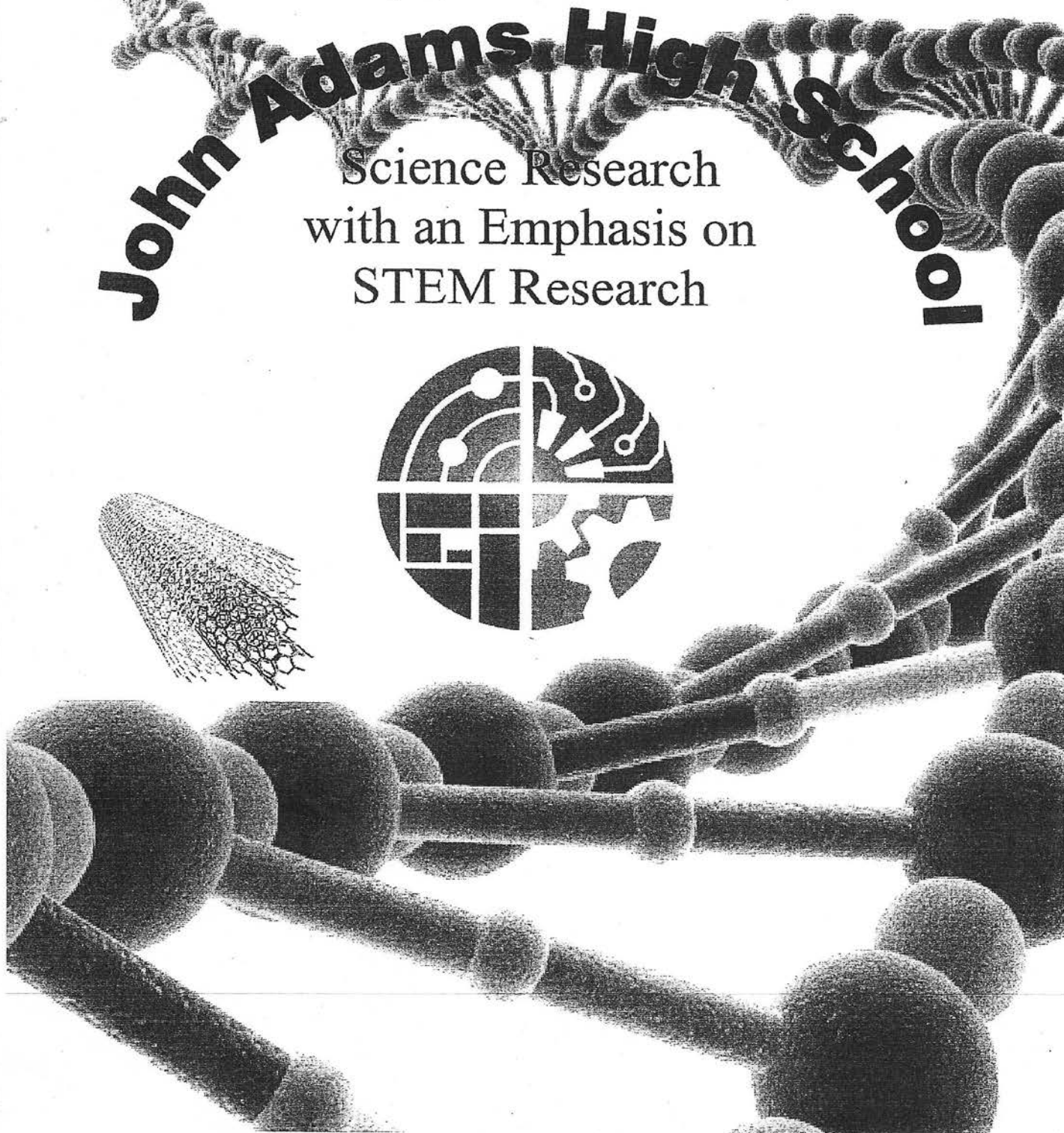
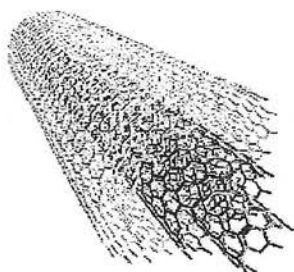




John Adams High School

Science Research
with an Emphasis on
STEM Research



Editor

Nevin Longenecker

Graphics Editor

Collin Daniels

An Introduction by the Author

The Science Research Course at John Adams High School began several years ago following repeated requests from many students over the years for an additional year of investigative science. The research course was designed to allow students to become familiar with reading research journals, writing research proposal and performing investigations. The course was not designed to provide advanced placement credit in college science courses, although, many of these students have received advanced placement at different universities.

Locally the class members have received much publicity due to the sophisticated nature of their work. Their research proposals have been funded by repeated grants from the American Lung Association, American Heart Association, Dow Chemical Company, Indiana Academy of Science and the American Association for the Advancement of Science. Further, many of these students have been awarded scholarships, trips and monetary awards totaling more than two million dollars.

Over the past several years many requests have come to the science department to provide an outline of the course syllabus. Although these letters have been handled individually, an inordinate of time has been involved with the replies and the responses have not always been identical or complete in scope. Dr. Jay Brockman, Associate Dean of Engineering at Notre Dame University, encouraged me to compile a booklet which could answer many of the routine questions about the course.

The course is offered as a honors science elective to both juniors and seniors who have completed two years of science course work, usually one year of biology and one year of chemistry, maintaining an A or B average in these courses. Further, for seniors, the research course cannot be taken in lieu of physics, but can be taken concurrently with physics. In addition, teachers of the previous two years concur about the students selecting the research course. Not all students who perform well in a structured class setting do well in this course when given the necessary freedom to perform independent research work.

Additional information about this course and the recognition gained by these students is listed on the internet at www.sbcsc.k12.us/adams/default1.html

Nevin E. Longenecker
John Adams High School
January 2017

Table of Contents

| | |
|---|--------|
| An Introduction by the Author | i |
| Table of Contents | ii-iii |
| Support letters from science researchers. | |
| Dr. Molly Duman Scheel-cancer researcher at IU Medical Center | iv |
| Dr. Fil Randazzo-deputy director of Bill and Melinda Gates Foundation | v |
| Dr. John Rearick- Attorney for protection of intellectual property-Boston | vi |
| Forward to the Interested Teacher | vii |
| Research rewards at John Adams High School | viii |
| Science Research tentative schedule (2 pages) | ix-x |
| Brief Reports of Science Research [Ch.1] | 1 |
| Science News (Society For Science & The Public) | |
| Understanding Research Papers[Ch. 2] | 2 |
| Precision in Measurement[Ch.4] | 3-4 |
| Electronic balance(readability to 0.001g) recommend 2 | |
| Triple Beam or electronic balance (readability to 300g) recommend 2 | |
| The Centrifuge[Ch.7] | 5-7 |
| Variable speed for test tubes (suggest 2) | |
| Ultra highspeed centrifuge for small test tubes (expensive-recommend 1) | |
| Histological Techniques of Staining (General) [Ch.8] | 8 |
| Compound microscopes with 400X –or 1000X | |
| Stereoscopes up to 30X | |
| Histological Techniques of Staining (Plant Tissue) [Ch.8] | 9-10 |
| Carolina Redistain Kit | |
| Histological Techniques of Staining (Animal Tissue)[Ch.8] | 11-12 |
| Carolina Redistain Kit | |
| Microscopic Measurements [Ch.8] | 13-14 |
| Microscope with (micrometer eyepiece) recommend 2 | |
| Introduction to Colorimetry [Ch.9] | 15-16 |
| Digital spectrophotometers recommend 2 Cuvettes | |
| Using the Spectrophotometer [Ch.9] | 17-18 |
| Digital spectrophotometers recommend 2 Cuvettes | |
| Manometric Lab Techniques – Seeds [Ch.10] | 19-20 |
| Cell Respiration Kit Flinn Scientific recommend 2-4 | |
| Manometric Lab Techniques – Mice.[Ch10] | 21-23 |
| Metabolism Equipment Kit Finn Scientific recommend 2 | |
| Paper Chromatography.[Ch.11] | 24-25 |
| Paper Chromatography Kit Flinn | |
| Thin Layer Chromatography.[Ch.11] | 26-27 |
| Thin Layer amino acids Kit | |

Table of Contents continued

| | |
|--|-------|
| Analyzing Central Tendencies in Raw Data.[Ch. 15]..... | 28 |
| Data are in this syllabus | |
| Calculating Central Tendencies for Lists of Data with Excel[Ch. 15]..... | 29 |
| Spec 20 data are in this syllabus (and access to Excel and Internet) | |
| Standard Deviation Problems – Set 2.[Ch.15]..... | 30 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Analyzing the Variance with Student T-Test.[Ch. 16]..... | 31-32 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Student T-Test Problems – Set 1.[Ch.17]..... | 33 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Probability Table.[Ch.17]..... | 34-35 |
| In this syllabus | |
| Student T-Test Problems – Set 2.[Ch.17]..... | 36-37 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Altering T values by changing variables Worksheet- [Ch.17.]..... | 38 |
| Computerized T-Test Results [Microsoft Office] ..[Ch.17]..... | 39 |
| Data are in this syllabus (and access to Excel and Internet) | |
| ANOVA Briefly Explained..... | 40-41 |
| Data are in this syllabus (and access to Excel and Internet) | |
| The Mann-Whitney U-Test for Behavioral Studies..... | 42-43 |
| Data are in this syllabus (and access to Excel and Internet) | |
| U-Test Probability Table..... | 44 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Mann-Whitney Problems-set 1..... | 45 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Computerized Mann-Whitney Worksheet set 2..... | 46 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Linear Regression with Problem.[Ch.17]..... | 47-48 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Correlation Coefficient with Problem.[Ch.17]..... | 49-50 |
| Data are in this syllabus (and access to Excel and Internet) | |
| Computerized Linear Regression Worksheet [Microsoft Office] ..[Ch.17]..... | 51-52 |
| Pie Charts in Excel [Ch.15.]..... | 54 |
| Data are listed on the worksheet in this syllabus | |
| Extensive lists of possible investigations (8 pages)[Ch.7.]..... | 55-71 |
| Recommended References [Ch.3.]..... | 72-76 |
| Appendix | |
| Power Point Matrix..... | 77-78 |
| A. Siemens Science Talent Search..... | 79 |
| B. Indiana Academy of Science Talent Search..... | 80 |
| C. Regenon Science Talent Search..... | 81 |
| D. Indiana Science and Humanities Symposium..... | 82 |
| E. Indiana Junior Academy of Science Fall Meeting..... | 83 |
| F. Indiana Research Grants..... | 84 |

INDIANA UNIVERSITY SCHOOL OF MEDICINE — SOUTH BEND

A partnership with the University Notre Dame August 19, 2013

Mr. Nevin
Longenecker John
Adams High School
808 S. Twyckenham
Dr. South Bend, IN
46615

Dear Mr.
Longenecker,

The research program at John Adams High School in South Bend, Indiana had a profound impact on my life and career choice. As a high school researcher, I studied the impacts of chemical agents on animal growth and development. Through this experience, I developed a passion for developmental biology and research that resulted in my decision to pursue a career as a developmental geneticist. In the Adams research course, we learned about lab techniques, the statistical analysis of data, literature searches, grant and manuscript preparation, and delivering oral research presentations. Now, as an Associate Professor of Medical and Molecular Genetics at the Indiana University School of Medicine at the University of Notre Dame, these activities still occupy most of my time on a daily basis! My laboratory studies genes that regulate development with the goals of elucidating a better understanding and potentially a means of preventing mosquito-borne illnesses and cancer.

In addition to pursuing research, I am also responsible for the equally important task of helping to train the next generation of scientists. I have observed that young researchers who had early research training experiences are the most likely to thrive in my laboratory and beyond. It is therefore critical that today's high school students have excellent scientific training, including opportunities for research, if they intend to pursue careers in science. Research experiences help students develop the critical thinking skills which are prerequisites for a career in scientific research, but are also beneficial to all students regardless of their ultimate career paths. Yet, such opportunities are unfortunately lacking in most high school, and even undergraduate curricula. The John Adams program serves as an excellent model for the development of such programs at other institutions.

