL. Sediment and water samples were collected 3-5 meters from the shoreline. Individual bacterial strains are maintained in the Weber State University Halophile Culture Collection.

## Culture Media and Preparation

Halophile agar (HB agar) was prepared using the following formulation: NaCl, 120 g; agar, 15 g; MgSO4-7H2O, 25 g; casamino acids, 5 g; yeast extract, 5 g; proteose peptone, 2.5 g; trisodium citrate, 3 g; KCl, 2 g; agar, 20 g per liter of distilled water (Atlas, 1993). The pH was adjusted to  $7.2 \pm 0.2$  prior to sterilization. Isolates were grown in halophilic broth (HB) of the same formulation without agar. The HB broth was inoculated with a 48-hour culture and incubated for 24 hours in a 30°C water bath. Isolate identity was determined previously by 16SrRNA sequencing (Bowcutt et al., 2007).

### Antibiotic Disc Assay

The antibiotic assay was done on HB agar plates, each inoculated with one of 21 different halophilic broth cultures. Each isolate, after being incubated in HB at 30oC overnight, was inoculated onto plates with a sterile cotton swab to create a bacterial lawn. After inoculation, six sterile 10 mm diameter antibiotic discs (Sensi-Disc, Becton, Dickinson and Co., Sparks, MD) containing different antibiotics were applied to the plate using a BBL Sensidisc Dispenser. The antibiotics tested were tetracycline (30  $\mu g$ ), ampicillin (10 $\mu g$ ), vancomycin (30 $\mu g$ ), oxacillin (1 $\mu g$ ), sulphamethazole (23.75  $\mu g$ )/trimethoprim (1.25 $\mu g$ ), and doxycycline (30  $\mu g$ ). Each halophilic isolate was tested in triplicate. Plates were incubated at 30° C for 48 hours. The areas around the disks with no bacterial growth were considered the zones of inhibition and were measured in mm for each antibiotic. Isolates were scored as sensitive (S), intermediate (I), or resistant (R), according to the scale established by the disc manufacturer for each antibiotic (Table 1).

Each antibiotic has its own mechanism for inhibiting bacterial growth. Some antibiotics target some bacteria better than others. The general characteristics of the antibiotics used in this study are summarized in Table 1.

**Table 1.** List of antibiotics, scale used to determine resistance (R) or sensitivity (S), and their modes of actions against bacteria.\*

Antibiotics	R	S	Mode of Action
Vancomycin	< 14 mm	>17 mm	Inhibits cell wall synthesis by binding to terminal D-alanyl-D-alanin ends of the NAG and NAM peptides in the peptidoglycan matrix. It also alters membrane permeability and RNA synthesis.
Tetracycline	< 14 mm	> 19 mm	Inhibits action of the prokaryotic 30S ribosome, by binding the 16S rRNA thereby blocking the aminoacyl-tRNA.
Ampicillin	< 11 mm	> 14 mm	Competitively inhibits transpeptidase which inhibits the third stage of cell wall synthesis thus leading to cell lysis.
Oxacillin	< 10 mm	> 13 mm	Inhibits the synthesis of the peptidoglycan layer of bacterial cell walls by inhibition of the Penicillin Binding Proteins preventing the final cross linking (transpeptidation) of the peptidoglycan layer, disrupting cell wall synthesis.
Sulphamethoxazole/ Trimethoprim	< 10 mm	> 16 mm	Sulphamethoxazole inhibits dihydropteroate synthetase which inhibits dihydropteroic acid in the folate synthesis pathway. Trimethoprim interferes with the action of bacterial dihydrofolate reductase inhibiting tetrahydrofolic acid which is used in thymidine and uridine that is necessary in DNA replication and transcription.
Doxycycline	< 12 mm	> 16 mm	Doxycycline is a semi-synthetic tetracycline so it's mode of action is closely related to tetracycline above.

<sup>\*</sup>Information on drugs obtained per package inserts for each drug: Vancmycin-Viropharma, Tetracycline-Pfizer, Sulphmethoxazole/Trimethoprim-Roche, Doxycycline-Pfizer, Oxacillin-Baxter, Ampicillin-Sandoz.

### **RESULTS**

Each halophilic isolate was resistant to at least one antibiotic. All were resistant to tetracycline, doxycycline, vancomycin, and oxacillin (Tables 2 and 3 on following pages). Sensitivity to ampicillin was seen among the *Idiomarina* and some of the *Halomonas* isolates. Several of the *Salinivibrio* isolates were intermediately sensitive to sulphamethazole/trimethoprim. All of the isolates, except S29, were gram-negative gamma-*Proteobacteria*. S29 was related to *Salinicoccus*, a gram-positive bacteria, however, this isolate was resistant to all of the tested antibiotics.

### DISCUSSION

Overall, the halophilic isolates were resistant to the antibiotics tested (Tables 1 and 2). It was not surprising that halophilic microorganisms displayed resistance to some therapeutically used antibiotics. Antibiotic resistance is common in organisms from natural environments such as soil, and fresh and marine water (D'Costa et al., 2006; Martinez, 2008;

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**Table 2.** List of halophilic isolates, their strain number, and the zones of inhibition in mm with standard deviation caused by interaction with each antibiotic.

	Te	Am	Va	Ox	SXT	D
SV2	5 ± 0	9 ± 2	0	0	12 ± 2	0
Salinivibrio costicola SV4				· ·		
Salinivibrio costicola	1 ± 1	6 ± 1	0	0	11 ± 1	0
SV9 Salinivibrio costicola	1 ± 1	2 ± .5	0	0	11 ± 1	0
SV14 Salinivibrio costicola	0	11 ± 1	0	0	10 ± 1	0
SV17 Salinivibrio costicola	0	9 ± 1	0	0	11 ± 1	0
SV22 Salinivibrio costicola	0	0	0	4 ± 0	5 ± 1	0
SV24 Salinivibrio costicola	0	3 ± 1	0	0	3 ± 0	0
S3 Idiomarina sp.	0	20 ± 1	0	2 ± 0	10 ± 1	0
S6 Idiomarina sp.	0	21 ± 1	0	5 ± 1	2 ± 1	0
S11 Idiomarina loihiensis	0	18 ± 2	0	3 ± 0	2 ± 1	0
S21 Idiomarina sp.	0	0	0	0	10 ± 2	0
S29 Salinicoccus sp. D23.3	0	10 ± 0	0	1 ± 0	8 ± 1	0
S15 <i>Halomonas</i> sp.	0	27 ± 1	0	0	17 ± 1	0
S25 Halomonas ventosae	0	5 ± 1	0	0	11 ± 1	0
S26 Halomonas sp. LCKS0	0	2 ± 1	0	1 ± 0	2 ± 0	0
S27 <i>Halomonas</i> sp. LCKS0	0	8 ± 1	0	0	2 ± 0	0
S31 <i>Halomonas</i> sp. AJ275	0	25 ± 1	0	7 ± 0	7 ± 1	3 ± 0
S32 Halomonas sp. LCKS0	0	12 ± 1	0	0	3 ± 1	0
S33 Halomonas sp. LCKS0	0	9 ± 1	0	0	3 ± 1	0
S34 Halomonas sp. LCKS0	0	9 ± 1	0	1 ± 0	9 ± 1	0
S23 Unknown	0	5 ± 1	0	0	2 ± 1	0

Te-Tetracycline 30  $\mu$ g, Am-Ampicillin 10 $\mu$ g, Va-Vancomycin 30 $\mu$ g, Ox-Oxacillin 1 $\mu$ g, SXT-Sulphamethazole 23.75  $\mu$ g/Trimethoprim 1.25 $\mu$ g, D-Doxycycline 30  $\mu$ g.

Riesenfeld et al., 2004). The ability of many environmental microorganisms to deactivate, avoid, destroy, or consume antibiotics is a natural consequence of living in complex communities of organisms that interact through chemical signaling (Dantas et al., 2008).

Isolates from the GSL were largely resistant to vancomycin and oxacillin, both inhibitors of peptidoglycan synthesis in growing **Table 3.** Antibiotic resistance and sensitivity profiles of halophilic isolates. Organisms were coded as resistant (R), intermediate (I), or sensitive (S) according to the scale determined by the antibiotic disc manufacturer.

**Table 3.** Antibiotic resistance and sensitivity profiles of halophilic isolates. Organisms were coded as resistant (R), intermediate (I), or sensitive (S) according to the scale determined by the antibiotic disc manufacturer.

	Te	Am	Va	Ox	SXT	D
Resistant if < mm	14	11	14	10	10	12
Sensitive if > mm	19	14	17	13	16	16
SV2	R	R	R	R	I	R
Salinivibrio costicola	K	K	K	N.	1	K
SV4	R	R	R	R	I	R
Salinivibrio costicola						
SV9	R	R	R	R	I	R
Salinivibrio costicola						
SV14	R	I	R	R	I	R
Salinivibrio costicola SV17			<u> </u>			
Salinivibrio costicola	R	R	R	R	I	R
SV22						
Salinivibrio costicola	R	R	R	R	R	R
SV24						
Salinivibrio costicola	R	R	R	R	R	R
S3						
Idiomarina sp.	R	S	R	R	I	R
S6			_	_		_
Idiomarina sp.	R	S	R	R	R	R
S11		c	n	n	ъ	n
Idiomarina loihiensis	R	S	R	R	R	R
S21	R	R	R	R	I	R
Idiomarina sp.	K	K	K	I.	1	K
S29	R	R	R	R	R	R
Salinicoccus sp. D23.3						
S15	R	s	R	R	s	R
Halomonas sp.						
S25	R	R	R	R	I	R
Halomonas ventosae						
S26 Halomonas sp. LCKS0	R	R	R	R	R	R
S27	-		1			
Halomonas sp. LCKS0	R	R	R	R	R	R
S31	_		1			
Halomonas sp. AJ275	R	S	R	R	R	R
S32			_	_	_	_
Halomonas sp. LCKS0	R	I	R	R	R	R
S33	R	R	R	R	R	R
Halomonas sp. LCKS0	к	К	К	К	К	K
S34	R	R	R	R	R	R
Halomonas sp. LCKS0	К	К	К	Л	К	Л
S23	R	R	R	R	R	R
Unknown	K	K	Λ	Λ	A	K

microorganisms (Table 1). These antibiotics are generally used to treat gram-positive bacterial infections. Vancomycin is most often used to treat multi-drug resistant strains of *Staphylococcus aureus*. Because most of the isolates were gram-negative, these antibiotics are expected to be less effective.

Interestingly, some strains of *Idiomarina* and *Halomonas* were sensitive to ampicillin, another peptidoglycan synthesis inhibitor. Ampicillin is a broad-spectrum antibiotic, targeting gram-positive and gram-negative infections. Among other halophiles, ampicillin resistance is variable.

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Salinicoccus halodurans, isolated from a saline soil in China and related to isolate S29, was also sensitive to ampicillin (Wang et al., 2008). Other studies, however, have shown that the halophiles from the Java Sea (Joseph, et al., 1978) and Shira Lake (Lobova, et al., 2002) were found to be resistant to ampicillin.

The GSL isolates were also resistant to broad-spectrum antibiotics including tetracycline, doxycycline, and sulphamethazole/ trimethoprim, a folic acid inhibitor (Table 3). The mechanisms of resistance to these antibiotics is unclear, but are likely different than those found in non-pathogens. The patterns of antibiotic resistance and sensitivity may be useful for further characterizing our isolate library with greater resolution than 16S rRNA gene sequencing alone.

### CONCLUSION

Halophilic microorganisms could serve as a reservoir of antibiotic resistance genes that could be passed to other human or wildlife pathogens through lateral gene transfer. Resistance to a wide range of antibiotics suggests mechanisms that include an effect of high salt concentrations on the antibiotics or the metabolic mechanisms are used for survival in a high saline environment. These mechanisms may or may not be genetically coded and able to be passed to other microorganisms. However, considering the local sewage plants, antibiotics could be present in the GSL, driving selection for resistant strains of halophiles. Further, animal waste in runoff water and migratory birds are potential sources of both enteric bacteria and antibiotics. Enteric bacteria such as Escherichia coli and Enterococcus sp. are a source of disease in humans, birds, and other animals. If they obtain antibiotic resistant genes from halophiles, this could reduce antibiotic effectiveness. Further studies must be done to see if halophiles readily transfer genes to enteric organisms and to further describe the nature of antibiotic resistance in halophiles.

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## COLLEGE OF SCIENCE MICROBIOLOGY

## Method Development to Study Prophage Induction in Halophiles from the Great Salt Lake

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Eccles Undergraduate Research Scholarship

### **ABSTRACT**

Limited research has been performed on lysogenization in halophilic bacteria found in moderately halophilic (8-12% NaCl w/v) environments. Thirteen halophilic bacterial strains were isolated from the South Arm of the Great Salt Lake. Broth cultures of each strain were exposed to either ultraviolet (UV) radiation at various time intervals or to mitomycin C to cause prophage induction. Various growth conditions including NaCl concentrations, nutrient levels, and incubation temperatures were also examined for their effect on prophage induction. A comparison between untreated control tubes and UV treated tubes showed much slower growth in UV-treated tubes over time for many cultures, indicative of prophage induction. Of note, many halophilic strains were very sensitive to UV treatment, which was surprising since these isolates are constantly exposed to UV light in their environment. Spot tests utilizing the soft agar overlay method with filtrates from UV and mitomycin C treated cultures have been inconclusive. Growth curve results suggest the existence of prophage in some halophiles. Continued method development to induce prophage induction will include varying the concentration of mitomycin C and utilizing modifications in UV exposure intensity.

### INTRODUCTION

The Great Salt Lake (GSL) is a hypersaline environment ideal for moderate to extremely (12-26% NaCl w/v) halophilic microorganisms (Baxter et al., 2005). A high concentration of nutrients, intense

seasonal sunlight exposure, and wind-generated aeration allows the GSL to support a significant number of halophilic microorganisms, primarily bacteria and archaea (Ventosa, 2006). These microorganisms contribute significantly to the ecological balance within saline lakes since they form the base of the food web, act as decomposers, and facilitate nutrient recycling, thus playing a significant role in trophic networks within the lake and surrounding environment (Dyall-Smith et al., 2005; Jiang and Paul, 1998). Many environmental bacterial isolates harbor lysogenic bacteriophages (phage). The lysogenic bacteriophage genome (termed prophage in this context) is integrated into the bacterial genome for long-term survival but can be induced, resulting in replication of the phage and eventual lysis of the infected bacterium. Activation of DNA repair mechanisms within host bacteria due to environmental factors or chemical exposure is generally responsible for initiation of the lytic phase of the phage life cycle with release of phage into the environment to infect other bacteria (Cochran & Paul, 1998). Following bacterial cell lysis in a laboratory setting, the phage can be collected and used to infect similar bacteria, providing a means to study the phage.

The role of lysogenic prophages and their impact on microbial populations in these unique environments has been an area of interest for those who study saline-lake ecology (Daniels & Wais, 1990). Lysogenic phages can integrate into the chromosomal DNA of host bacteria so the integrated phage genome (prophage) is replicated during cell division, thus perpetuating the phage (Jiang & Paul, 1998; Williamson et al., 2002). Lysogenic bacteria (bacteria that harbor a prophage) also appear to have a competitive advantage over their non-lysogenic counterparts under nutrient-limited conditions (Jiang & Paul, 1998).

Bacteria predation by bacteriophages may play a significant role in controlling bacterial populations, but this effect in hypersaline environments is unknown (Dyall-Smith et al., 2005). Bacteriophages are thought to help maintain equilibrium between bacteria and resources for growth, and between related bacterial cultures. There are few reports of bacterial predation by bacteriophage in the GSL (Kauri et al., 1991). One study in the Dead Sea found 1 x  $10^7$  virus particles per mL of water (Oren et al., 1997). A follow-up study suggested the titer of virus particles in the Dead Sea changed seasonally, falling as low as  $1 \times 10^4$  per mL during winter (Ventosa et al., 1998). In marine environments, it is estimated that more than 40% of bacterial isolates

contain inducible prophage, suggesting this bacteriophage survival strategy is widespread in moderately halophilic aqueous environments (Jiang & Paul, 1998).

In this study, bacterial isolates from the GSL were tested for the presence of prophages using the mutagenic agents UV light and mytomicin C (Jiang & Paul, 1998; Cochran & Paul, 1998). In addition, two induction procedures using UV light, along with variations in UV exposure times, and a method using mitomycin C were tested in an attempt to develop an effective technique for prophage induction in halophilic bacteria.

### **METHODS**

### Culture Isolation

Samples were collected along the north shore of Bridger Bay on Antelope Island, located in the South Arm of the GSL. Sediment and water samples were collected four meters from the shoreline, and plated on halophilic agar (Atlas, 1993).

### Culture Media and Preparation

Halophile agar (HA) was prepared using the following formulation (NaCl, 120 g; agar, 10 g; MgSO $_4$ ·7H $_2$ O, 10 g; casein hydrolysate, 5 g; KCl, 5 g; trisodium citrate, 3 g; KNO3, 1 g; yeast extract, 1 g; CaCl2·6H2O, 0.2 g per liter of distilled water) with trisodium citrate substituted for disodium citrate (Atlas, 1993, p.421). Halophile broth (HB) was prepared using the same formulation without agar addition. Halophile broth tubes were inoculated with a 48-hour culture and incubated for 24 hours in a 30°C water bath. Other media used in the induction methods included 2X HB broth (NaCl, 120 g; MgSO $_4$ , 50 g; casamino acids, 10 g; yeast extract, 10 g; proteose peptone, 5 g; KCl, 4 g; trisodium citrate, 6 g per liter distilled water) and MgSO $_4$  buffer (MgSO $_4$ , 25 g; NaCl, 120 g per liter distilled water).

## Prophage Induction

Induction Method 1- UV exposure of cell concentrates. Halophilic cultures were incubated overnight at 30°C and then centrifuged at 3,000 rpm for 10 minutes in a Beckman Model GS-6 (Beckman Inc., Palo Alto, CA). The pellet was resuspended in 10 ml of sterile

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0.1M MgSO, and then centrifuged at 3,000 rpm for 10 minutes. After the washing step, the pellet was resuspended in 10 ml of sterile 0.1M MgSO<sub>4</sub>. Seven tubes of each culture were individually transferred to sterile 100 x 15 mm polystyrene Petri dishes (VWR International, West Chester, PA). The UV light used for both Method 1 and Method 2 was a 30-watt light bulb with a wavelength of 252 nm. One tube of each culture was swirled by hand 32.0 cm away from the UV light source for either 20, 30, 40, 50, 60, or 80 seconds. Irradiated cultures were transferred to test tubes containing sterile 2X HB (12% NaCl) and incubated in a 28°C water bath (light exposure not controlled). A control culture for each halophilic strain was prepared with 0 seconds UV exposure. Optical density (OD<sub>600</sub>) readings (Spectronic 20D+, Thermo Fisher Scientific Inc., Waltham, MA) were taken at 0.5, 1, 1.5, 2, 2.5, 3, 3.5, and 4 hours. For the growth curve of any culture, if there was an initial increase in the  $\mathrm{OD}_{600}$  readings followed by a decrease in the  $OD_{600}$  for a period of an hour or more; this was interpreted as evidence of prophage induction. When a prophage is induced in a cell, the lytic replicative cycle is initiated and eventually results in lysis of the bacterial cell. This results in an observable decrease in  $OD_{600}$  if a sufficient portion of the bacterial population is infected by the phage. The effect of various exposure times on individual cultures was also examined in the same manner.

Induction Method 2- UV exposure of broth cultures. Halophile cultures were incubated overnight at 30°C and then back diluted to an  $\mathrm{OD}_{600}$  reading between 0.5 and 0.6 with sterile HB broth. Diluted cultures were transferred to sterile  $100 \times 15$  mm polystyrene Petri dishes and swirled by hand 19.0 cm away from the UV light source for either 20, 30, 40, 50, or 60 seconds. A control culture for each halophilic strain was prepared with 0 seconds UV exposure. UV exposed cultures were incubated at  $28^{\circ}\mathrm{C}$  in the dark using a covered incubator.  $\mathrm{OD}_{600}$  readings were taken at 2, 4, 6, 20, 22, and 25 hours. Growth curve results for each culture at each exposure time were interpreted as described in Method 1.