Sincerely,

Molly Duman Scheel, Ph.D.

Associate Professor, Department of Medical and Molecular
Genetics, Indiana University School of Medicine, South Bend, IN
Eck Institute for Global Health and Department of Biological
Sciences, University of Notre Dame, Notre Dame, IN

Dr. Fil Randazzo-Deputy Director of Bill and Melinda Gates Foundation
May 23, 2013

The John Adams High School Research Biology Program under Nevin Longenecker was one of the best learning experiences in my entire career. As a high school student, I participated in the program from 1978-1980. It was a seminal experience in my life, instilling and nurturing a passion for research.

The program provided an outlet for my curiosity and energy as well as an opportunity to nurture my creativity. It helped build a tremendous amount of self-confidence, particularly in the areas such as independent thinking and taking on difficult challenges and tasks. It taught me how to define, attack and solve complex problems in a systematic, methodical and informed way. It also taught me the importance of communicating results and how to do so effectively. Fundamentally, the approaches I learned in the program are approaches that I use today in my own work as a Deputy Director of Global Health at the Bill & Melinda Gates Foundation. In my current role, I contribute to strategy and fund research and development programs that focus on developing products and interventions to address the needs of the underserved in the developing world.

I cannot imagine a more enriching and outstanding experience for any student, whether or not they further pursue biology, science or any other field of endeavor. The lessons are fundamental to success in today's complex and fast changing world and those lessons are not easily gained in a traditional learning environment.

Sincerely,

Fil Randazzo, Ph.D.
Deputy Director
Global Health Discovery & Translational Science
Bill & Melinda Gates Foundation

Dr. John Rearick- Attorney for protection of intellectual property (Boston)

C H O A T E

CHOATE HALL & STEWART LLP

Dear student thinking about a possible career in science,

If you are like I was in high school, you probably liked the introductory biology or chemistry courses you have taken and find yourself wanting to learn more. Or maybe you are just looking for the sort of extracurricular activity that with a lot of hard work and a little luck, you can really shine as an individual. Either way, I strongly encourage you to consider the research program described in this book. I participated in the program during my junior and senior years, and it had a tremendously positive effect on both my interest and success as a student of science. No other single academic experience was more influential on my career path.

As soon as I started the research program, I was bitten by the research bug and knew my path forward. From day one, the program imparts the same methods of scientific research in place at any higher academic research institution, from the formulation of ideas to the procurement of funding to the presentation of experimental findings. My time in the program afforded me a "toolbox" of skills that I applied throughout my academic career, including undergraduate coursework and research, authoring grant proposals, and graduate teaching and dissertation research. The program initiated a snowballing of scholarships and opportunities in higher education including a full academic scholarship to Purdue University and full five-year graduate funding as a chemistry PhD student at Harvard University.

While I ultimately decided that a career in scientific research was not my calling, I found a career that allows me to apply my science background in a way that drives the development of life-saving technologies. As a patent attorney at Choate Hall & Stewart in Boston, I help universities and life science companies translate their inventions into valuable intellectual property assets. These assets attract the investments necessary to drive the development of the next generations of cancer drugs and antibiotics. What is most exciting about my job is that I get to learn about cutting-edge research before the rest of the world hears of it.

Working with the top scientists in the pharmaceutical and biotech industries requires being able to "talk the talk." I have to not only understand the technology, but also assure my clients that I understand the challenges of science. While I have refined the necessary skills for interpreting and understanding scientific research over the years, these skills are all rooted in my experience in the research program. I wholeheartedly encourage students who have a strong interest in science to participate in the research program. At a minimum, it will put you at an advantage as college applicants. It will also be a starting point for a whole host of careers, including both traditional research careers as well as other nontraditional paths.

Best regards,

John P. Rearick, PhD, JD

Forward to the Interested Teacher

Teachers who have supervised only a few students in research often feel at a loss for direction when they consider simultaneous supervision of 15-20 students as is done in some high schools. Teachers also wonder how many students they could reasonably expect to pursue research in their setting, if given the opportunity. The following thoughts, by this author, may help in providing the interested teacher with some guidelines.

Several years ago the science teachers at John Adams High School offered guidance to interested students for conducting investigations on an individual basis, usually one or two students per year. The students received no academic credit for the work and the guidance was usually given during the teacher's preparation period or after school. Most of the research work was performed at home. Following repeated requests from students, the department members decided to offer to the students a research course in which they could receive direct supervision and also receive academic credit. In our high school, of approximately 1800 students we have 15-20 students per year directly involved in research.

This booklet "A Guide to High School Science Research with an Emphasis in STEM Education", will be most useful if an interested teacher approaches the topic of research supervision with the attitude that he "wants to" incorporate a research program into his school's curriculum. If a teacher is "looking for" reasons why he cannot offer such a course in his school, he will have no difficulty compiling such a list. A positive attitude concerning modifying and incorporating this program into a science curriculum will be well rewarded. It would be useful to read the three letters of support from practicing scientists who had completed extensive investigations while in high school. (Section II.) In addition, many opportunities exist for your students to maximize their research experience and to profit in monetary ways. Many of these opportunities are listed in Sections IV. and X. of this book.

The cost of the program varies considerably from student to student. In some cases the cost is minimal, as with a study of varying temperatures on the germination rates of wild weed seeds vs. cultivated seeds. At the upper extreme is the cost involved with purchasing diagnostic test equipment. Another variable related to cost concerns the types of equipment presently available in the school. In general, however, the research course is more expensive to fund than a typical science class. Local industries and universities are in most cases helpful, related to the use of their equipment and personal guidance. Funding is also available from sources such as AAAS and state Academies of Science. In other cases the student's parents fund the investigation.

Science Research Tentative Schedule

A. Yearly Schedule

$\frac{1}{4}$ year-Introduction to Research

$\frac{1}{8}$ year- Library/Internet Research and Proposal Formation

$\frac{1}{2}$ year- Active Research

$\frac{1}{8}$ year- Research Report

B. Daily Outline Introduction to Research (1/4 year)

Day 1. General Introduction to Research and Course Objectives-Ch. 1

2. Subdivisions of the Sciences and Realistic Expectations-Ch. 2

3. Approaches to Research and Powerpoint Presentations- Summary
Ch1 and 2

4. Introduction to Chemical Measurements- Ch. 4 Discussion

5. Measurement lab work Day 1

6. Measurement lab work Day 2

7. Measurement lab work Conclusion- Ch. 5 and 6 Assigned

8. Research Techniques and Organisms-Ch. 7 Discussion

9. Introduction to Centrifugation-Types and Separations Discussion

10. Centrifuge lab work Day 1

11. Centrifuge lab work Day 2

12. Centrifuge lab work Conclusions

13. Introduction to Microscopy and Histology-Ch. 8 and pp. 109-111

14. Plant tissue histology work (1/3 of students alternate)

15. Animal tissue histology work (1/3 of students alternate)

16. Microscopy measurements (1/3 of students alternate)

17. Introduction to Colorimetry/Spectrophotometry Ch. 9 Discussion

18. Light adsorption and solution concentration lab work

19. Spectrophotometry lab work

20. Test #1

21. Manometric Techniques-Ch. 10 Discussion

22. Manometer work with seeds or animals (1/2 of students)

23. Manometer work with seeds or animals (1/2 of students)

24. Introduction to Chromatography Ch.11 Discussion

25. Chromatography lab work -TLC

26. Chromatography lab work- paper

27. Test 2

28. Circuitry consideration- Ch.13 Discussion

29. Circuitry lab work completing circuits with multimeters

30. Altering variables in circuits lab work with multimeters

Science Research Tentative Schedule (cont.)

Daily Outline Introduction to Research (1/4 year)

Day 30. Measuring variables using Vernier circuits- lab work

31. Introduction to Statistics Ch. 15 Discussion

32. Statistical Treatment of Data- Measurements of central tendencies

33. Statistical Treatment of Data- Using Excel with central tendencies

34. Standard deviation- problem solving discussion

35. Standard deviation -problem solving

36. Test #3

37. T-test of Significance Ch. 17 Discussion

38. T-test of Significance -problem solving

39. T-test of Significance- problem solving using Excel

40. ANOVA Discussion Ch. 16

41. ANOVA problem solving

42. Mann Whitney Stats Test Ch.16

43. Mann Whitney -problem solving

44. Mann Whitney -problem solving with Excel

45. Linear regression Discussion Ch.16

46. Linear regression- problem solving

47. Correlation coefficient Discussion Ch.16

48. Calculating linear regression and correlation coefficient with Excel

49. Test #4

50. Selecting a topic to investigate-Ch. 2 Discussion

51. Examining Brief Reports in Science News and other publications.

52. Selecting a topic to investigate-- Powerpoint presentations

53. Understanding Science Journal Articles Ch.18 and Ch.3 Reading and examining, science papers, reports and articles.

54. Examining Your Proposal Outline

Examining Brief Reports of Science Research in *Science News*

ILLINOIS STANDARDS -SCIENCE AND INQUIRY FOR ENGINEERING

Aligned with National Science Education Standards

Content Standard A--As a result of their activities in grades 9-12, all students should develop understanding of:• Abilities necessary to do scientific inquiry^[17]_[SEP]

- Identify questions and concepts that guide scientific investigations.
- Design and conduct scientific investigations
- . ○ Formulate and revise scientific explanations and models using logic and evidence.
- Communicate and defend a scientific argument.

Supplies- recent issues of Science News.

It has been stated that there is nothing new “under the sun”. To a considerable extent this is true related to science research. Almost no one conducts research on an entirely new topic. Most often what is investigated is the imposition of different variables on the same subject. Sometimes investigators attempt to identify new relationships among organisms.

Homework assignment

1. Over the next two days read this issue of Science News.
2. Write a paragraph summary for each of FIVE articles indicating briefly
 - a. the problem being investigated
 - b. the researcher’s findings or conclusions.
3. ALSO using your own reasoning ability, discuss what would or could be TWO additional areas of investigation on each of these topics

Be sure to list the title for each of the articles. Also list the date of the issue of *Science News* you reviewed.

Understanding Science Journal Articles

(with emphases on measurements and units)

ILLINOIS STANDARDS SCIENCE AND INQUIRY FOR ENGINEERING

Alignment with National Science Education Standards

Content Standard A--As a result of their activities in grades 9-12, all students should develop understanding of:

- Abilities necessary to do scientific inquiry^(SEP)
 - Identify questions and concepts that guide scientific investigations.
 - Design and conduct scientific investigations.
 - Formulate and revise scientific explanations and models using logic and evidence.
 - Communicate and defend a scientific argument.

All researchers wish to communicate with other interested scientists. They are concerned that the science community learns about their research findings. Since there are many research investigations being reported each day and from many places on the earth, the articles must be technical, exact, concise but understandable.

During this year you will likely be reading 8-12 science journal articles. You will be writing reviews on at least 8 articles which will provide much of the background information for your research report of next May.

The articles handed out today are representative of science research reports. You will likely need to read the article at least two times. Some of the specific details of equipment and procedures you may not completely understand but you should be able to generally understand the researcher's hypothesis, methods and results.

Write a two page review of the article in your own words. Your paper should include the following information;

1. List the titles of article and author(s)
2. Rephrase the hypothesis(es) being tested or implied.
3. Describe the Type(s) of organism used, the equipment used or the prototype developed.
4. Contrast their control and experimental groups.
5. List the procedural steps which implied that the researcher made specific concentrations of solutions or administered agents which had a specific quantitative aspect. List quantities and/or units associated with the administered and/or agents.
6. List by name or description the types of tests performed or measured to obtain their results (data). List quantities and/or units associated with the collected data.
7. Discuss their results.
8. In which science subdivision would this work be classified? Explain your choice.
9. What scientific approach was used by this researcher? Explain your choice.
10. Was this a pure science or an applied science investigation? Explain your choice.