Induction Method 3 - Mitomycin C treatment. Overnight cultures grown at 30°C and back diluted to  $OD_{600}$  readings between 0.5 and 0.6 with sterile HB broth. Cultures were then treated with mitomycin C (Sigma-Aldrich Corp., St. Louis, MO) to a final mitomycin C concentration of 0.5  $\mu$ g/ml and then incubated in the dark at 28°C in a covered incubator.  $OD_{600}$  readings were taken at 2, 4, 6, 20, 22, and 25 hours. Growth curve results for each culture were interpreted as

previously described. Growth curves for each culture used in each method were then compared to determine which method could cause prophage induction in individual cultures.

### Spot Test Method

Selected UV and mitomycin C treated cultures from all three induction methods were filtered through  $0.45\mu m$  filters. Next, 0.1 ml of the appropriate halophilic bacterial host culture was transferred to 3 ml of soft HB agar (0.8% agar) and mixed thoroughly. After mixing, contents of the soft agar tube were poured onto a Petri plate containing HB agar. Finally, 0.1 ml of the phage filtrate was placed on the solidified soft agar surface. Plates were then incubated at  $37^{\circ}C$  for 72 hours.

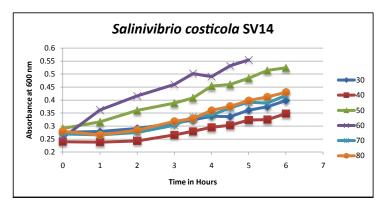
### **RESULTS**

Three methods were evaluated for their potential to cause prophage induction in halophilic bacteria isolated from the GSL. Two methods utilized UV light as the inducing agent while the third used the antibiotic, mitomycin C, as the inducing agent. Several other modifications to general prophage induction methods used for non-halophilic bacteria were also incorporated in the test methodology, including incubation in the dark (to prevent light repair mechanisms for damaged DNA from being used by exposed cells) and changes in incubation temperature (to more closely approximate individual culture's optimal incubation temperatures).

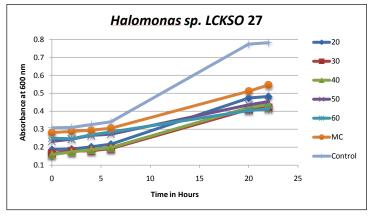
Method 1 was an adaptation of a prophage induction method used for lactic acid bacteria. Results showed very minimal growth of treated cultures out to 4 hours and no decrease in cell concentration was observed out to 4 hours, indicating that no prophage induction had taken place (Figure 1).

Method 2 is a modification of methodology used to induce prophage in marine isolates (Jiang & Paul, 1998) and seemed to alleviate many of the difficulties encountered with Method 1. In this method, cells were grown to higher densities then back diluted prior to UV exposure. Other changes from Method 1 included taking readings out to 25 hours of incubation and incubating cultures in the dark. A number

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**Figure 1.** *Salinivibrio costicola* SV14 growth curve utilizing induction Method 1. Lines represent UV exposure time in seconds.



**Figure 2.** *Halomonas* sp. S27 growth curve after induction Method 2. Lines represent UV exposure time in seconds.  ${\rm OD}_{600}$  readings taken at 2, 4, 6, 20, 22, and 25 hours of irradiated culture with no drop after 25 hours. The control absorbance was higher than the treated organisms suggesting some lysogenization.

of halophilic isolates showed characteristic growth curves for phage induction not observed with concentrated cultures treated using Method 1 (Figure 2).

In Method 3, the use of mitomycin C as an inducing agent seemed to provide results most consistent with prophage induction. After 6 hours, the growth curves of mitomycin C treated isolates showed a

considerably lower OD $_{600}$  reading than untreated controls (Figure 3). While not all halophilic isolates responded in the same manner to this concentration of mitomycin C, use of this inducing agent resulted in growth curves that matched growth curves of lysogenic marine bacteria (Jiang & Paul, 1998).

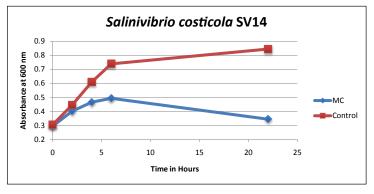


Figure 3. Salinivibrio costicola SV14 growth curve after induction Method 3. Lines represent UV exposure time in seconds. OD $_{600}$  readings taken at times 2, 4, 6, 20, 22, and 25 hours. After 3 hours, the absorbance of the mitomycin C treated culture started to drop while the control organism continued to increase.

An attempt was made to use filtered supernatant from induced cultures to observe bacteriophage plaque formation by the Spot Test. After  $\mathrm{OD}_{600}$  readings were taken, UV and mitomycin C treated cell suspensions were centrifuged and then the supernatant was filtered to remove debris. Spot tests to confirm those lytic bacteriophages were released during induction provided only very tentative results with several faint plaques observed on soft agar overlay plates.

When Methods 1 and 2 were performed, it was observed that the initial  $\mathrm{OD}_{600}$  (time 0) of most halophilic cultures was lower than that culture's adjusted OD just prior to the UV treatment. A comparison was done between non-halophilic cultures and halophilic isolates. A significant decrease in absorbance was noted for halophilic cultures immediately after UV exposure that suggested cell lysis, an observation not seen with non-halophile control bacteria (Table 1).

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Table 1. Comparison of UV exposure between a halophile (S21) and non-halophilic
bacteria. ODs were taken just prior to UV exposure and immediately after UV exposure.

Organism	UV exposure (sec)	Pre-UV	Post-UV
Staphylococcus	40	0.412	0.422
Streptococcus	40	0.772	0.770
Escherichia coli	40	0.690	0.706
S21 (Idiomarina sp.)	0	0.672	0.704
	20	0.706	0.472
	30	0.698	0.482
	40	0.708	0.562

### DISCUSSION

Method 1 proved ineffective because a number of the method's requirements were difficult to perform with halophilic bacteria. Many halophilic isolates did not readily form a pellet when centrifuged at speeds available on a tabletop centrifuge even with extended centrifuge times. This cell-harvesting problem resulted in low cell counts in resuspended cultures prior to UV exposure. Even though UV treated cultures were grown in HB broth, the lag phase before an increase in growth could be observed often took more than 3 hours. Method 2 seemed to alleviate many difficulties encountered with Method 1. Since cells were grown to higher densities and then back diluted prior to UV exposure, greater control could be maintained on determining the initial OD<sub>600</sub> reading. Increasing the incubation time to 25 hours and incubating cultures in the dark allowed for greater separation in growth curves between control and UV exposed cultures that were slow growing or had prolonged lag times. Method 3 provided growth curves most indicative of prophage induction indicating that mitomycin C was a more effective inducing agent than UV light.

Attempts to use UV radiation as the inducing agent yielded limited results with a number of halophile isolates showing little overall effect following a variety of UV exposure times. In a previous study with marine bacteria, researchers used a 15-watt light bulb to induce the bacteriophage (Williamson et al., 2002). Since the UV light source used in these induction studies was a 30-watt light bulb with a wavelength of 252 nm, it may have damaged the bacteria and could have been more than required for prophage induction. A significant decrease in absorbance was noted immediately after UV exposure for halophilic cultures, suggesting cell lysis, an observation not seen with non-halophile control bacteria (Table 1). Yet after several

hours of incubation, either in the light or in the dark, UV-exposed cultures appeared to be growing at the same rate as non-exposed control cultures. Our results suggest the UV light source used in these experiments was too strong and may have even induced cell lysis, as manifested by the initial decrease in absorbance. Wilcox and Fuhrman (1994) used continuous sunlight or pulses of sunlight to induce marine isolates, but no detectable virus production was seen using this method, indicating natural UV light did not induce lysogens. Using Method 2, which is more like natural UV exposure in the environment, we observed growth curves representative of prophage induction for several cultures. Cochrane, Kellogg, and Paul (1998) found that mitomycin C was the inducing agent of choice for marine lysogens even though it is not a natural component of the marine environment. Our results also indicate that mitomycin C is an effective inducing agent for halophilic bacteria because the most significant decreases in growth over time were observed with mitomycin C treated isolates.

### CONCLUSION

While it has been suggested that more than 40% of marine bacteria contain inducible prophage, under normal conditions it has been estimated that lysogenic viral production contributes less than 0.02% of the total viral load (Jiang & Paul, 1998). This suggests that prophage induction is rare under normal environmental conditions and only triggered under unusual circumstances. Since there is intense UV exposure during the summer when our isolates were obtained, this may be considered normal for these halophiles and few lysogens may exist in a bacterial population during this time period. Induction in the marine environment has been noted in months of lowest bacterial and algal production, suggesting lysogeny is favored under conditions for poor host growth (Williamson & Fuhrman, 2002). The majority of halophilic isolates in our collection were obtained in late spring and early summer, times of favorable growth in the GSL. Unpublished results in another study by our research group found high titers of phage for Salinivibrio in water samples during late spring and early summer collection times, suggesting elevated lytic cycle replication at the expense of lysogenization.

Unfortunately, Spot tests utilizing supernatant from induced cultures were not confirmatory for lytic bacteriophage. Interestingly, Calvo et al. (1988) never observed plaques after induction (only decreases in

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absorbance) while Jiang and Paul (1998) could only confirm prophage induction by SEM observation of filtrates. Only Mei et al. (2007) observed plaque formation after induction of *Natrinema* which they concluded were temperate phage. One explanation of our results is that the host cultures used in the Spot Test were predominantly lysogens, which already contained the prophage. Lysogens are resistant to superinfection by the same bacteriophage already integrated into the bacteria's genome.

Jiang and Paul (1996) state that environmental factors, which cause prophage induction in the marine environment, are unknown and it appears this may also be the case in halophilic environments. Our investigation of prophage induction for halophiles provided some promising results that warrant further investigation. Continued method development to induce prophage induction will include varying the concentration of mitomycin C and utilizing modifications in UV exposure intensity and UV wavelengths.