PRECISION IN MEASUREMENT

In scientific measurement, the degree of precision required varies with the type of research being performed. Sometimes very precise measurements are necessary and minutely calibrated equipment, such as an analytical balance and micrometer eyepieces are used. At other times “rough estimates” are adequate. For example, determining the mass of a mouse to 0.001g is not reasonable since the mass would differ depending upon when the mouse had last eaten. In general, one should use precise measurements, since the work may be repeated at another time or at another location by another researcher.

Ref. Experimental Biology Ch. 3 van Norman,
World of Chemistry by Zumdah/Zumdahl Ch.9

Content Standard 1.5: Describe solutions in appropriate concentration units (be able to calculate these units) such as molarity, percent by mass or volume, parts per million (ppm), or parts per billion (ppb).



Procedure

- A. Work in pairs if you like, however, each person should complete his own lab write-up and list his own calculations.
- B. Use the equipment as it becomes available. Complete exercises 1-8 in any order but do keep your numbering the same as below.

1. Concentration-Molarity (solid)

The formula for sodium bicarbonate is NaHCO_3 . Make up 100 mL of 0.25M sodium bicarbonate solution. List your reasoning and show your calculations. Submit 5mL of your final solution.

2. Concentration-Molarity (liquid)

Make up 200 mL of 0.1M ammonium hydroxide solution. The stock solution is 8M. Use the relationship $C_1V_1 = C_2V_2$. Submit 5 mL of your final solution. Show your calculations.

3. Percentage problem (solid)

Produce 150 mL of a 3.2% sucrose solution. List your reasoning and show your calculations. Submit 5mL of your final solution.

4. Percentage problem (liquid)

Begin with 5 mL of a 10% methylene blue dye stock solution. Dilute the original quantity of the dye until you have produced 0.2% dye solution. $C_1V_1 = C_2V_2$ List your reasoning and show your calculations. Submit 5mL of your final solution.

5. Dilution problem (concentration/mL)

The commercial bottle of Vitamin C contains 3mg Vitamin C/5mL of solution. Prepare a solution containing 1mg vitamin C/1mL of solution. List your reasoning and show your calculations. Submit 5mL of your final solution.

6. Concentration exercise. (parts per ---- volume)

Making up solutions with pp volume (i.e. ppt, ppm, ppb) are really % problems. So as not to make up billions of mL of solution a student would usually make up 100 mL of a solution and then dilute that solution to produce the desired low concentration. Use 1mL of methyl red dye in 99mL of water =1pph. Remove a quantity of this 100 mL and make a solution that has 3ppm of red dye. List your reasoning and show your calculations. Submit 5mL of your final solution.

7. Mass Determination (% error exercise)

Using a triple beam balance determine the combined mass of one of the containers of sand. (Mass of container and sand-don't empty the sand.) How many significant digits are possible in your answer using this balance? Refer to pages 53-55, if you are unsure. Check your unknown mass # with the instructor. Determine your percent error by using the following formula.

$$\% \text{ error} = \frac{\text{Difference between analytical and triple beam mass}}{\text{analytical mass}} \times 100$$

8. Temperature Conversions

Become familiar with the following temperature comparisons. Show calculations to convert from F to C and from C to F.

- Normal body temperature
- Average room temperature
- Typical outdoors summer temp. in South Bend
- Freezing temperature of water
- Boiling temperature of water

The Centrifuge

A centrifuge is an instrument designed to separate materials of different density from each other by virtue of a centrifugal force. Since the centrifugal force is similar in its effects to gravity, most things that can separate in a centrifuge would eventually settle out because of gravity, but a very long period of time may be required. The centrifuge speeds up this separating effect by supplying the force of several hundred or thousand units of gravity. In research at least one of the components to be separated is a liquid. The other material may be a solid(s) or some other liquid(s). The following lab exercise is designed to acquaint you with the operation of the centrifuge, the types of solutions that can be separated and with the calculations involved in determining the force on the particles.

I. Separation of plant components (differential centrifuge)

1. Use a scalpel to cut a cherry tomato in half.
2. Scrape out all of the tomato contents except the skin from each half of the tomato and add 5mL of tap water.
3. Use a pestle and grind up the contents for about one minute.
4. Transfer the contents to a test tube, add water to fill the test tube up to 1cm below the top, shake well and place in centrifuge well.
5. Add a corresponding tube of water in the opposite well to balance the centrifuge, if needed.
6. Centrifuge the tubes for 10 min. Remove tubes AFTER the centrifuge has stopped.
7. Identify and Compare the percentages of the distinct layers.
8. Clean/brush out the test tubes and clean up the lab area.

Questions to answer while you wait and later.

- Q.1. What does RCF represent?
- Q.2. Why is the term RCF a more meaningful term than centrifugal force?
- Q.3. Calculate the number of "g-forces" acting on the tomato components
Refer to Page 85 $R=13\text{cm}$ $N=1750\text{rpm}=29\text{rps}$
- Q.4. What pattern developed with the tomato components in the test tube due to the centrifugation? How many distinct layers are there?
Describe these layers.
- Q.5. What does this pattern in the test tube tend to imply about the original mixture?
- Q.6. Why is particle mass less important than particle density in discussing centrifugation?
- Q.7. Compare the percentages of the distinct layers.

II. Separation of whole cells from a liquid suspension

1. Stir up the stock yeast solution and transfer 7mL to a small test tube.

2. Remove one drop of the suspension and place it on one end of a clean slide. Observe under the high power magnification of the microscope (400X) and estimate the number of cells in $\frac{1}{4}$ of the field, and use the factor of 4x.
3. Centrifuge the 7mL sample for 10 minutes.
4. Remove the tube and use a clean dropper to transfer a drop of the supernatant fluid to the opposite end of the original slide. Estimate the yeast population in the field of view. Compare the two population counts.

- Q.8. What force acting within the test tube while it is being centrifuged tends to impede or retard the sedimentary process? Describe the action of this force.
- Q.9. List three situations in which a person doing a research investigation might want to separate out or concentrate whole cells.

III. Percentage of Separation by Centrifugation

Almost all of the use of centrifugation involves the separation of differing masses in a solution. It is sometimes important to note the quantitative differences in the centrifuged components. The percentage relationship between blood plasma and the solid components of the blood is critical to the health of the person.

Procedure

1. Transfer 8mL of simulated whole blood to a small test tube.
2. Centrifuge the "blood" for 10 minutes.
3. Using a pipette, carefully remove the supernatant plasma to a graduate and determine the volume of both components.

- Q.10. Calculate the percentages of plasma and solid components.
- Q.11. How do these percentages compare with the known percentages for Normal blood?

Clean/Brush out the test tube.

IV. Separation of Enzymes in the supernatant

Enzymes usually operate best at a specific concentration. Enzyme molecules are often needed in a pure state and thus must be separated from the substrate on which they are acting. Washing the substrate and then centrifuging out the substrate can often separate the enzyme.

Procedure

1. Stir well the stock solution of starch-amylase-water and glucose
2. Transfer 8mL of this solution to a small test tube.
3. Remove a drop of this solution and test the substrate for starch on one end of a slide with iodine solution. KEEP this slide and the drop of solution.
4. Centrifuge the solution within the tube for 10 minutes.
5. Transfer a drop of the supernatant to the other end of the slide and retest for starch. Compare your two starch tests.

Clean/Brush out the test tube.

- Q.12. Knowing that starch was originally in the solution, explain your starch tests.
- Q.13. The starch molecules are very large molecules, the enzyme molecules are moderately large and the end product is glucose, which is a relatively small molecule. What will be the main problem associated with trying to separate out a pure enzyme quantity for the supernatant? How could this problem be solved?

Conclusions

1. Refer to the flow chart on page 81 and discussion on pages 81 and 82. The first time the mixture was centrifuged, the sediment was discarded and the chloroplasts were in the supernatant fluid. With the second period of centrifugation the supernatant fluid was discarded and the chloroplasts were in the sediment. What was different about those two periods of centrifugation? List two variations.
2. The force acting on a particle is calculated from the following formula:

$$F_c = m(2\pi N)^2 R$$

What are three factors which can vary and affect the centrifugal force acting on particles being separated?



Ultra-Centrifuge



Variable Speed
Centrifuge



Fixed Speed Centrifuge

HISTOLOGICAL TECHNIQUES OF STAINING

In general, the following procedures are used for mounting and staining tissues for permanent slides.

- | | |
|---|--|
| 1. Fix tissue in preservative | 6. Dissolve paraffin with xylol |
| 2. Dehydrate through a series of alcohols | 7. Pass slide through alcohols and water |
| 3. Clear tissue | 8. Stain tissue |
| 4. Embed tissue in paraffin | 9. Dehydrate with alcohol |
| 5. Section tissue with a microtome | 10. Mount in balsam |

1. Plant and animal tissues which have been removed from their natural surroundings change so quickly that within a few minutes they are no longer suitable for microscope examination. This degenerative process is called autolysis and you must try to slow down that process. As soon as the tissues or structures are removed from the host organism transfer them to a fixing solution which kills the cells but also kills associated bacteria. In order that the fixing solution will penetrate the object equally from all sides, place it in a glass vessel with the tissue raised up on a small quantity of glass wool. There are many types of fixing solutions and one should refer to a research manual to determine the best type for the tissue being preserved. Ethyl alcohol, formaldehyde and Fleming's solution are the most common fixatives. The time of fixing varies with the tissue and with the agent, however, 24hrs. is a usual time period.
2. In the fixative the tissues are quickly killed and also hardened. The next step is dehydration of the tissues. Water must be removed from the tissue and the specimen separated from the embedded wax. Usually the tissues are transferred from the fixative to a 70% ethyl alcohol solution. It may later be transferred to a 90% ethyl solution. The time spent in the alcohol ranges from 3hrs to 24hrs.
3. Next the specimen is transferred to a container holding xylol which acts as a clearing agent to rid the tissue of the alcohol. Here it should remain for 1-3 hrs. If the xylol becomes cloudy, return the tissue back to the alcohol solution for another 3 hrs. When the solution has become clear the tissue is ready for embedding in wax for slicing.
4. A small paper box, of a size that will fit into the microtome holder should be formed and filled with wax and kept warm in an oven. The specimen should be placed into the melted paraffin wax and left for two hrs. or until hardened. After two hrs. place the paper box containing the wax and the specimen into a shallow tray of cool water, but do not permit the water to cover the top of the wax. Blow on the surface of the paraffin until a film begins to form. When the paraffin has hardened sufficiently on the surface, submerge the box and let it harden throughout. Then remove the block and trim it so it can fit into the microtome holder.
5. Attach the block to the microtome holder and spread a small amount of melted wax around the edges. There are many types of microtomes, therefore, follow the instructions given in class concerning its operation. Set the microtome so that the sections are from 6-10 microns thick. These paraffin slices are very thin and delicate. It is best to fix them on a glass slide with Meyer's albumen and place in an incubator at 37C for 24 hrs.
6. Follow the procedures listed on one of the next two pages for staining and counterstaining the tissue.

HISTOLOGICAL TECHNIQUES OF STAINING-plant tissues

Many hours of time are usually involved from the time plant and animal tissues are removed from the parent organism until they are permanently mounted on a microscope slide. Some of you will become involved in this rather long and complex process when you get started with your research investigation. The general pattern of the steps are listed on the other handout, although, there are many modifications and exceptions to the procedure. To some extent, "one learns by doing."