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## **COLLEGE OF SCIENCE**

**ZOOLOGY** 

# The Effect of Cheatgrass on Deer Mouse Abundance

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Phyllis Crosby Gardner Undergraduate Research Scholarship

### **ABSTRACT**

Invasive species can negatively affect the biodiversity of native ecosystems. Cheatgrass (Bromus tectorum) has invaded much of the Intermountain West and has been implicated in native biodiversity loss. This study addressed the effects cheatgrass has on deer mouse (Peromyscus maniculatus) populations at Antelope Island State Park, Utah. Deer mice were trapped during two four-night surveys along seven, 100-m traplines established in habitats representing a range of cheatgrass coverage from 19-77%. Linear regression showed a significant negative relationship between cheatgrass cover and the relative abundance of deer mice. These findings concur with the conclusions of others suggesting that cheatgrass negatively impacts the abundance of rodents and the composition of their communities.

### INTRODUCTION

Invasive species can have detrimental effects on ecosystems (Vitousek, 1990). They can negatively impact native biodiversity (Davis, 2003) and can contribute to species eradication (Pimentel et al., 2000). Cheatgrass (*Bromus tectorum*), an exotic annual grass native to Eurasia, has successfully invaded much of the Intermountain West of North America over the last century (Mack, 1981) and has been implicated in habitat loss and the subsequent decline of several animal species (Rickard, 1970; Gano and Rickard, 1982; Knick and Rotenberry, 2000; Newbold, 2005; Young and Clements, 2009).

Cheatgrass alters soil dynamics (Belnap and Phillips, 2001; Evans et al., 2001; Sperry et al., 2006) and increases fire frequency (Stewart and Hull, 1949; Young and Clements, 2009). Following overgrazing and wildfires, cheatgrass can become integrated as an understory member of

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the shrub-steppe landscape by out-competing native perennial grasses in root growth, seedling germination, and adult survivorship (Harris, 1967; Humphrey and Schupp, 2004), allowing it to displace native grasses and occupy spaces under and around shrubs (Billings, 1990). In many cases, it can become the dominant plant species in a community, perpetuating the cheatgrass-wildfire cycle (D'Antonio and Vitousek, 1992) and leading to increased cheatgrass dominance (Knapp, 1996).

Reduced native plant diversity, in turn, can affect abundance and diversity of animal species by reducing food resources (e.g., seeds, herbaceous forage, invertebrates) and homogenizing the overall structure of plant communities (e.g., shrub fragmentation/loss). Cheatgrass also indirectly affects foraging ability, thermoregulation, refuge sites, and movement patterns (Gano and Rickard, 1982; Parmenter and MacMahon, 1983; Stapp and Van Horne, 1997; Newbold, 2005).

The deer mouse (*Peromyscus maniculatus*) is a common and important species because of its role in food webs and seed dispersal (Handley Jr., 1999; Gitzen et al., 2001; Vander Wall et al., 2001). Yet, little work has been done to explore effects cheatgrass may impose on deer mouse populations as well as other animal species inhabiting the shrubsteppe habitat (Davidson et al., 1996; Ostoja, 2008, but see Rieder et al., 2009). Cheatgrass is widespread on Antelope Island and may be affecting deer mice populations, reducing an important prey species for mammalian mesopredators, raptors, and snakes. Furthermore, fewer rodents that distribute seeds could indirectly alter native plant communities (Ostoja, 2008).

In a preliminary study in which rodents were trapped using species-indiscriminate funnel traps, fewer deer mice were retrieved from traps in areas with higher percentages of cheatgrass cover (Hall et al., 2009). These preliminary observations were the basis for the main objective of the current study: to examine the relation between deer mice abundance and cheatgrass cover. I also examined other ecological aspects (e.g., breeding, demographics) of deer mice to provide a more complete assessment of the potential effects of cheatgrass on this species.

### MATERIALS AND METHODS

### Study Area

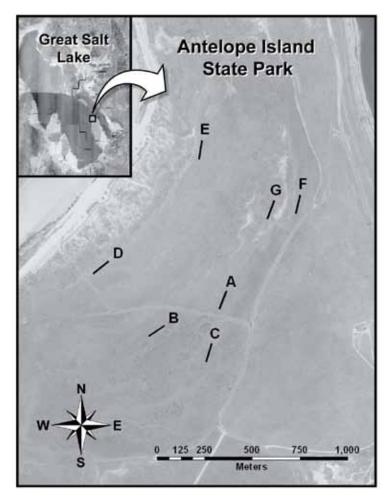
Antelope Island (41°02'N, 112°14'W, elevation 1,310 m) in Davis Co., Utah (Figure 1) is the largest island (113 km2) in the Great Salt Lake, although it is not a true island due to a causeway at the north end and to a naturally occurring land-bridge at the south end during dry years. Common native vegetation on Antelope Island consists of bunchgrasses—*Poa longiligula*, *P. spicatum*, *Agropyron spicatum*—and shrubs—*Artemisia tridentate*, *Chrysothamnus nauseosus*. Non-native cheatgrass is one of the most abundant plants on the island (Marshall, 1940; Hall et al., 2009).

Seven 100-m traplines were established with 10 stations each, one per 10 m of trapline. Their GPS coordinates were uploaded into a GIS (ArcMap, version 9.2, Environmental Systems Research Institute, Redlands, California) (Figure 1). At each station, two Sherman live traps  $(7.6 \times 8.9 \times 22.9 \text{ cm})$  were placed about 1 m on opposing sides of the trapline (Matlack et al., 2001), totaling 20 traps per trapline. Sherman traps were opened and baited every evening with rolled oats and peanut butter balls approximately 1 cm in diameter. Traps were checked and occupants were removed each morning prior to or at sunrise to avoid heat stress of trapped animals.

Trapline areas were selected to reflect a spectrum of cheatgrass cover present on the island by roughly estimating cheatgrass areas. However, this did not allow me to assess rodent abundances in non-cheatgrass areas, because this habitat type does not exist in the general habitat that I sampled. Due to the dominant nature of cheatgrass (Aguirre and Johnson, 1991) traplines with reduced cheatgrass cover had higher vegetation diversity (i.e., shrubs and grasses) whereas traplines with higher cheatgrass cover had lower vegetation diversity.

## Cheatgrass Surveys

Cheatgrass cover for each trapline was measured by placing a 1 m  $\times$  1 m wooden frame every 5 m along the trapline and estimating the percent of cheatgrass found within the square to the nearest 5% (Daubenmire, 1959). I took 20 individual cheatgrass samples per trapline, and 20% of each trapline was directly sampled during the cheatgrass survey.



**Figure 1.** Map of Great Salt Lake with enlarged picture of the northwestern portion of Antelope Island. Traplines are depicted with actual length and spatial arrangement, and are denoted by trapline letter.

## Rodent Surveys

Two four-night surveys were conducted from 23–26 May and 19–22 August 2006 during new moon periods. Surveys were scheduled to avoid full moon periods when nocturnal predators could potentially discourage rodent activity (Blair, 1943; Daly et al., 1992).

**Table 1.** Trapline metrics for deer mice (*Peromyscus maniculatus*).

Traplines	A	В	С	D	Е	F	G	OVERALL
Cheatgrass %	19	26	32	43	59	68	77	
SPRING								
Sex Ratio % (n)	42 (36)	6 (22)	23 (25)	34 (29)	40 (20)	37 (19)	22 (18)	29
% Breeding (n)	36 (36)	77 (22)	36 (25)	31 (29)	10 (20)	47 (19)	39 (18)	39
% Adults (n)	100 (36)	71 (31)	83 (30)	100 (29)	100 (20)	100 (19)	100 (18)	92
♂ Weight (g) (±SE)	25.9 (±1.4)	28.0 (±0)	24.6 (±1.6)	24.8 (±1.0)	23.6 (±1.0)	26.4 (±0.7)	27.3 (±2.6)	25.4 (±0.5)
n	15	2	7	10	8	7	4	53
♀ Weight (g) (±SE)	28.9 (±1.1)	27.4 (±0.9)	25.9 (±1.0)	26.0 (±0.8)	23.8 (±1.0)	28.4 (±1.0)	28.0 (±1.0)	27.0 (±0.4)
n	21	20	18	19	12	12	14	116
SUMMER								
Sex Ratio % (n)	58 (35)	56 (27)	58 (24)	44 (18)	50 (11)	44 (16)	41 (16)	52
% Breeding (n)	14 (35)	11 (27)	4 (24)	11 (18)	9 (11)	6 (16)	19 (16)	11
% Adults (n)	97 (36)	100 (27)	100 (24)	100 (18)	92 (12)	100 (16)	94 (17)	98
♂ Weight (g) (±SE)	23.6 (±0.6)	22.9 (±0.9)	22.6 (±0.6)	21.4 (±1.2)	22.2 (±0.6)	26.6 (±1.7)	22.3 (±0.6)	23.1 (±0.4)
n	20	15	14	8	5	7	7	76
♀ Weight (g) (±SE)	22.3 (±0.7)	22.8 (±1.0)	21.7 (±1.1)	24.6 (±1.4)	21.7 (±1.2)	24.7 (±1.0)	24.9 (±1.5)	23.2 (±0.4)
n	15	12	10	10	6	9	9	71

I recorded trap station, species identification, age, sex, breeding condition (e.g., males with scrotal testes, pregnancy or size of mammae in females), and weight to the nearest g using a  $100 \text{ g Pesola}^{\circ}$  spring scale ( $\pm 0.1 \text{ g}$ ) for each trapped rodent (Table 1). Each individual was marked by hair-clipping using a battery-powered beard trimmer (Matlack et al., 2001). Recaptured individuals were not included in the statistical analyses since the marking technique used was not individually unique. Rodents captured during the spring census were

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marked above the right shoulder and those captured in the summer census were marked above the right side of the rump. Once marked, rodents were released on site.

## Statistical Analyses

Mean percent cover (±SE) of cheatgrass was calculated for each trapline. The following metrics were calculated for May (spring) and August (summer) surveys for each trapline: relative abundance (number captured per four night trapline), percentage representing breeding adults, age distribution (% adults), mean weights for males and females (±SE), and sex ratios (% of adult males in the total sample of adult males and females). Overall percentages were also acquired for breeding individuals, adults, sex ratios, as well as overall means for male and female weights. Linear regression was used to determine if there was a relationship between rodent relative abundance and cheatgrass cover (%). Additional linear regressions were used to assess whether sex ratios, breeding percentages, age distributions, and mean weights were associated with cheatgrass cover (%).

A chi-square analysis was used to determine if sex ratio was equal between survey periods. Juvenile data of deer mice were removed from sex ratios, percent of breeding individuals, and mean weights so that these factors could be accurately compared. Statistical Package for the Social Sciences (SPSS) software, version 15.0, was used for all non-parametric analyses. All statistical tests were evaluated at the  $\alpha=0.05$  level.

### **RESULTS**

## Cheatgrass Cover

The results of the cheatgrass survey produced a range of cheatgrass percentages across the trapline sites (Figure 2).

## Species Captured

One-hundred and eighty-three deer mice (62 males, 121 females) were trapped during May and 150 (78 males, 72 females) were captured during August for a total of 333 deer mice in 1,120 trap-nights. Only one mortality was recorded. Eight montane voles (*Microtus montanus*; 2 males, 6 females) were captured in May, and three Ord's kangaroo rats

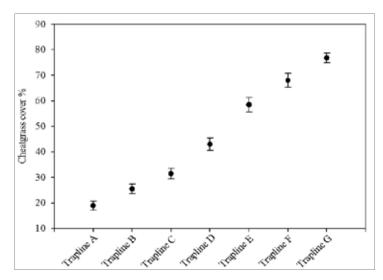


Figure 2. Mean ( $\pm$  SE) cheatgrass (*Bromus tectorum*) cover (%) of each trapline determined by a cheatgrass transect survey (n = 20 samples per trapline).

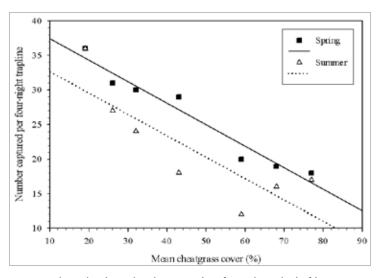


Figure 3. Relative abundances (number captured per four night trapline) of deer mice (Peromyscus maniculatus) trapped along cheatgrass (Bromus tectorum) percentage gradient on Antelope Island in May (spring) and August (summer) of 2006. The linear regression equations were Y = -0.31X + 40.5 (F1,6 = 97.2, P = 0.0002,  $R^2 = 0.95$ ; n = 183) and Y = -0.31X + 35.7 (F1,6 = 11.8, P = 0.019,  $R^2 = 0.70$ ; n = 150) respectively.

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(Dipodomys ordii; 1 male, 2 females) were captured in August. Because of rarity, these two species were excluded from analyses.

### Relative Abundance

Deer mouse relative abundance was negatively associated with cheatgrass cover (%) in spring and summer surveys (F1,5 = 97.2, P = 0.0002,  $R^2 = 0.95$ , n = 183; F1.5 = 11.8, P = 0.019,  $R^2 = 0.70$ , n = 150, respectively; Figure 3).

### Demographics & Breeding

In summer, males were less likely to be in high cheatgrass areas than females  $(F_{1.5} = 17.6, P = 0.0085, R^2 = 0.78)$ , but sexes were equally abundant in spring ( $F_{1.5} = 0.14, P = 0.72, R^2 = 0.027$ ). However, more females were present in spring than males ( $\chi^2 = 23.5$ , df = 1, P < 0.0001). The sex ratio was equal in summer ( $\chi^2 = 0.17$ , df = 1, P = 0.68).

The percentage of adult deer mice captured on each trapline was not significantly related to cheatgrass cover (%) during spring or summer  $(F_{15} = 1.97, P = 0.22, R^2 = 0.28; F_{15} = 1.36, P = 0.30, R^2 = 0.21,$ respectively). In addition, there was no relation between the percent of breeding deer mice and cheatgrass cover (%) during spring or summer ( $F_{1.5} = 0.51$ , P = 0.51,  $R^2 = 0.093$ ;  $F_{1.5} = 0.16$ , P = 0.70,  $R^2 = 0.032$ , respectively).

## Mean Weights

The mean weights of male and female deer mice for each trapline were not significantly associated with cheatgrass cover (%) during spring  $(F_{1.5} = 0.002, P = 0.97, R^2 = 0.0004; F_{1.5} = 0.022, P = 0.89, R^2 = 0.004)$ or summer  $(F_{1.5} = 0.19, P = 0.68, R^2 = 0.037; F_{1.5} = 2.80, P = 015,$  $R^2 = 0.36$ ).

### DISCUSSION

Several studies have shown that small mammals and particularly rodents are impacted by cheatgrass (Rogers and Hedlund, 1980; Gano and Rickard, 1982; Groves and Steenhof, 1988; Yensen et al., 1992; Gitzen et al., 2001; Hanser and Huntly, 2006; Ostoja, 2008). The impact may

be due to several factors. One is that areas invaded by cheatgrass are prone to wildfire (Young et al., 1987). Gano and Rickard (1982) found that rodents were less abundant in an area that had burned four years previous to their study, whereas McGee (1982) observed that rodents were only temporarily (i.e., 1-3 yrs) affected by fires. After a wildfire, if an area has sufficient time and potential to return to normal preburn status, then reestablishment of rodents is conceivable. But often, cheatgrass increases fire intervals (Knapp, 1996), decreasing recovery time between burns. Due to the self-perpetuating cycle of wildfire and cheatgrass succession, rodent populations may become depleted over time because the landscape would not have ample time to recover.

Reduced abundances of deer mice in high cheatgrass cover could be attributed to at least three factors: 1) cheatgrass may affect diet; 2) cheatgrass indirectly reduces shrubs that are important for foraging, cover from predation, and movement; and 3) cheatgrass may impede locomotive abilities critical for breeding, foraging, and predator avoidance.

First, landscape homogenization due to cheatgrass can result in less diverse and abundant food sources. This would most likely affect specialists more than generalists. This may partly explain why more deer mice, an omnivorous generalist (Wolff et al., 1985; Sieg et al., 1986), were captured in my study in cheatgrass-dominated habitats compared to other rodent species. Deer mice are considered "selective opportunists," primarily exhibiting granivorous and insectivorous habits in the West, with coleopterans and hymenopterans comprising the majority of their diet (Kritzman, 1974; Sieg et al., 1986). Cheatgrass alters the composition and abundance of beetle populations (Rickard and Haverfield, 1965; Rickard, 1970), an effect consistent with lower abundances of deer mice in areas of high cheatgrass cover in my study.

Since cheatgrass can attain high abundances and the deer mouse diet can vary seasonally and differ in composition (Vaughan, 1974; Wolff et al., 1985; Sieg et al., 1986), cheatgrass could have a large dietary importance for deer mice. But laboratory and field results show cheatgrass seeds are not preferred (Everett et al., 1978; Kelrick et al., 1986).

Second, loss and fragmentation of shrubs due to increased fire frequency and subsequent colonization by cheatgrass (Baker, 2006) may be affecting the small mammal community (Gitzen et al., 2001). 96 · Hall & Zeveloff Hall & Zeveloff • 97

Deer mice are more abundant in shrub communities (Rogers and Hedlund, 1980; Gano and Rickard, 1982; Stapp and Van Horne, 1997) using open microhabitats with less herbaceous cover (Belk et al., 1988) perhaps for ease of capturing insects (Pearson et al., 2001). Shrubs serve as cover permitting rodents to move about during times of high predator activity (Price et al., 1984). Yet Parmenter and MacMahon (1983) found that following shrub removal, deer mice still inhabited shrubless areas whereas other rodent species migrated elsewhere. They argued that shrubs may still be important for food sources and foraging ability over longer time periods, specifically since deer mice utilize shrubs for foraging.

Third, deer mouse mobility may be affected by cheatgrass (Rieder et al., 2009), which in turn may negatively affect the reproductive success of rodents in cheatgrass areas (Gano and Rickard, 1982; Groves and Steenhof, 1988). Lack of a relation between percentages of breeding mice and cheatgrass cover did not indicate that the proportions of breeding deer mice suffered because of encumbered mobility. Although I could not assess if reproductively active mice were successful at mating, dense cheatgrass may make it difficult for breeding mice to encounter one another. This might explain why relative abundance was less in cheatgrass areas. Reduced mobility could also affect foraging success for arthropods and the ability to escape predation.

### CONCLUSION

Much of the landscape that is susceptible to invasion by cheatgrass has likely already been affected to some degree. The extent to which cheatgrass has affected plant and animal species remains unclear. Animal species inhabiting areas invaded by cheatgrass are at risk (Billings, 1990) and unless cheatgrass can be effectively managed, it will not surrender its established territory in the semi-arid grasslands of the Intermountain West (Mack, 1989). This leaves the future of many shrub-steppe species uncertain, especially those that may be more sensitive to disturbance than the deer mouse.

### **ACKNOWLEDGEMENTS**

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## COLLEGE OF ARTS AND HUMANITIES PERFORMING ARTS

## MTNA Division Competition

#### Moriah Wilhelm

I am currently a Violin Performance Major at Weber State University and a student of Dr. Shi-Hwa Wang. On October 23, I had the opportunity to participate in the annual MTNA (Music Teachers National Association) National Competition where I was awarded first place in the Senior Strings Division at the state level. Because of this achievement, I have been asked to represent Utah, as well as Weber State University, at the University of Hawaii at Manoa – Honolulu, Hawaii on January 8-10, 2010.

MTNA is an organization founded to advance the study of music and strives to support the professionalism of music teachers as well as the performance of music to society. At this annual competition, I will have the fortuity of associating with professional musicians from throughout the southwestern portion of the United States and to contest for the prospect of qualifying for the national round held in Albuquerque, New Mexico convening later in March.

Participating in this competition will tremendously reinforce my personal performance abilities. Through the tutelage of Dr. Wang and the multiple performance venues that are available at Weber State, I have studied the skills needed to compete effectively and enjoy the aspirant atmosphere of this event. I will also strengthen my leadership and social skills in the field of music as I associate with great performers and pedagogues of the southwest.

The program that I have prepared for this competition is as follows: Concerto No. 5 by Henri Vieuxtemps (1820-1881), Tzigane by Maurice Ravel (1875-1937), and Johann Sebastian Bach's (1685-1750) Sonata No. 1. This encompasses a forty-minute program. The entire program needs to be performed memorized and it lasts 40 minutes.