Structures and Functions of Living Systems3.1: Describe features that all cells have in common and contrast those with distinctive features that allow them to carry out specific functions. Relate organelles and other distinctive cellular structures (e.g., plasma membranes, ribosomes) with their functions. [Cell Structure](#), [Osmosis](#), [Paramecium Homeostasis](#), [RNA and Protein Synthesis](#)

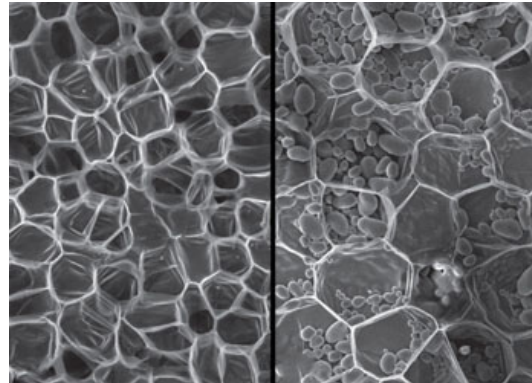
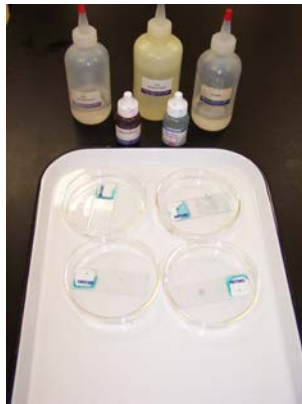
[Supplies -Histological Kit from Carolina Biological Supply](#)

The selected materials we are going to work with have been killed, fixed, dehydrated, infiltrated with wax, embedded with wax, sectioned and mounted on glass slides. These steps would have required about 10 days to complete. Six processes remain to be completed, to dissolve the wax, stain the tissue, remove this primary stain, add a secondary stain, remove that stain and make a permanent mount.

Procedures for using on plant tissues

- | | |
|--|----------------------------|
| 1. Secure a slide identified as noted to the right. | A. dicot root (ranunculus) |
| | B. monocot root (corn) |
| 2. Place the slide in one half of a glass petri dish with the tissue side up. | C. dicot stem (sunflower) |
| | D. monocot stem (corn) |
| 3. Place 4 drops of xylene on the tissue for 1 min. or until the paraffin dissolves. | E. dicot leaf (privet) |
| | F. monocot leaf (corn) |
| 4. Pour off the xylene on a paper towel and again add 4 drops of xylene to the tissue for a period of 1 min. Pour off any remaining xylene. | |
| 5. Add 8 drops of anhydrous isopropyl alcohol for 1 min, then pour off the alcohol onto a paper towel. | |
| 6. Add 8 drops of 70% alcohol for 1 min, then pour off alcohol. | |
| 7. Add 4 drops of safranin stain and continue adding the stain as necessary to keep the tissue wet for 8 minutes, then pour off any excess on a paper towel. | |
| 8. Add 8 drops of 50% alcohol for 1 minute. Remove excess stains from the glass slide with a paper towel. DO NOT touch the tissue. | |

9. Add 8 drops of 70% alcohol for 1 minute. Pour off alcohol.
10. Add 4 drops of the secondary fast green stain and add additional stain to keep the tissue wet for 8 minutes. Pour off excess stain.
11. Add 8 drops of anhydrous alcohol for 1 minute, then pour off excess by tilting the slide on a paper towel.
12. Add 4 drops of xylene. If cloudiness occurs, continue adding additional drops of xylene until the fluid is clear.
13. Exhale on the slide until dry. Place the slide on a new paper towel and add a drop of balsam on the tissue. Add a cover glass starting on a 45-degree angle so as not to trap air under the cover glass.
14. Examine your slide under the microscope. Identify by name the structures which absorbed different stains. Include a labeled 100X view or a labeled 430X view.



Histological Techniques of Staining Animal Tissues

Structures and Functions of Living Systems3.1: Describe features that all cells have in common and contrast those with distinctive features that allow them to carry out specific functions. Relate organelles and other distinctive cellular structures (e.g., plasma membranes, ribosomes) with their functions. [Cell Structure](#), [Osmosis](#), [Paramecium Homeostasis](#), [RNA and Protein Synthesis](#)

B. Procedural Steps for Animal Tissues

- | | |
|--|-----------------------------------|
| 1. Secure a slide identified as noted to the right. | A. Liver tissue from congo eel |
| 2. Place the slide in one half of a glass petri dish. | B. Small intestine from congo eel |
| 3. Place 4 drops of xylene on the tissue for 1 minute or until the paraffin dissolves. | C. Skin tissue from congo eel |
| 4. Pour off the xylene on a paper towel and again add 4 drops of xylene to the tissue for a period of 1 minute. Pour off any remaining xylene. | D. Testes from grasshopper |
| | E. Taste buds from rabbit tongue |
| | F. Mammal spinal cord |
-
5. Add 8 drops of anhydrous isopropyl alcohol for 1 minute, then pour off the alcohol onto a paper towel.
 6. Add 8 drops of 70% alcohol for 1 minute. Pour off excess alcohol.
 7. Add 8 drops of 50% alcohol for 1 minute. Pour off excess alcohol.
 8. Gently dip the slide 4 times into a beaker of distilled water.
 9. Add 4 drops of the primary hematoxylin dye and then continue adding this dye as necessary to keep the tissue wet.
 10. Gently dip the slide 4 times into a beaker of distilled water.
 11. Add 8 drops of 50% alcohol for 1 minute. Remove the stains from the glass slide with a paper towel. DO NOT touch the tissue!
 12. Add 4 drops of the counterstain-erythrosin and continue adding this stain as necessary to keep the tissue wet for 3 minutes. (Fast green can also be substituted as a counterstain.)
 13. Add 8 drops of anhydrous alcohol for 1 min, then pour off and tilt the slide.
 14. Add 4 drops of xylene. If cloudiness occurs, continue adding additional drops of xylene until the fluid is clear.

15. Exhale on slide until dry. Place the slide on a paper towel and add a drop of balsam. Place a cover glass in such a fashion so as not to trap air under the glass.
16. Examine the prepared slide under a scope. Identify the structures and note the specific colors absorbed by the structures. View under 100X and 430X.



MICROSCOPIC MEASUREMENTS

The researcher often wishes to know if the object being viewed is normal in size as compared with a control organism (object). Because these objects (cells) are usually very small, metric units of length such as centimeter or millimeter are too large to be used. The unit of length commonly used in a light microscope is the micron, which is equal to 0.001mm. Cellular components such as membranes or vacuoles are usually measured in angstrom units which are one-ten-thousandth of a micron. Although we will not be measuring with the micron unit, you will see it listed in the literature.

Reference Experimental Biology Ch -8

Structures and Functions of Living Systems3.1: Describe features that all cells have in common and contrast those with distinctive features that allow them to carry out specific functions. Relate organelles and other distinctive cellular structures (e.g., plasma membranes, ribosomes) with their functions. [Cell Structure](#), [Osmosis](#), [Paramecium Homeostasis](#), [RNA and Protein Synthesis](#)

Procedure

- A. Secure a microscope containing a micrometer scale in the eyepiece. Clean all three lenses including the eyepiece with lens paper. Make all measurements based on the following data submitted by the microscope company: diameter of 100X field = 1760 μ , 400X = 435 μ .

B. Monocot-Corn Stem - a prepared slide. Use 100X (yellow lined lens)

1. Determine the metric value of the numbered markings and the subunits between the numbered markings. Record on your lab paper.
3. Observe the corn stem under 100X. What does the printed X.S. or C.S. represent?
3. Xylem and phloem cells occur in bundles in monocot plants. Determine the average size of four typical bundles. Record calculations.
4. You will notice within each bundle two very large cells called metaxylem cells. Determine the average diameter of 4 of these large cells. Record calculations.

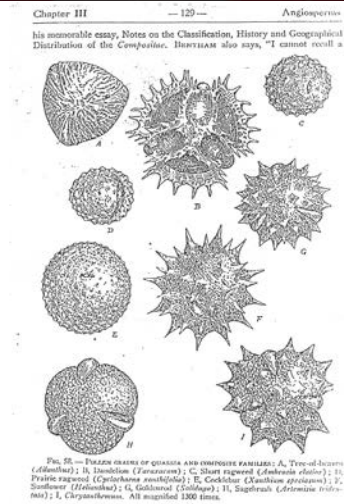
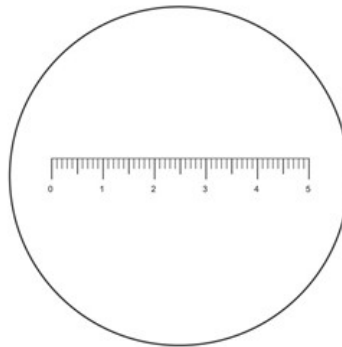
B. Allium Root Mitosis Slide --Use 100X lens

1. Using the prepared root mitosis slide position the slide so that you are observing a section about half way between the upper and lower end of the root.
2. What does the printed L.S. represent?
3. How many cells wide is the midregion of the root? What is the average cell width? Record calculations.
4. Switch to the high power lens 400X (blue lined lens) Determine the metric value of the numbered markings and the subunits between the numbered markings. Record on your lab paper. (Note, these sizes are not the same as in section A.

5. Measure the widest and narrowest cell in the field of view. Record.
6. Rotate the slide so that it is perpendicular to the original position. Record the lengths of the longest and shortest cells in the field of view.
7. Record the diameter of a typical nucleus.

C. Pollen Cell Volumes Use 400X lens.

1. Prepare a wet mount of the ragweed pollen solution. Add a cover glass.
2. Determine the diameters of the largest and smallest pollen cells. Show calculations of the volumes of these two cells.
3. Percentage-wise how much larger is the one cell compared to the smaller cell? Show calculations.



Introduction to Colorimetry

Colorimetry is very important for analyzing the composition of different liquids or solids that are dissolved in a liquid. All colorimetric measurements are relative to a known concentration. For example, if we wish to learn how much glucose is in a urine sample, we would first determine the light absorbance for a known concentration of glucose of urine. By comparing the absorbance of the known concentration with the absorbance of the unknown concentration we can calculate the glucose concentration. For many analyses a reactive compound must be added to the sample in order to produce a colored solution.

The following exercise demonstrates the relationship between the concentration of methylene dye and its absorbance of light as well as its transmittance of light.

IL Engineering Content Standard A--As a result of their activities in grades 9-12, all students should develop understanding of:

- Scientists rely on technology to enhance the gathering and manipulation of data. New techniques and tools provide new evidence to guide inquiry and new methods to gather data, thereby contributing to the advance of science. The accuracy and precision of the data, and therefore the quality of the exploration, depends on the technology used.

References Experimental Biology by Van Norman – Ch.9 Chemistry and Chemical Reactivity by Kotz and Treichel - Ch 23

Materials/equipment- Spectrophotometer, 10 ml graduated cylinders, methylene dye, droppers

Procedure Remember to handle the cuvettes ONLY by the top 1 cm .

1. Secure approximately 20mL of the stock methylene dye solution in a beaker.
2. Consider the stock solution to be 100% dye solution. Transfer 8mL of the 100% dye to a cuvette. Place this cuvette in position #1 in test tube rack.
3. Transfer 4mL of the stock solution from your supply beaker to a second cuvette. Add water to make a 50% solution.
4. Transfer 2 mL of the stock dye into cuvette #3. Add water to make a 25% solution.
5. Continue this type of dilution until you have at least 6 cuvettes to compare. Be sure to keep these cuvettes IN ORDER of decreasing concentrations.
6. Prepare a water blank by placing 8 mL of distilled water in a cuvette.
7. Obtain 2 of the unknown tubes from your instructor and estimate the concentrations of the unknowns by comparing them with your standards.
8. Using a piece of bibulous paper, wipe the outside of each cuvette.
9. List your unknowns by tube # and list your estimated concentrations.

10. Set the Spec. 20 on a wavelength of 355nm. With the lid closed, use the left knob and set the meter on 0% trans. Open the lid and properly insert a WATER BLANK . Close the lid. Use the right knob and set the % transmittance at 100%.(water/NO dye) Then remove this water blank.
11. Insert your 6 cuvettes into the colorimeter and read %Transmittance.
Adjust the concentration knob to Absorbance and read units of Absorbance for each of the your cuvettes.
12. Plot two graphs. Dye Concentration vs % Transmittance
Dye Concentration vs Units of Absorbance
13. Calculate the concentrations of your TWO unknowns by using your graphs.
14. Determine the percent transmittance and the absorbance for each of your unknown
15. Compare this %transmittance with your original estimates. Determine your % error for each tube.
16. Additional activity.
List the basic assumptions about light which underlie the operation of a colorimeter instrument. Explain how each assumption is related to the quantification of a compound in solution.