Preparation for this event includes thorough rehearsal with my accompanist, Fan-Ya Lin to develop accurate technique, correct musical style and most importantly, effective musicality. We have both experimented with compelling ensemble efficacy and distinct

interpretation of genres ranging from baroque, classical, romantic, to impressionistic periods. My pianist and I have devoted many hours of rehearsal developing these principles and are continuing rigorous practice for this division competition.

This research was presented at the Music Teachers National Association Division Competition in Honolulu, Hawaii, January 7–11, 2010.

## JERRY AND VICKIE MOYES COLLEGE OF EDUCATION TEACHER EDUCATION

# The Effects of Indiscrimindable Contingencies on the Independent Work Completion and Accuracy of Students with Emotional and Behavioral Disorders

Kira Hafen-Westbroek & Natalie A. Williams, PhD.

This research describes the use of indiscriminable contingencies (IC) in a classroom of elementary students with behavioral disorders. The dependent variable was the accuracy and completion of independent seat work assignment, also known as self-start. The independent variable was an IC, determined by the number rolled on a pair of dice. This study attempted to answer the question: What are the effects of IC on the independent work completion and accuracy for students with behavioral disorders? The study implemented a single-subject reversal design using a visual analysis of the data to interpret the results and potential success of the IC as a successful classroom intervention.

This research was presented at the ABAI 35th Annual Convention in Phoenix, Arizona, May 22–26, 2009.

## JERRY AND VICKIE MOYES COLLEGE OF EDUCATION TEACHER EDUCATION

## Does Caring Matter in Education

Loretta Walker, Rick Walker & Susan Kenney

Anecdotes of university and elementary music students who do or do not thrive in educational settings. A discussion of research literature from the converging fields of education, psychology, and brain research is cited, exploring how the nature of critical interpersonal relationships affects students' ability to succeed in a several key aspects of education, broadly defined, such as learning and memory, trust and healthy risktaking required for growth, the cultivation of supportive relationships that nurture learning, and cultivation of the disposition to apply what one has learned for the betterment of one's own life and the lives of others. The role of metacognition in executive function is discussed in some detail. These issues are examined through the lenses of trauma studies (clinical and neuroscientific), attachment theory, emotional intelligence, positive psychology, and educational philosophy. Implications for educational practice are discussed, particularly in the interrelated arenas of public education, teacher education, and the ongoing growth of teacher educators.

This research was presented at the Mountain Lake Colloquium in Mountain Lake, Virginia, May 17–20, 2009.

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## DR. EZEKIEL R. DUMKE COLLEGE OF HEALTH PROFESSIONS CLINICAL LABORATORY SCIENCES

# Prolonging Reconstituted Coagulation Control Stability with the Addition of Trehalose

Chelsie Buckner, Natalie Heath, Mechelle Sargent, Satoko Yamamoto & Kara Hansen-Suchy

The objectives of this research were to prolong the expiration of reconstituted coagulation controls used by medical laboratories by adding a unique sugar molecule called trehalose and also to determine if the shelf life of lyophilized controls could be extended. Trehalose was chosen because it has previously been shown to protect proteins from denaturation thus making it appropriate to use with proteinbased coagulation controls. Prothrombin time (PT) and activated partial thromboplastin time (aPTT) tests were performed on various levels of coagulation controls at different time intervals over a period of 72 hours. Additionally, controls with a stated expiration date from more than three years ago were also analyzed. This study illustrated that trehalose provided no benefit in prolonging the stability of reconstituted controls. It is important to note that regardless of the addition of trehalose, all controls stayed within +/-2 standard deviations for at least 72 hours, surpassing the manufacturers' recommendation of only eight hours. Furthermore, the expired controls still yielded valid results suggesting the stability of the lyophilized controls could be years beyond the stated expiration date. These findings could lead to less waste of coagulation controls and significant monetary savings for medical laboratories.

This research was presented at the American Society for Clinical Laboratory Science National Conference in Chicago, Illinois, July 21–24, 2009.

## DR. EZEKIEL R. DUMKE COLLEGE OF HEALTH PROFESSIONS DENTAL HYGIENE

# Are You at Risk? An Investigation Into the Use and Communication of Risk Assessments to Patients in Dentistry

Elisabeth Pyper, Katie Poulsen, Amanda Ferguson & Kami Hanson

The dental profession works to broaden public understanding of the importance of oral health and its relevance to general health. Oral health is evaluated using data obtained from conducting clinical assessments for the risk of certain dental diseases. A summary of dental and overall health risk based on clinical assessments is referred to as a "risk assessment calculator" or RAC. Risk assessment calculators are important because they are a uniform, consistent way to measure the various overall risks that dental patients face. They also allow patients to understand the clinician's explanation of risks and how they relate to their body, life and future. The purpose of this research was to find out how risk assessments are being performed and communicated in private dental offices. It was hypothesized that the majority of dental clinicians do not use a type of risk assessment tool and do not communicate risk to their patients. Also, when risk assessments are performed, there is a general lack of patient education and understanding. Methodology included a survey of dental professionals and dental patients. Research data was evaluated for descriptive statistics. Evidence revealed that 83% of dental professionals did not use a type of risk assessment tool and that 53% of the time risk assessment was communicated to the patient verbally. Most dental professionals were satisfied with the current method they are using for risk assessment and that they are educated about current options available for this purpose. Dental patients stated a high awareness, 69%, of their risk for dental disease and overall health and that 63% found this information helpful. Patients further communicated that they did not care how risk was communicated to them, but 63% of them are not willing to pay extra for the service. In conclusion, dental professionals conduct risk assessments routinely without the benefit of a RAC tool. Dental risk is communicated to patients verbally and not related to overall health. Dental patients state an understanding of information communicated to them and are

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not willing to pay for a more extensive RAC that correlates oral health to overall health. A general taxonomy needs to be set for the dental community on risk assessment and awareness for new RAC that can synthesize oral and health information for patients.

This research was presented at the American Dental Hygienists Association's Annual Session in Washington, D.C., June 18–23, 2009.

### Wright, et al. · III

## DR. EZEKIEL R. DUMKE COLLEGE OF HEALTH PROFESSIONS DENTAL HYGIENE

## A Collaborative Multi-year Project in the Creation of a Mixed-Reality System to Teach Techniques for Local Anesthesia

Michelle Wright, Megan Dahl, Amber Allen and Holly Burton & Kami Hanson

Starting in 2005, dental hygiene students and faculty at Weber State University have been working on a project that would allow students to learn techniques for administering local anesthetic in a virtual environment. The virtual system that we have today called LAMRS (Local Anesthesia Mixed-Reality System) has gone through multiple iterations in design and research. Our presentation will be on the phases of development for LAMRS from 2005 to present and on the most current research outcomes for learning using LAMRS. Research is set to begin April 6, 2009 with the latest iteration of LAMRS. Outcomes will be ready to present by the June 2009 ADHA annual session. Research questions that will be addressed are centered on: Can students learn techniques for administering local anesthetic using LAMRS? Can students demonstrate a level of competency and verbalize confidence after using LAMRS? Will students gain a greater sense of anatomical spatial and dimensional acuity of human anatomy?

This research was presented at the American Dental Hygienists Association's Annual Session in Washington, D.C., June 18–23, 2009.

112 · Hall, et al.

McCormick, et al. · 113

## DR. EZEKIEL R. DUMKE COLLEGE OF HEALTH PROFESSIONS DENTAL HYGIENE

## Do I Need a Second Life? An Investigation into Student Use of Second Life for Dental Hygiene

Kara Hall, Denver Allen and Pamela Carranza & Kami Hanson

Second Life (SL) is an online 3-dimensional virtual world. About 3 years ago it had approximately 100,000 members, now it registers about 100,000 new members each month. Of those members, large numbers are educators and K-20 institutions that embrace SL as an instructional and social tool. As students in dental hygiene, we want to explore SL with our faculty for its potential use in our education. Our first efforts will focus on gaining basic literacy within the environment and then to investigate those best practices for education in use by other institutions. Second, we have several activities planned in SL with other students and educational groups. Our presentation will focus on presenting those outcomes. Our research is currently ongoing but will be completed and ready to present in June 2009 for the ADHA annual session. Our expectation is to share our knowledge and experience with other dental hygiene students and educators so that SL can be used for teaching and potential collaborations in the future. We have found that virtual worlds like SL are not games, but complex-evolving models that are significant for multi-disciplinary applications.

This research was presented at the American Dental Hygienists Association's Annual Session in Washington, D.C., June 18–23, 2009.

## COLLEGE OF SCIENCE BOTANY

## Germination of Cypripedium Parviflorum (Orchidaceae) Seeds Using Crude Soil Inoculum

Meghan A. McCormick & Dr. Ron Deckert

Cypripedium parviflorum is a species of temperate terrestrial orchid. The entire genus Cypripedium is listed on Appendix II of the Convention on International Trade in Endangered Species (CITES). C. parviflorum has a widespread distribution in the United States, but is listed by the US Forest Service (USFS) Rocky Mountain Region as a sensitive plant species that occurs rarely in the far western and southwestern US. C. parviflorum seeds are lacking in nutrient reserves and must form an association with specific soil fungi to germinate. The effect of soil from 10 different habitat types on C. parviflorum germination rates was studied. The hypothesis that germination rates of C. parviflorum will vary when inoculated with soil from different sites, due to the presence or absence of particular mycorrhizal fungal symbiont species, was tested. Sites ranged in quality from known orchid sites (based on herbarium specimens) to orchid hostile habitats, and it was predicted that higher rates of germination would occur in soils from orchid sites. Seeds were exposed to crude soil inoculum using one of three germination techniques: Petri plates containing cellulose agar, Petri plates containing a simplified soil microcosm, and ex situ seed baits in pots with soil collected from test sites. Asymbiotic germination of seeds on nutrient agar was used as a base line comparison for germination. Preliminary results from the simplified soil microcosm plates showed high seed vitality rates with treatment of soil from known orchid sites, suggesting that germination rates will be correspondingly higher in those sites than in other sites. Soils from sites that correlate with high seed vitality may indicate potential C. parviflorum habitats.

This research was presented at the 23rd National Conference on Undergraduate Research in La Crosse, Wisconsin, April 16–18, 2009. It was also presented at the 2009 Botany and Mycology Conference in Snowbird, Utah, July 25–29, 2009.