Using the Spectrophotometer (Spec 20)

In the great majority of tests performed in clinical laboratories quantification is based on relationship between the absorption or emission of light and the concentration of the substance being tested. The instruments used for such measurements are called spectrophotometers. We can observe visually that the more concentrated a solution the more light that is absorbed and the less that is transmitted. In practically all colorimetric procedures the substance being tested, i.e. glucose, protein, uric acid, cholesterol, etc. are not themselves colored, but form a colored complex when mixed with specific color reagents.

IL Engineering Content Standard A--As a result of their activities in grades 9-12, all students should develop understanding of:

- Scientists rely on technology to enhance the gathering and manipulation of data. New techniques and tools provide new evidence to guide inquiry and new methods to gather data, thereby contributing to the advance of science. The accuracy and precision of the data, and therefore the quality of the exploration, depends on the technology used.

References Experimental Biology by Van Norman – Ch.9 Chemistry and Chemical Reactivity by Kotz and Treichel - Ch 23

Materials/equipment- Spectrophotometer, 10 ml graduated cylinders, phenol red dye, droppers



Procedures

1. Prepare 10mL of a standard phenol red dye solution using appropriate amounts of distilled water and phenol red dye. (Use a range of 35-65%.)
2. Pour this STANDARD into a cuvette filling it about 2/3 full. (DON'T Overfill)
3. Secure an UNKNOWN and estimate its concentration based on the concentration in your STANDARD.
4. Prepare a distilled water BLANK cuvette.
5. Set the Spec 20 on 545nm using the top knob.
6. Adjust the Spec 20 with the left Knob to 0% transmittance with NO cuvette in the well and the lid closed.
7. Wipe the outside of the BLANK (0% dye) with a Kwik Wipe. Insert the blank into the well and close the lid. With the right lower knob set the transmittance to 100%.
8. Press the mode key to read absorbance units.
9. Wipe each cuvette with a kwikwipe before inserting into the well. Record the absorbance value for the STANDARD and the UNKNOWN cuvettes.
10. Determine the unknown concentration by using the following formula;

$$\frac{\text{Absorbance units of UNKNOWN}}{\text{Absorbance units of STANDARD}} \times K = \text{concentration of unknown dye}$$

K is the concentration of the STANDARD

11. Prepare 10mL of a STANDARD methylene blue dye solution using appropriate amounts of distilled water and methylene dye. (use 35-65%)
12. Secure an UNKNOWN methylene dye solution and estimate its concentration based on your STANDARD.
13. Set the spec 20 on 355nm using the top right knob.
14. Using steps 6-10 determine the concentration in your UNKNOWN.
15. Additional questions. In most testing, at least three cuvettes are used
 - a.) cuvette A with a color reagent but none of the compound being tested.
 - b.) cuvette B with a color reagent and a known conc. of the compound being quantified.
 - c.) cuvette C with a color reagent and an unknown quantity of the compound being quantified.Explain why cuvettes A and B were prepared before reading cuvette C.
16. Using drawings and discussion, explain basically how a Spec 20 instrument functions.

Measurement of Gas Exchange (in seeds)

Frequently, it is desirable to follow the progress of some biological reaction in which gases are used up or produced. Two such processes are respiration and photosynthesis. Carbon dioxide and oxygen are exchanged in both of these reactions. These gases can be measured in several different ways. However, the manometric method of measuring rates of metabolic gas exchange is used in almost every cell physiology laboratory in the world. You will construct a small manometer for the measuring gas consumption in seeds and then use a commercial manometer for measuring gas consumption in mice.

Standard 5: Behavior of Gases

Core Standard^[SEP] Using the kinetic molecular theory, describe and explain behavior of ideal gases.

Core Standard^[SEP] Examine the relationship between number of moles, volume, pressure, and temperature for ideal gases, using the ideal gas equation of state $PV = nRT$.

C.5.1 Use kinetic molecular theory to explain changes in gas volumes, pressure, moles, and temperature.

C.5.2 Using the ideal gas equation of state, $PV = nRT$, calculate the change in one variable when another variable is changed and the others are held constant.

C.5.3 Given the equation for a chemical reaction involving one or more gases as reactants and/or products calculate the volumes of gas assuming the reaction goes to completion and the ideal gas law holds.

References: Experimental Biology (Ch.10)
Chemistry (Ch.4)
High School Biology (Ch.6)

Procedure: Day One – Respiration in seeds (1/2 Class) – Mouse Respiration (1/2 Class)

1. Refer to the drawing on the next page and assemble the manometer.
2. Support the extended end of the capillary tube with a ring stand in such a manner that it is level.
3. Place a meter stick behind the capillary tube in such a way as to be able to measure gas uptake as the meniscus moves through the tube.
4. Blot off 20 wet seeds and determine the mass to the nearest 1/100 of a gram.
5. Place the seeds into the bottom of a large test tube.
6. Add a cotton plug above the seeds. Add 5 grams of Caustic KOH above the cotton (about 1 spoonful).
7. Lower the test tube assembly into the bath water. The water temperature should be equal to that of the air.
8. With a dropper, add a drop of water to the end of the capillary tube.
9. Record starting time and meniscus location in the tube.
10. While you wait:
 - a) Calibrate the volume of the test tube. (Remember: $\text{vol.} = \text{length} \times \pi R^2 = \text{mm}^2$)

- b) Determine the current barometric pressure in mm of Hg (via Internet)
 c) Determine the temperature of the water in the bath.
11. Determine the O₂ consumption of the seeds for a 20 min. time period.
12. Using the following formula, determine the volume of O₂ used to correct STP.

$$\frac{P_1 V_1}{P_2 V_2} = T_1 / T_2$$

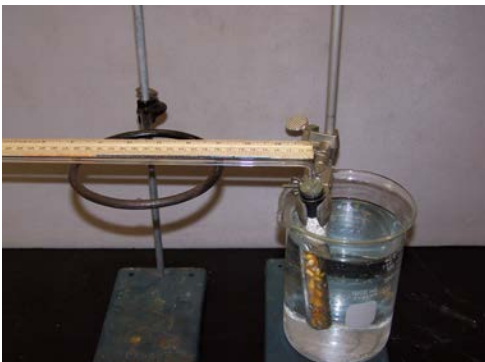
$$\frac{T_2 P_1 V_1}{T_1 P_2} = V_2$$

P₁= Atmospheric pressure of today in the room
 V₁= Calculated volume of gas used in manometer
 T₁= Temp. in water bath converted to 273+ room temp
 P₂= Standard atmos. Pressure = 760mm of Hg
 T₂= 273 C
 V₂= The corrected Volume to STP

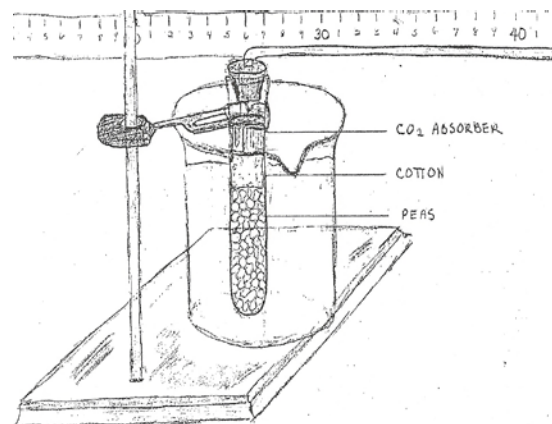
1. Why is it necessary to convert gas quantities to STP?
2. Why is the water bath needed? Explain with examples of temp. changes.
3. What would likely happen to a manometer without KOH? Explain your answer.
4. Why is 200 mm³/1g not a complete BMR?
5. Express 2000 m³/ 100g/ 20 min. in 5 other correct values of comparable values.
6. V₂ is the corrected volume for specific mass of seeds for 20 min.
 Determine the average O₂ consumption of the seeds in mm³/10 g, 20 min.
Show calculation or setup
 Determine the average O₂ consumption of the seeds in mm³/1 g/ 1min.
Show calculation or setup
7. Compare the respiration rate of your seeds with that of another type of seed.
 Calculate the percentage difference between the rates.

$$\frac{\text{Difference in rates}}{\text{Smaller rate}} * 100\%$$

Show calculations or setup



METABOLIC



MEASUREMENT OF SMALL ANIMALS

References Experimental Biology Ch-10 Chemistry Ch-4

Standard 5: Behavior of Gases

Core Standard^[SEP] Using the kinetic molecular theory, describe and explain behavior of ideal gases.

Core Standard^[SEP] Examine the relationship between number of moles, volume, pressure, and temperature for ideal gases, using the ideal gas equation of state $PV = nRT$.

C.5.1 Use kinetic molecular theory to explain changes in gas volumes, pressure, moles, and temperature.

C.5.2 Using the ideal gas equation of state, $PV = nRT$, calculate the change in one variable when another variable is changed and the others are held constant.

C.5.3 Given the equation for a chemical reaction involving one or more gases as reactants and/or products calculate the volumes of gas assuming the reaction goes to completion and the ideal gas law holds.

Procedure-Construct a 9 column table in your log book before you begin.

| Animal ID | Mass (g) | Present Bar Pressure (cm) | Present Chamber Temp (oC) | pixr ² (cm ²) | Distance Meniscus Moved | Calc Vol. (cm ³) | STP Corrected Vol (cm ³) | cc/10g/10min (cm ³) |
|--------------|-------------|------------------------------------|------------------------------------|---|-------------------------------|------------------------------------|---|------------------------------------|
|--------------|-------------|------------------------------------|------------------------------------|---|-------------------------------|------------------------------------|---|------------------------------------|

1. Cover the bottom of the metabolic chamber with a thin layer of soda lime (1 tablespoon or 18g so that the exhaled CO₂ will be absorbed. Caution; Do not Permit the animal or student to made contact with the caustic chemical. Keep the soda-lime bottle closed at all other times. Pic at this location****
2. Accurately weigh a small mammal to 0.01 g and place it in the wire holding cage of the metabolic chamber. Carefully add a wooden dowel to confine the animal.



3. Place the thermometer in the holder and slide the animal into the metabolic chamber.
4. If the device has not been recently used, wet the inside of the long glass tube with several drops of tap water.
5. After the animal has been in the chamber for at least 5 min. insert the glass tube with the rubber stopper into the chamber. Add a drop of colored soap to the far end of the glass tube. Place the meter stick behind the glass tube .
6. Record the exact time including seconds. Record the barometric pressure in cm. from Weather channel.
7. Record the total length (in cm) the soap bubble (meniscus) moves in a 10 min. time period.
8. If the meniscus does not move, check the following locations for error.
 - a. Stopper not inserted far enough into the chamber due to an air leak.
 - b. Failure to wet the interior of the long glass tube.
 - c. Saturated soda lime.
 - d. Dirty or blocked glass tube.
9. Record the Temp inside the chamber (ave. temps of steps 3 and 9) Record current bar pressure.
10. Record diameter of inside of glass tube
11. Calculate vol. of this tube cylinder $\pi \times R^2 \times \text{length meniscus moved}$. $\pi \times R^2 \times L$
12. Calculate volume of oxygen consumed.
13. Oxygen consumption must be corrected to STP (standard temp and standard pressure).

14.

$$\text{Corrected O}_2 \text{ vol.} = \frac{\text{Calculated vol\#12}}{1} \times \frac{\text{Present Bar Pressure}}{76\text{cm}} \times \frac{273\text{C}}{\text{Chamber Temp} + 2}$$

15. Oxygen consumption needs to be expressed in terms of a mass unit of the animal. This is usually expressed as cc/10g/10min. Correct your gas volume from step 14 to these units.

16. If time permits, the test should be repeated and gas volumes averaged.

17. Usually a student can evaluate two mice simultaneously using two metabolic chambers and two meter sticks.

Questions to answer while you wait;

Situation #1

- What happens to a volume of gas if the temp. is kept constant and the air pressure increases during the measurement period?
- How will this increase in pressure affect the number of oxygen molecules/cc? Explain your answer
- How will this change in pressure affect the volume of air consumed? Explain your answer.

Situations#2

- What happens to a volume of gas if the air pressure is kept constant and the temperature increases during the time period?
- How will this increase in temp. affect the number of oxygen molecules/cc? Explain your answer.
- How will this change in temp. affect the volume of air consumed? Explain your answer

PAPER CHROMATOGRAPHY

Chlorophyll contained in chloroplasts in green plant cells is the substance that makes photosynthesis possible. In addition, to two different chlorophylls (a and b), chloroplasts may contain a pale yellow pigment called xanthophyll and a deep yellow or orange pigment called carotene. Minute quantities of closely related substances can be isolated by using paper chromatography.

References: Experimental Biology—Ch 11 and BSCS Lab Block-Molecular Basis of Metabolism –Ch 2

IL Engineering Content Standard A--As a result of their activities in grades 9-12, all students should develop understanding of:

- Scientists rely on technology to enhance the gathering and manipulation of data. New techniques and tools provide new evidence to guide inquiry and new methods to gather data, thereby contributing to the advance of science. The accuracy and precision of the data, and therefore the quality of the exploration, depends on the technology used.

Procedure

1. Grind up or blend several leaves from different types of plants in an alcohol and xylene mixture 50/50%.
2. Filter the solution to separate the leaf fragments from the solvent using a funnel and small beaker.
3. Using a fume hood, vaporize the solution in order to concentrate the chlorophyll solution.
4. Attach a strip of filter paper to a paperclip hook on the underside of cork which fits in a test tube which is approximately 20 x 150mm. Make sure the paper will hang down straight in the tube without touching the tube sidewalls. The strip should be reach to within about 0.5cm from the bottom.
5. Take the cork and paper out of the tube and place them on a clean paper towel. Try not to touch the chromatography paper with your fingers. Handle paper with forceps.
6. In pencil, not ink, draw a line across the paper strip about 2cm from the bottom of the strip. Along this line spread a drop of the pigment extract from step 3 with a fine tipped pipette. Allow this line to dry, then, add another drop of the extract along the line and allow it to dry. Repeat this process four times. You may blow air over the strip to speed up the drying process.
7. Add enough of the alcohol/xylene solution (2 ml) to the test tube so the paper will dip into the solution. Be sure the pencil line with the mixed pigments does NOT come in contact with the solution.
8. Place the paper with the line of dried pigment extract in the test tube. Be sure the bottom of the paper dips into the solvent mixture, but try to keep the paper from touching the sides of the test tube. Allow the solvent mixture to rise almost to the top of the paper.

THIN LAYER CHROMATOGRAPHY (TLC)

Thin layer chromatography (TLC) is a simple, fast, and relatively inexpensive method for qualitatively separating a mixture into its components. The thin layer is prepared by spreading a slurry of an absorbent material on a glass plate and drying it to form a layer about 250 microns thick.

REFERENCE; Experimental Biology (Chapter 11)

IL Engineering Content Standard A--As a result of their activities in grades 9-12, all students should develop understanding of:

○ Scientists rely on technology to enhance the gathering and manipulation of data. New techniques and tools provide new evidence to guide inquiry and new methods to gather data, thereby contributing to the advance of science. The accuracy and precision of the data, and therefore the quality of the exploration, depends on the technology used.

Lab Procedure: Cleanliness is extremely important in this lab. DO NOT touch the surface of the TLC plates with your fingers. Handle the plates only by their top edge or with forceps.

1. Place 25 drops of the solvent in each of two developing chambers and cap. (Note: the solvent is a premixed solution of butanol/acetone/ninhydrin/ acetic acid and water)
2. Spread out a paper towel on the lab table. WITHOUT TOUCHING the surface of the TLC plates, place two plates (white side up) on the paper towel.
3. Using a pencil, place a small #1 at the top of one of the plates. Also place a small identification mark (initials) at the top. Place a very thin 1mm line up 1cm from the bottom of the strip as indicated in class.
4. Repeat step #3 on the second plate except use a #2 at the top.
5. Using a micro-applicator, spot the unknown amino acid solution A or B five or six times forming a line out from the 1mm baseline. BE SURE TO RECORD THE LABEL ON THE SMALL BOTTLE.
6. Repeat step #5 on TLC #2 using the other unknown amino acid solution A or B. RECORD THE BOTTLE LABEL.
7. Place the plates into the developing chambers, which you set up in step #1.
8. Observe the wet solvent front move up the plate and answer question while you wait.
9. Remove the plates when the wet front is about 1cm from the upper end of the plate. (About 30-55 min.) WITH A PENCIL PLACE A SMALL MARK ON THE PLATE WHERE THE FRONT STOPPED.

JWT
Front —

Base
Line

10. Allow the plates to air dry by leaning them against the developing chambers. (About 5 min.)
11. Then transfer the strips to the drying oven for 5-10 minutes at 80C.
12. Remove strips from oven and note the 2 or 3 distinct bands on each plate. Determine the Rf values of each band and identify the amino acids. Show Calculations.



From amino acid mixture A

| <u>Rf Value</u> | <u>Amino Acid</u> |
|-----------------|-------------------|
| 0.95 | Norleucine |
| 0.84 | Leucine |
| 0.62 | Tyrosine |
| 0.50 | Tryptophan |
| 0.30 | Serine |

From amino acid mixture B

| | |
|------|---------------|
| 0.82 | Isoleucine |
| 0.76 | Phenylalanine |
| 0.65 | Histidine |
| 0.44 | Alanine |
| 0.22 | Threonine |

Questions

1. List the three primary factors affecting the degree of separation of the mixed solutes.
2. Discuss how each one acts to separate the compounds in the mixture.
3. Consider your understanding of #1, how do you explain the Rf values of Tryptophan (mol. Wt. = 204) and Serine (mol. Wt. =105)
4. Why do Rf values always include an associated solvent?
5. Why are most solvents mixed solvents?

Analyzing Central Tendencies In New Data

REFERENCES: Experimental Biology (Ch. 16), Biological Science (Ch. 5),
Fundamental Statistics for Psychology (Ch. 3)

The two most important characteristics of a set of data are its central tendencies and how varied are the data points. Four different quantities help us to evaluate the collected data. The MEAN, which is the arithmetic average, gives a general impression of a representative result. The MEDIAN, is a value that divides the distribution of results into two parts, such that, there is an equal number of data points that fall above, as well as, below this value. The MODE represents the most frequently recorded result. In addition to these three measures, the RANGE of the results is also important, in that, it provides us with some impression of the extremes of the results.

Procedure

1. Calculate the MEAN, MEDIAN, MODE, and RANGE for each of the following four sets of data.

The data listed below represents the quantity of oxygen used by different types of germinating seeds.

| Corn | Peas |
|-------|-------|
| 0.200 | 0.500 |
| 0.220 | 0.620 |
| 0.210 | 0.540 |
| 0.250 | 0.70 |
| 0.240 | 0.660 |
| 0.230 | 0.500 |
| 0.200 | 0.560 |
| 0.210 | 0.500 |
| 0.200 | 0.600 |
| 0.260 | 0.630 |
| 0.250 | 0.940 |
| 0.800 | 0.580 |

The following data represent the net CO₂ uptake for Chlorella plants exposed to full and half sunlight.

| Full sun | Half sun |
|----------|----------|
| 1.800 | 1.200 |
| 0.200 | 2.200 |
| 2.800 | 1.400 |
| 2.000 | 1.000 |
| 1.500 | 1.700 |
| 2.300 | 1.200 |
| 2.300 | 1.800 |
| 1.800 | 1.600 |
| 2.400 | 1.200 |
| 1.700 | 6.100 |
| 2.200 | 1.400 |
| 2.100 | 0.100 |
| 2.900 | 1.600 |
| 1.800 | 1.300 |
| 1.800 | 1.500 |

2. Look again at the lists of data.

- Which columns have data which look “questionable”? Identify these columns and the specific values.
- Do you think the questionable values should be included with the remainder of the data for that research? Explain your answer.
- Eliminate the “questionable data points and then recalculate the new MEAN and the new RANGE.

3. Look at your mean values for the two seed groups. Do you believe there is a significant difference between their respiration rates? Explain why or why not.

4. Look at your values representing the CO₂ uptake of the Chlorella. Do you believe there is a significant difference between these two groups? Explain why or why not.

Central Tendencies Worksheet for Excel

Follow the directions for the Excel program to evaluate these data for the following central-tendencies mean, median, mode, range, and standard deviation.

Arrange the data and results in a format similar to the recommended form on the reverse side of the Excel Direction sheet. When completed, print your processed data.

TRIALS REQUIRED TO PRODUCE CONTROLLED RESPONSE

| Worm # | Original Worm | Retested Worm | Retested Worm |
|--------|---------------|---------------|---------------|
| 1 | 58 | 31 | 27 |
| 2 | 58 | 43 | 49 |
| 3 | 51 | 25 | 27 |
| 4 | 78 | 21 | 25 |
| 5 | 83 | 55 | 42 |
| 6 | 109 | 31 | 27 |
| 7 | 92 | 32 | 36 |

Height in cm at +30 days

| Plant | Insecticide #1 | Insecticide #2 |
|-------|----------------|----------------|
| 1 | 40.5 | 33.5 |
| 2 | 44.5 | 51.5 |
| 3 | 26.5 | 36.5 |
| 4 | 35.5 | 39.5 |
| 5 | 44.5 | 41.0 |

BMR in cc of O₂/10g/10 min.

| Mouse | Control | 50ppm | 100ppm |
|-------|---------|-------|--------|
| 1 | 6.48 | 5.84 | 4.09 |
| 2 | 6.07 | 5.20 | 5.61 |
| 3 | 5.39 | 5.37 | 4.54 |
| 4 | 5.73 | 5.54 | 4.09 |
| 5 | 5.83 | 5.21 | 5.08 |
| 6 | 4.97 | 5.15 | 4.45 |
| 7 | 6.07 | 6.05 | 5.15 |

STANDARD DEVIATION PROBLEMS –SET 2

Directions-Set up all problems as previously described in class.

- Two groups of pea plants were kept at different temperatures and the amount of oxygen used per hour in respiration recorded. Determine the mean values, variance and standard deviation for each group. Eliminate all values outside the +2 and -2 sd. units and recalculate the new variance for each group.

| Vial Number 26C | Pea Group #1-22C | Pea Group #2- 26C |
|--------------------|------------------|----------------------|
| 1 | 0.20cc | 0.25cc |
| 2 | 0.24 | 0.23 |
| 3 | 0.22 | 0.33 |
| 4 | 0.21 | 0.27 |
| 5 | 0.25 | 0.23 |
| 6 | 0.25 | 0.23 |
| 7 | 0.24 | 0.33 |
| 8 | 0.23 | 0.25 |
| 9 | 0.20 | 0.30 |

- There is some variation in the white blood cell counts (WBC) of normal healthy persons. However, persons having appendicitis usually also have an elevated WBC count. Determine the mean values, variance and standard deviation for each group. Eliminate all values outside the +2 and -2 sd. units and recalculate the new variance for each group.

| Normal WBC Counts | Appendicitis WBC Counts |
|----------------------------|-----------------------------|
| 8,300cells/mm ³ | 11,200cells/mm ³ |
| 5,660 | 10,600 |
| 8,450 | 11,800 |
| 10,460 | 11,000 |
| 7,000 | 11,200 |
| 6,570 | 10,800 |
| 4,360 | 9,800 |
| 8,900 | 7,600 |

- To test two promising new corn hybrids under normal farming conditions, a seed company selected nine farms at random in Iowa and planted both hybrids in experimental plots on each farm. The yields per acre in lbs. for the locations are as follows. Determine the mean values, variance and standard deviation for each group. Eliminate all values outside the +2 and -2 sd. units and recalculate the new variance for each group.

| Farm # | Hybrid S | Hybrid T |
|--------|------------|------------|
| 1 | 86lbs/acre | 80lbs/acre |
| 2 | 89 | 79 |
| 3 | 56 | 58 |
| 4 | 93 | 91 |
| 5 | 84 | 77 |
| 6 | 93 | 82 |
| 7 | 73 | 74 |
| 8 | 79 | 66 |
| 9 | 56 | 52 |

ANALYZING THE VARIANCE WITH STUDENT T-TEST

A researcher, after collecting a considerable amount of data, is interested in knowing if the data do, in fact, support the original hypothesis, if they represent an inverse function of the stated hypothesis, or if they represent no clear trend at all. Since no two organisms are exactly alike and since a living organism changes with time, there will be variations in the collected data even within the same experimental group.