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## COLLEGE OF SCIENCE CHEMISTRY

# Analysis of Titanium Oxide in Liquid Paint by X-ray Fluorescence Spectroscopy

Rachelle E. Maass, Hyun Ah Choi & Edward B. Walker

Pigments are incorporated into paint to enhance color, opacity, and durability. Titanium oxide accounts for about one-third of all pigments used in paints, often formulated into liquid paint emulsions at relatively high concentrations. An important aspect of quality testing is the determination of titanium oxide in liquid paint emulsions. Traditional methods of analysis require extensive sample preparation, such as ashing and subsequent dissolution, prior to atomic absorption or emission spectroscopy. In an effort to provide a more rapid and simplified test procedure, X-ray fluorescence was successfully applied to whole liquid paint samples. A method was developed using a mobile, hand-held x-ray fluorescence instrument that accurately determined total titanium levels in just 30 seconds. The method exhibits linear correlation across a range of 0-10%(w/w) TiO2. Higher concentrations deviated somewhat from linearity, but were readily measured based upon a second-degree polynomial calibration curve.

This research was presented at the American Chemical Society 237th National Meeting in Salt Lake City, Utah, March 22–26, 2009.

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## **COLLEGE OF SCIENCE**

**CHEMISTRY** 

## Rapid Analysis of Tin in Zirconium Alloys by X-ray Fluorescence

J. Spencer Barrett, Rachelle E. Maass, Brandon J. Burnett, Kyle D. Ashby, & Edward B. Walker

Zirconium is an important metal used in the nuclear fuels industry. Adherence to rigid specifications for Zr alloys, called Zircaloy, is critical to ensure its performance and efficiency in the extreme environment of nuclear reactor cores. Tin is present in Zircaloy at low concentrations, typically 1.5% (w/w). The most common method for determination of Sn is ICP spectroscopy. We report the application of XRF for analysis of Sn in both solid metal samples and aqueous solutions of Zircaloy. By virtue of the different emission energies of Zr and Sn, this analytical method does not suffer from commonly encountered difficulties caused by high Zr levels during ICP analysis.

This research was presented at the American Chemical Society 237th National Meeting in Salt Lake City, Utah, March 22–26, 2009.

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## COLLEGE OF SCIENCE

**CHEMISTRY** 

## Rapid Determination of Gold During Plating Operations by X-ray Fluorescence

Andrew B. Giles, Brandon J. Burnett, Arthur L. Anderson, Terry Darger, Kyle D. Ashby & Edward B. Walker

The process of electro-plating gold onto conductive surfaces electrochemically deposits a very thin layer of metallic gold over the surface of the item to be plated. This process can impart some of the desired properties of the gold to the entire piece being plated at a fraction of the cost of solid gold. In many cases, the important mechanical properties of the substrate can be maintained to obtain the desirable benefit of an exposed gold surface. The concentrations of gold and various additives in cobalt-hardened plating solutions affect the cathode efficiency. The purpose of this study is to determine the influence of gold concentration on the cathode efficiency of the plating cell in the presence of typical additives. A mobile hand-held X-ray Fluorescence (XRF) instrument was utilized to successfully analyze gold in plating solutions at concentration ranges of 0-12 g/L in the presence of several additive ions including Co, Ni, K, Mn, and Cr. XRF was also utilized to determine the thickness of gold layers plated on copper substrate up to 10 microns. The cathodic efficiency of the plating process was then studied using this method, revealing that an optimum gold concentration of 7.4 g/L under our experimental conditions.

This research was presented at the American Chemical Society 237th National Meeting in Salt Lake City, Utah, March 22–26, 2009.

## COLLEGE OF SCIENCE GEOSCIENCES

## Paleontology and Sedimentology of a Turonian (Late Cretaceous) Lagoon, Southwestern Utah

Shayne Pearce & Jeff Eaton

A preliminary study of fossils recovered from UMNH (Utah Museum of Natural History) locality IP24 in Bryce Canyon National Park in the Smoky Hollow Member of the Straight Cliffs Formation (middle Turonian, Late Cretaceous) revealed previously unreported vertebrate and invertebrate taxa. The most fossiliferous horizon is in a carbonaceous siltstone immediately above one of the lignite layers about 15.5 meters above base of the member. Fossils were recovered by wet screen-washing, a technique traditionally not applied to invertebrate localities. Many of these taxa (including numerous brackish water gastropods and bivalves including oysters, and several possibly freshwater taxa including ostracods, pycnodontid fish, crocodilians, myledaphid rays, shark, amiids, and gars) have not been previously reported from brackish water localities in the Smoky Hollow Member. The recovered taxa suggest an intermingling of fresh and brackish water taxa as would be expected in a lagoonal environment. Stratigraphic sections from multiple sites were measured and depositional features analyzed. The Smoky Hollow Member overlies the regressive marginal marine sandstones of the Tibbet Canyon Member. The stratigraphic sequence in the lower part of the Smoky Hollow Member represents a continuation of that regressive sequence and is composed mostly of fine-grained sandstones and a highly variable sequence of organic rich siltstone-lignite couplets ~12.5 to 32 m above the base of the member, each about one meter thick. These couplets probably represent autocyclic or possibly Milankovitch driven cycles. As interpreted here, the sandstones represent the barrier beach environments, the organic rich siltstones the lagoonal environment, and the lignites the adjacent swamps. These facies are highly variable both laterally and vertically, suggesting local controls such as channel and bar migration. The absence of marine or brackish water fossils below the lignite and the lack of Sedimentological structures suggest reworking of sediment by

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wave-action. The lignite and organic rich siltstone layers along with taxonomic associations of oyster beds, other brackish water taxa as well as fresh water taxa suggest a coastal lagoon setting with adjacent barrier beaches and swamps.

This research was presented at the 61st Annual Meeting Rocky Mountain Section at Utah Valley University in Orem, Utah, May 11–13, 2009.

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## COLLEGE OF SCIENCE GEOSCIENCES

## Weathering Patterns and Dissolution Rates of Calcite Buried in Arctic Soils on Spitsbergen – AFM and SEM Study

Sara Summers, Marek Matyjasik, Maciej Manecki & Colin Inglefield

This study presents the results of Atomic Force Microscopy (AFM) and Scanning Electron Microscopy (SEM) imaging of calcite sample surfaces after they have been buried for one year and three years respectively, in the arctic soil of Spitsbergen. The objective of the study was to determine the pattern of calcite surface alterations and to compare the effects of various environments. Six samples of freshly cleaved calcite were buried in three different environments. Environmental effects include varying elevations (ranging from 1 to 590 m amsl), distances from the Greenland Sea (from 10 m to 3000 m), and distances from the Werenskiold Glacier (from 100 to 3500 m). The retrieved samples were analyzed using AFM and SEM and compared with a control sample that was freshly cleaved and had never been exposed to an arctic environment. The control sample is characterized by sharp step edges and smooth surfaces. Overall, all of the calcite samples recovered from Spitsbergen have more irregular surfaces with rounded edges. The development of numerous intersecting etch pits is common, both isometric and elongated. Distribution of etch pits is heterogeneous with some areas being less affected. Typically top step surfaces are altered more than bottom step surfaces. Rhombohedral weathering patterns are present on many surfaces and are more dominant in samples retrieved after three years. Calcite surfaces are covered locally by biofilms. The most prominent dissolution features (advanced weathering) were observed in the sample located at the Greenland Sea shore; characterized by the longest contact time with water, higher moisture air masses, and soils with more active microbial activity. The dissolution rates varied between 1.25E-08 mol/cm2 / yr to 5.97E-07 mol/cm2/yr. Annual dissolution rates were higher in all samples retrieved after 3 years than in samples retrieved 1 year. The average annual dissolution rate increased by 202% for samples retrieved after 3 years as compared to samples retrieved after 1 year. This dissolution rate increase can result from a larger calcite surfaces areas

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exposed to chemical reactions in all samples but there might also be additional regional factor (such as regional weather or climate pattern) that has influenced the dissolution rate for all samples.

This research was presented at the GSA Annual Meeting 2009 in Portland, Oregon, October 18–21, 2009.

Canova, et al. · 121

## COLLEGE OF SCIENCE MICROBIOLOGY

## Development of an Assay to Characterize Chitinase Utilization in Euryhaline Halophiles

Travis J. Canova, Craig J. Oberg & Michele D. Zwolinski

Chitin is composed of repeating units of the monomer N-acetyl-D-glucosamine (GlcNAc). It serves as a significant carbon and nitrogen source in the Great Salt Lake (GSL) due to the large biomass of brine fly cases and brine shrimp exoskeletons. Degradation of chitin by euryhaline halophilic bacteria is important in recycling carbon and nitrogen in the GSL, but the diversity of chitinolytic organisms remains unknown. This assay, adapted from a filter paper technique, can screen up to 96 samples simultaneously for chitinolytic activity. Assay substrates are the monomer, dimer, and trimer forms of 4-methyllumbelliferyl-N-acetyl-β-D-glucosaminide (4-MUF. GlcNAc). When these compounds are enzymatically cleaved, 4-methyllumbelliferyl (4-MUF) is released and fluoresces. This assay was used to screen halophilic microorganisms isolated from the GSL. The cleavage pattern for the three substrates helps characterize the chitinolytic enzymes each isolate may possess. Results indicate some halophile isolates metabolize all three forms of the GlcNAc derivatives while others prefer just the dimer and trimer forms. This assay can now provide data on the metabolic rates of carbon and nitrogen biogeochemical cycling in the GSL ecosystem. In addition, it can be used as an initial screen for organisms that may have interesting metabolic traits associated with chitinase production for use in biotechnology applications.

This research was presented at the 109th General Meeting of the American Society for Microbiology in Philadelphia, Pennsylvania, May 17–21, 2009.