The student t-test is used to determine the probability that the results obtained from two different groups was not due to chance alone. The typical form of the student t-test formula is listed below.

$$t = \frac{M1 - M2}{\sqrt{\frac{V1 + V2}{N}}}$$

M1 = mean of group 1
M2 = mean of group 2
V1 = variance of group 1
V2 = variance of group 2
N = number of individuals in the

After the t-value is calculated, based upon the mean difference, the sum of the variance and the number of organisms involved, the probability factor (P) is determined from the probability table found on the next page.

Probability indicates the likelihood of a mean difference this great being due to chance alone. For example, a probability of $p < 0.05$ indicates that in less than 5 times out of 100 would there be a mean difference this great due to chance alone, and that, more than 95 times out of 100 an investigator would have to alter a variable in order to produce a mean difference this great.

A probability of $p < 0.001$ indicates that in less than 1 time out of 1000 would there be a mean difference this great due to chance alone.

A probability of $p < 0.05$ is considered significant. A probability of $p < 0.01$ is very significant and a $p < 0.001$ is very highly significant. In some research a p-value < 0.1 is considered significant for BEHAVIORAL STUDIES.

Assignment

1. Read and review the two solved problems on the reverse side of this page.
2. Determine the t values for the Standard Deviation Problems, you processed.
3. Using the attached Probability Table, determine the p-values for each of your Standard Deviation Problems.
4. Assignment Sheet to be Submitted
 - a. Write out the name (title) of each problem.
 - b. List the mean and variance of each group.
 - c. List the T-Test equation (with values) as noted at the top of this page.
 - d. List the p-value as determined from the Probability Table
 - e. Write out the meaning of the p-value you calculated for each comparison.

ANALYZING THE VARIANCE WITH STUDENT T-TEST (continued)

Example # 1

Alan Engel studied the effects of altered diets on the aging processes in lab mice. Food consumption per group was one of the measured variables. In one series of tests he found that the control consumed 32.06 grams with a variance of 8.75 grams. The lipid group consumed 24.67 grams with a variance of 0.718 grams. The measurements were taken 4 times to determine the mean values.

$$t = \frac{32.06 - 24.67}{\sqrt{\frac{8.75 + 0.718}{4}}} = \frac{7.39}{\sqrt{2.3672}} = \frac{7.39}{1.57} = 4.7$$

The PROBABILITY N value is $(N_c + N_e - 2) = 6$

The t-value of 4.7 falls between 3.707 and 5.959

The probability value is $p < 0.01$

Therefore, the probability of getting mean values this different due to chance alone is less than 1 time out of 100 for such experiments. Or stated differently, in more than 99 times out of 100 such experiments, such a researcher would have to specifically alter the conditions for one of the groups in order to produce a mean difference this great.

The results could be stated as follow; there was a significant difference between the means of these two groups $p < 0.01$.

Example #2

Doug Brazy studied the possible effects of simulated acid rainfall on soybean plants. Normal rainfall has a pH of approx. 5.5. One of his experimental groups was watered with a simulated rainfall of pH of 2.0. At plus 30 days he determined the lengths of the plant stems. The mean stem growth for the 15 control plants was 43.65 cm with a variance of 2.857. The same number of experimental plants had a mean value of 29.07 cm with a variance of 3.401. Was there a significant difference between the two groups?

These values produce a t-value of 22.56, which places it to the right of 3.725 on the probability table. It will have a p-value of < 0.001 . There was a significant difference between these two means, since in more than 999 times out of 1000 such experiments, specific conditions would have to be imposed on one of the two groups in order to produce means this different. It could also be stated that, in less than 1 experiment out of 1000 would there be mean differences this great due to chance alone.

Data for T-test Comparisons- Set 1
Organisms/field of view in different water depths

| | Top 1 cm | at 10 cm depth | at 26cm depth |
|--|----------|----------------|---------------|
| | 20.4 | 7.8 | 4.6c |
| | 11.8 | 7.6 | 5.0 |
| | 13.6 | 11.8 | 4.4 |
| | 17.0 | 11.4 | 5.6 |
| | 19.6 | 6.6 | 5.2 |
| | 22.6 | 5.8 | 9.6 |
| | 24.4 | 6.2 | 9.8 |
| | 11.3 | 3.2 | 10.2 |
| | 18.5 | 1.2 | 7.6 |

Mean
St. Dev.
+2 SD
-2 SD

Possible
New mean

T-test results
List P Values below
Top 1cm vs 10cm

Indicate
Degree of significance

Top 1cm vs 26cm

10cm vs 26cm

Mice Metabolic Rates

CC O₂/ 10g/10 min

| Resting M Rate | Post Exercise M Rate |
|----------------|----------------------|
| 5.11 | 7.02 |
| 5.01 | 7.17 |
| 3.52 | 8.83 |
| 3.95 | 8.97 |
| 3.67 | 5.93 |
| 3.94 | 6.31 |
| 4.74 | 8.97 |

Mean
St. Dev.
+2SD
-2SD

Possible
New mean

T-test results
List P Value below
Resting vs Post Ex.

Indicate
Degree of Significance

PROBABILITY TABLE FOR STUDENT t-TEST

| Degree of Freedom | Probability <u>(p value)</u> | | | |
|----------------------|---------------------------------|-------------|-------------|--------------|
| | <u>0.1</u> | <u>0.05</u> | <u>0.01</u> | <u>0.001</u> |
| 1 | 6.314 | 12.657 | 63.657 | 636.619 |
| 2 | 2.2920 | 4.303 | 9.925 | 31.598 |
| 3 | 2.353 | 3.182 | 5.841 | 12.941 |
| 4 | 2.132 | 2.776 | 4.604 | 8.610 |
| 5 | 2.015 | 2.571 | 4.032 | 6.859 |
| 6 | 1.943 | 2.447 | 3.707 | 5.959 |
| 7 | 1.895 | 2.440 | 3.499 | 5.405 |
| 8 | 1.860 | 2.306 | 3.355 | 5.041 |
| 9 | 1.833 | 2.262 | 3.250 | 4.781 |
| 10 | 1.812 | 2.228 | 3.169 | 4.587 |
| 11 | 1.796 | 2.201 | 3.103 | 4.437 |
| 12 | 1.782 | 2.179 | 3.055 | 4.318 |
| 13 | 1.771 | 2.160 | 3.012 | 4.221 |
| 14 | 1.761 | 2.145 | 2.977 | 4.140 |
| 15 | 1.753 | 2.131 | 2.947 | 4.073 |
| 16 | 1.746 | 2.120 | 2.921 | 4.015 |
| 17 | 1.740 | 2.110 | 2.898 | 3.965 |
| 18 | 1.734 | 2.101 | 2.878 | 3.922 |
| 19 | 1.729 | 2.093 | 2.861 | 3.883 |
| 20 | 1.725 | 2.086 | 2.845 | 3.850 |
| 21 | 1.721 | 2.080 | 2.831 | 3.819 |
| 22 | 1.717 | 2.074 | 2.819 | 3.792 |
| 23 | 1.714 | 2.069 | 2.807 | 3.767 |
| 24 | 1.711 | 2.064 | 2.797 | 3.745 |
| 25 | 1.708 | 2.060 | 2.787 | 3.725 |

(Procedure for using the Probability Table on the following page)

Procedure for using the Probability Table

Step 1. Descend the Degrees of Freedom to a number representing (N-2)

$$N = (N_c - 1) + (N_e - 1) = (N_c + N_e - 2)$$

Step 2. Proceed to the right of the degrees of freedom number on a horizontal line until you locate the range of numbers within which your t value falls.

Step 3. Proceed up the column of numbers which borders the range of numbers on the left side. At the top of that column you will find the probability -p value.

Step 4. You indicate the p value as less than the number on the top of the left hand column.

Example $N_c = 8$, $N_e = 6$, $t = 3.5$ so $N - 2 = 12$

3.5 falls between 3.055 and 4.318. Thus, you would use the p value above the left hand column which is <0.01 . This is less than 1 chance in 100 such trials that you would find mean differences this great just due to chance alone, or that more than 99 times out of 100 of such test you have to alter a variable to get mean differences this great. Thus you can confidently say that there is a significant difference between these two means.

Open Stomate Area as a % of the Total Leaf Area

| Immature Small Leaf | Mature Large Leaf |
|--------------------------|------------------------|
| 2.3 | 5.7 |
| 2.4 | 5.1 |
| 2.5 | 4.9 |
| 2.5 | 4.1 |
| 3.0 | 3.9 |
| 1.9 | 4.0 |
| 2.7 | 5.3 |
| 2.2 | 8.9 |
| Mean | |
| St. Dev. | |
| +2SD | |
| -2SD | |
| Possible | |
| New mean | |
| T-test results | Indicate |
| List P Value below | Degree of Significance |
| Small leaf vs Large leaf | |

Effect of pH on Grass Seed Germination

| pH < 4.0 | pH 6-8 | pH>10.5 |
|----------------------------|--------|------------------------|
| 0 | 80 | 72 |
| 0 | 84 | 56 |
| 28 | 76 | 43 |
| 32 | 96 | 37 |
| 40 | 88 | 24 |
| 12 | 94 | 32 |
| Mean | | |
| St. Dev. | | |
| +2 SD | | |
| -2 SD | | |
| Possible | | |
| New mean | | |
| T-test results | | Indicate |
| <u>List P Values below</u> | | Degree of significance |
| <4.0 vs 6-8 | | |
| <4.0 vs >10.5 | | |
| 6-8 vs > 10.5 | | |

T-Test Problem Using Excel

Evaluation of Ideal Data

1. List on this paper 10 data points for each of the following groups. Keep the mean values of the three groups separated by approximately 20 units between each group. Select data points with very little variance.

| Control | Experimental 1 | Experimental 2 |
|---------|----------------|----------------|
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |
| _____ | _____ | _____ |

Determine the mean value and standard deviation for each group. CROSS OUT all values outside the $+2$ and -2 sd units. If values are eliminated, recalculate the new means and standard deviations. Analyze the differences of the means for significance using T-Test. Record all values on the reverse side of this worksheet.

Evaluation of data with a small (n) number

2. Eliminate the last 6 data points in each of the columns of #1. Determine the mean value and standard deviation for each group. CROSS OUT all values outside the $+2$ and -2 sd units. If values are eliminated, recalculate the new means and standard deviations. Analyze the differences of the means for significance using T-TEST. Record all values on the reverse side of this worksheet.

Evaluating data with a large variance

3. Add 6 data points in each of the columns of #2 to represent a very large variance. Determine the mean value and standard deviation for each group. CROSS OUT all values outside the $+2$ and -2 sd units. If values are eliminated, recalculate the new means and standard deviations. Analyze the differences of the means for significance using T-TEST. Record all values on the reverse side of this worksheet.

4. Compare your p-values. Discuss how these values changed as these 3 factors were altered.

ANOVA - BRIEFLY EXPLAINED

The one-way Analysis Of the Variance (ANOVA) is basically an extension of the Student T-Test for experiments involving more than two groups. It is a more stringent test than the Test. In one sense the ANOVA test of significance analyses the variance of the data where the T-Test analyses factors that influence how much the means differ.