## COLLEGE OF SCIENCE PHYSICS

# High Albedo Events Indicate Water Ice in Mars' Southern Polar Craters

Charla Boom & John Armstrong

Continuing the work done previously in the northern polar craters examining crater-interior water ice deposits, a sample of craters in the southern polar region of Mars has been analyzed using Mars Global Surveyor Thermal Emission Spectra (TES) temperature and albedo measurements. Using the JMARS program, we specifically analyze the depth, latitude, size, and elevation of these craters, hoping to gain better insight into the distribution and causes of these high albedo events. Variations in albedo distinguish 81 craters with high albedo events (HAEs) starting at Ls between 200 and 230 degrees. Data for morning albedo is less abundant than afternoon data, but appears consistent, supporting 52 of the 81 craters. The remaining craters did not have enough data taken in the morning to support or contradict the afternoon evidence. Data was taken external to each crater (to verify it is not an artifact of the entire pole), and HAEs were present in twelve of the 277 area observed. Eleven of these areas were found in a region that appears to be caused from variations in polar layered deposits from the seasonal ice cap. A  $\chi^2$  analysis was conducted to determine statistical significances. There is no difference between the depth of craters with or without HAEs, nor is there a difference in the diameters. The HAE craters are higher in elevation by 219 m. This could be an artifact of the distribution of the craters. HAE craters tend to be closer to the pole, which has higher elevation. As we compare the results and distributions to the northern polar region, we find the southern polar HAE craters appear to cluster toward the pole, where the northern HAE craters appear evenly distributed. HAE craters in the southern region did not extend below where the seasonal cap freezes, at ~65°S.

This research was presented at the Undergraduate Women in Physics Conference in Lincoln, Nebraska, October 31 – November 1, 2009.

## COLLEGE OF SCIENCE ZOOLOGY

# DNA Barcoding Reveals the Diversity of Shore Flies from the Great Salt Lake

Krystle J. Minear & Jonathan B. Clark

The Great Salt Lake (GSL) in northern Utah is one of the largest lakes in the United States, with a total surface area of 4400 square kilometers. Shore flies (Ephydridae) are among the most important components of the GSL ecosystem, removing an estimated 90 million kg of organic matter from the lake and serving as an important food source for bird populations. In spite of their essential role in this ecosystem there are few published studies on shore fly identity and diversity. DNA barcoding is a technique that examines a relatively short portion of the mitochondrial gene, cytochrome oxidase I (COI). This short sequence works as a barcode in that can be cloned and sequenced relatively cheaply; yet shows enough sequence variability to be useful in distinguishing closely related species. Among the other DNA sequences that can be used to differentiate species is a nuclear noncoding region known as ITS-1. For this sequence, the variability extends to size differences that can be revealed using gel electrophoresis. DNA barcoding is particularly useful for studying shore flies because some species are difficult to distinguish morphologically. This study examines how morphology and molecules can be used to compliment each other in the identification of shore flies. In addition, a comparison is made between COI and ITS-1 as species identifiers. The ultimate goal is to characterize all of the species of flies associated with the Great Salt Lake ecosystem.

This research was presented at the Sigma Xi Annual meeting and Research Conference at the Woodlands Waterway Marriot Hotel in Houston, Texas, November 12–15, 2009.

## COLLEGE OF SCIENCE ZOOLOGY

## Ecomorphological Relations between Cottus Species and their Environment in Northeastern Utah

Nathan V. Holmes & Christopher W. Hoagstrom

Factors inducing phenotypic plasticity of fishes are poorly understood. A recent study revealed a relation between sculpin (Cottus) morphology and stream bottom velocity. We investigated this relation in 16 streams of northeastern Utah, but included 16 additional environmental variables. We took 21 morphometric and two meristic measurements on all sculpin over 71 mm standard length, measuring a total of 373 individuals from 12 streams and 36 mesohabitats. Sculpin morphology varied substantially. One principle component (PC) revealed disparity in robustness, mouth size, and number of prickles among individuals and between species. Another PC revealed disparity in fin length, fin height, and head width among individuals, but not between species. Environmental variables significantly explained morphological variation suggesting phenotypic plasticity. Variation associated with the first and second PC was explained by many biotic and abiotic factors. Overall, our results support the earlier study because stream bottom velocity influenced morphology. However, inclusion of additional variables substantially improved the explanatory model.

This research was presented at the American Fishes Society – Western Division Annual Meeting in Albuquerque, New Mexico, May 2–8, 2009.

## COLLEGE OF SCIENCE ZOOLOGY

## Metagenomic Analysis of Brine Fly Larvae from Great Salt Lake, an Extreme Environment

Christy R. Cottrell, Mohammed Sondossi & Jonathan B. Clark

Brine flies of the family Ephydridae are particularly adaptable and inhabit extreme environments throughout their worldwide distribution. Brine flies constitute an important component of northern Utah's Great Salt Lake ecosystem, removing organic matter and serving as a food source for millions of birds. In spite of their biological and economic importance, little is known regarding the adaptation of brine flies to the extreme conditions associated with Great Salt Lake. It is possible that bacteria play a role in allowing the aquatic brine fly larvae to thrive in this high saline environment. Because it may not be possible to culture these bacteria free of their larval hosts, a culture-independent method, metagenomics, was used to assess the microbial diversity associated with brine fly larvae. Metagenomics involves the study of DNA isolated directly from environmental samples. In this study, primers specific for the 16S ribosomal RNA (rRNA) gene were used to amplify bacterial DNA isolated from larval samples. 16S rRNA gene sequences were compared to the genetic database and phylogenetic analysis was used to determine the affiliations of the larval samples. A comparison can also be made between the identity of culturable and non-culturable bacteria. Ultimately, information gained from this study will help determine whether these microbes contribute to the brine fly's ability to thrive in this extreme environment.

This research was presented at the 2009 Sigma Xi Annual Meeting & International Research Conference in Houston, Texas; November 12–15, 2009

## COLLEGE OF SCIENCE ZOOLOGY

## Morphological Variation among Humpback Whitefish in Three Alaskan Creeks

Michael Cranney & Christopher W. Hoagstrom

Morphology is a major component of an organism's phenotype. It affects important traits such as locomotion, feeding, and reproduction. The success of an organism is directly linked to its morphology. Between 22 June and 3 July 2008 we sampled humpback whitefish, Coregonus pidschian, from Scottie, Desper, and Gardiner creeks on Tetlin Nation Wildlife Refuge, Alaska to quantify morphological variation within and among populations. We captured whitefish with monofilament gill nets placed in major pool areas. To get geometricmorphometric data, we captured a left-lateral image of each fish and digitized 12 landmarks on each image using the program software tpsDig. Geometric-morphometric analysis was done with tpsRelw. Overall, morphology varied in body depth, hump prominence, and fin positioning. All morphotypes were found in Scottie Creek (n = 33), whereas only relatively shallow-bodied morphotypes were found in Desper (n = 12) and Gardiner (n = 2) creeks. Lower morphological diversity in Desper and Gardiner creeks may be due to lower sample size or morphology-related niche partitioning. Niche partitioning is supported by substantial differences among creeks. For example, deeper-bodied morphotypes in Scottie Creek may use more extensive pelagic habitat present in large pools. Such habitat was rare or absent in Desper and Gardiner creeks.

This research was presented at the American Fishes Society – Western Division Annual Meeting in Albuquerque, New Mexico, May 2–8, 2009.

## COLLEGE OF SOCIAL & BEHAVIORAL SCIENCES CRIMINAL JUSTICE

# The Influences of Fatigue and Suggestibility on Child Witnesses

Patrick Q. Brady, Erin F. Swedish & Julie A. Buck

When children are required to testify in court or recall information about an experienced event for forensic purposes, it is extremely important that the most accurate information is obtained from the child witness. Individuals of all ages are somewhat partial to suggestion and it has been shown that pre-school aged children are more susceptible to suggestive questioning than adults (Ceci & Bruck, 1993). In order to better forensic interview qualities, past research has shown how different suggestive interview techniques have led to inaccurate information obtained from the child witness (Garven et al., 1998). In this study we examined whether acute fatigue (as measured by time of day) affects preschool age children's susceptibility to suggestive questioning and their memory accuracy. Children (N=80) participated in a staged magic show and were interviewed one week later about specific details of the magic show. The children were randomly assigned to be interviewed at three different times of day to assess what time of day children provided the most accurate memory recall. The interview was broken up in to five stages that involved different acts throughout the show. We implemented nine true reminders of actual events that happened in the show, seven false reminders, and eight open-ended free recall questions. Results showed that children were least accurate in the afternoon and most susceptible to suggestive questioning in the afternoon. The results of this study can be applied towards forensic interview practices, by interviewing children at the appropriate time of day to obtain more accurate information from child witnesses.

This research was presented at the Western Association of Criminal Justice Conference in Las Vegas, Nevada, October 13–16, 2009.

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## COLLEGE OF SOCIAL & BEHAVIORAL SCIENCES PSYCHOLOGY

## Framing on Prejudice and Predictability of Personality Traits on Openness to Diversity

### Nathan Taylor

There has been an increased interest in investigating the differential impact of multicultural and assimilation strategies (Richeson & Nussbaum, 2004; Wolsko, Park, Judd, & Wittenbrink, 2000). Research has found that presenting multicultural ideology in a positive way reduces both explicit and implicit prejudice but increases stereotyping. The current study wished to replicate Richeson & Nussebaum's study investigating whether priming participants with multicultural or assimilation ideology would similarly affect implicit and explicit prejudice. In addition, the current study wanted to assess the predictive ability of certain personality traits (such as open mindedness, cultural empathy, emotional stability, flexibility, and social initiative) that have been demonstrated to predict attitudes about openness to diversity of prejudice, desire to learn and to accept of other cultures, and symbolic threat experienced towards immigrants.

The current study manipulated the ideological information presented to participants. Participants were presented with either a vignette that described the current situation in the U.S. as being culturally diverse OR culturally assimilated. Next, participants completed an implicit measure of prejudice followed by an explicit measure of prejudice, and symbolic threat. Finally, participants completed the Multicultural Personality Questionnaire (van Oudenhoven & van der Zee, 2002). Results demonstrated that contrary to previous findings and predictions (i.e., Richeson & Nussbaum, 2004), the manipulations did not have a significant effect on either explicit or implicit measures of prejudice. Nor did the manipulations have a significant effect on symbolic threat. Regression analysis did demonstrate that the personality trait of openmindedness significantly predicted desire to learn about and accept other cultures whereby greater openmindedness predicted greater desire to learn about other cultures. In addition, it was found that the personality trait of social initiative significantly predicted symbolic

threat, whereby greater social initiative predicted greater symbolic threat. Future research and implications of findings will be discussed.

This research was presented at the South Western Psychology Association in San Antonio, Texas, April 1–4, 2009.