The ANOVA test assumes a NULL hypothesis; that is to say that the groups means are all equal or that there is no significant difference between any two means of the groups. ANOVA compares the variance of the means BETWEEN the groups with the mean WITHIN the total group of tests.

The equation to make this comparison is very complex and very time consuming to perform in longhand or in using a calculator, therefore, almost everyone uses a commercial computer program to analyze data with ANOVA.

A comparison of the means squared is one way of analyzing the variance. The ANOVA calculates a "F" value. In an over simplification the equation becomes:

$$F = \frac{\text{Square of the sum of the differences BETWEEN the group's means}}{\text{Square of the mean WITHIN the total group values}}$$

If these two squared values are the same, then the F value is 1 and there is no significant difference between the mean values. As the means become separated, the F value becomes greater than 1.

If a significant difference exists, then the Sheffe's Test is used to determine between which two groups the significance exists.

The values are usually written as $F(3,44) = 14.7, p < 0.01$

This is read, that there were 4 groups (N-1) and that there were in total 48 data points recorded (n-1 in each group). Since 14.7 is much greater than 1, a Sheffe's probability tables would indicate a significant difference does exists. The $p < 0.01$ means the same as in the T-Test. A more complete explanation and the steps involved in solving a problem is available in a separate handout.

An ANOVA Problem

Subjects in the four groups were given different amounts of a sedative and at +30min. their reaction times were recorded. Their reaction times are listed below.

| Control No Sedative React. Time Seconds | Exp.1 0.1 mg Sedative React. Time Seconds | Exp. 2 0.2 mg Sedative React. Time Seconds | Exp. 3 0.4 mg Sedative React. Time Seconds |
|--|--|---|---|
| 9 | 2 | 18 | 22 |
| 6 | 7 | 11 | 13 |
| 8 | 6 | 15 | 15 |
| 7 | 4 | 13 | 17 |
| 14 | 5 | 6 | 11 |
| 2 | 9 | 7 | 12 |
| 7 | 11 | 12 | 15 |
| 8 | 3 | 9 | 16 |
| 10 | 6 | 18 | 18 |
| 8 | 5 | 8 | 13 |
| 4 | 4 | 14 | 15 |
| 16 | 14 | 13 | 16 |

Procedure

1. Determine the mean value for each group separately. There are four different means.
2. Determine the mean value WITHIN the total group.
3. Determine the differences BETWEEN the groups. There are six differences.
4. Determine the sum of the differences BETWEEN the groups.
5. Square the mean value WITHIN the total group.
6. Square the sum of the differences BETWEEN the groups' means (4)
7. Calculate the F value.

$$F = \frac{\text{Square of the sum of the differences BETWEEN the groups' means}}{\text{Square of the mean WITHIN the total group}}$$

THE MANN-WHITNEY U-TEST (for behavioral studies)

P.A. 3.1 Compute and use confidence intervals to make estimates.

P.A. 3.2 Understand hypothesis tests of means and differences between means and use them to reach conclusions.

The Mann-Whitney U-Test is an alternative to the Student t-test for analyzing behavioral data. It is commonly used where the experimenter draws two random samples from the same parent population subjects each to a different experimental treatment and compares the two on a single criterion. Of course, the experimenter may designate one of his samples as a control group, in which case the experimental treatment is no treatment at all. The U Test may also be used in the situation where independent random samples are drawn from two different parent populations and compared on a single criterion to determine whether the two populations differ. The null hypothesis tested by the Mann-Whitney U-Test is that the two populations from which the samples are drawn are identical. In an experiment concerned with the effect of caffeine on spatial associations a researcher found students required the following time periods (sec) before correctly identifying all of the pictured objects. Was the null hypothesis supported by these results?

| Control (no Caffeine) | | Caffeine (500 mg caffeine/day) | |
|--------------------------|------|-----------------------------------|------|
| Score | Rank | Score | Rank |
| 54 | 11 | 62 | 19 |
| 52 | 8 | 57 | 15 |
| 46 | 3 | 59 | 17 |
| 42 | 1 | 55 | 13 |
| 53 | 9 | 54 | 11 |
| 49 | 6 | 58 | 16 |
| 56 | 14 | 63 | 20 |
| 47 | 4.5 | 65 | 21 |
| 54 | 11 | 60 | 18 |
| 50 | 7 | | |
| R1 = 81 | | R2 = 150 | |
| N1 = 12 | | n2 = 9 | |

Steps in solving this problem;

1. Assign the rank value of 1 to the numerically lowest score, the rank value of 2 to the next higher score and so on. For tied scores, assign the average of the two rank values. Thus rank values of 5 and 6 would each be assigned 5.5. For tied rank values of three data points, use the average of the three values.

2. You will need to compute two rank value where;

$$U1 = n1 n2 + \frac{n1(n1 + 1)}{2} - R1$$

$$U2 = n1 n2 + \frac{n2(n2 + 1)}{2} - R2$$

$$U1 = 12(9) + \frac{12(n1 + 1)}{2} - 81$$

$$= 108 + 78 - 81$$

$$= 105$$

$$U2 = 12(9) + \frac{9(9 + 1)}{2} - 150$$

$$= 108 + 45 - 150$$

$$= 3$$

| EX. POP. 1 | EX. POP. 2 |
|------------|------------|
| 26 | 128 |
| 16 | 146 |
| 4 | 56 |
| 12 | 46 |
| 98 | 128 |
| 40 | 65 |

3. The value used in the Mann-Whitney test is the smaller of the two U values. In this case it is 3.
4. Refer to the attached Critical Value Table to evaluate the smaller U value. Move across the top horizontal row (n1) until you reach the n1 value. Proceed down this column until you reach the correct horizontal row corresponding to the n2 value on the left side. There may be up to 4 values corresponding to the p values listed in the two tailed probability column. To be significant at the p value listed, the U value must be equal to or less than the number listed. Since the U value 3 is less than 18, there is a p value of <0.01. Thus the null hypothesis is rejected since less than 1 time out of 100 would these rank values differ this much due to chance alone.

Internet-Mann-Whitney U Test (for behavioral Studies)

The Mann-Whitney U-Test is an alternative to the Student t-Test for behavioral studies. It is commonly used where the experimenter draws two random samples from the same parent population, subjects each to a different experimental treatment, and compares the two on a single criterion. The U Test may also be used in the situation where independent random samples are drawn from two different parent populations and compared on a single criterion to determine whether the two populations differ.

Indiana State Standards- Statistical Inference--Students use confidence intervals and hypothesis tests, fit curves to data and calculate correlation coefficients.

P.A. 3.1 Compute and use confidence intervals to make estimates.

P.A. 3.2 Understand hypothesis tests of means and differences between means and use them to reach conclusions.

1. Access the Internet and enter the following address

[//elegans.som.vcu.edu/~leon/stats/utest.html](http://elegans.som.vcu.edu/~leon/stats/utest.html)
(select-Mann-Whitney/Wilcoxon Rank Sum Test)

2. Skip the top three boxes (**n1, n2, U**)
3. Enter your data points horizontally, **separated by commas**, for the two groups in the boxes indicated as
Dataset 1:
Dataset 2:
4. After entering the data, select **Calculate Level of Significance**.
5. In your lab notebook, record the **names of groups compared**, **the U value** and **the p value- (one tailed)**.
6. At the bottom of the page select
Return to Mann Whitney U Test page
7. Continue comparing the groups in pairs.
8. Open Microsoft Excel and generate a bar graph to represent the Mann Whitney results for one comparison. Print out your graph

Critical Values For The Mann-Whitney U-Test Statistic

| N2 ↓ | Two Tailed Probability N1→ | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
|---------|----------------------------------|---|---|---|---|----|----|----|----|----|----|----|----|
| | 0.10 | | | | | | | | | | | | |
| 1 | 0.05 | | | | | | | | | | | | |
| | 0.02 | | | | | | | | | | | | |
| | 0.01 | | | | | | | | | | | | |
| | 0.10 | | | | | | | | 1 | 1 | 1 | 1 | 2 |
| 2 | 0.05 | | | | | | | | | | | | 1 |
| | 0.02 | | | | | | | | | | | | |
| | 0.01 | | | | | | | | | | | | |
| | 0.10 | | | 0 | 0 | 1 | -2 | 2 | 3 | 3 | 4 | 5 | 5 |
| 3 | 0.05 | | | | | | -1 | 1 | 2 | 2 | 3 | 3 | 4 |
| | 0.02 | | | | | | | 0 | 0 | 1 | 1 | 1 | 2 |
| | 0.01 | | | | | | | | | 0 | 0 | 0 | 1 |
| | 0.10 | | | 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 |
| 4 | 0.05 | | | | 0 | 1 | 2 | 3 | 4 | 4 | 5 | 6 | 7 |
| | 0.02 | | | | | 0 | 1 | 1 | 2 | 3 | 3 | 4 | 5 |
| | 0.01 | | | | | | 0 | 0 | 1 | 1 | 2 | 2 | 3 |
| | 0.10 | | 0 | 1 | 2 | 4 | 5 | 6 | 7 | 9 | 11 | 12 | 13 |
| 5 | 0.05 | | | 0 | 1 | 2 | 3 | 5 | 6 | 7 | 8 | 9 | 11 |
| | 0.02 | | | | 0 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 |
| | 0.01 | | | | | 0 | 1 | 1 | 2 | 3 | 4 | 5 | 6 |
| | 0.10 | | 0 | 2 | 3 | 5 | 7 | 8 | 10 | 12 | 14 | 16 | 17 |
| 6 | 0.05 | | | 1 | 2 | 3 | 5 | 6 | 8 | 10 | 11 | 13 | 14 |
| | 0.02 | | | | 1 | 2 | 3 | 4 | 6 | 7 | 8 | 9 | 11 |
| | 0.01 | | | | 1 | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 9 |
| | 0.10 | | 0 | 2 | 4 | 6 | 8 | 11 | 13 | 15 | 17 | 19 | 21 |
| 7 | 0.05 | | | 1 | 3 | 5 | 6 | 8 | 10 | 12 | 14 | 16 | 18 |
| | 0.02 | | | 0 | 1 | 3 | 4 | 6 | 7 | 9 | 11 | 12 | 14 |
| | 0.01 | | | | 0 | 1 | 3 | 4 | 6 | 7 | 9 | 10 | 12 |
| | 0.10 | | 1 | 3 | 5 | 8 | 10 | 13 | 15 | 18 | 20 | 23 | 26 |
| 8 | 0.05 | | 0 | 2 | 4 | 6 | 7 | 10 | 13 | 15 | 17 | 19 | 22 |
| | 0.02 | | | 0 | 2 | 4 | 6 | 7 | 9 | 11 | 13 | 15 | 17 |
| | 0.01 | | | | 1 | 2 | 4 | 6 | 7 | 9 | 11 | 13 | 15 |
| | 0.10 | | 1 | 3 | 6 | 9 | 12 | 15 | 18 | 21 | 24 | 27 | 30 |
| 9 | 0.05 | | 0 | 2 | 4 | 7 | 10 | 12 | 15 | 17 | 20 | 23 | 26 |
| | 0.02 | | | 1 | 3 | 5 | 7 | 9 | 11 | 14 | 16 | 18 | 21 |
| | 0.01 | | | 0 | 1 | 3 | 5 | 7 | 9 | 11 | 13 | 16 | 18 |
| | 0.10 | | 1 | 4 | 7 | 11 | 14 | 17 | 20 | 24 | 27 | 31 | 34 |
| 10 | 0.05 | | 0 | 3 | 5 | 8 | 11 | 14 | 17 | 20 | 23 | 26 | 29 |
| | 0.02 | | | 1 | 3 | 6 | 8 | 11 | 13 | 16 | 19 | 22 | 24 |
| | 0.01 | | | | | | | | | | | | |

DIRECTIONS FOR USE OF CRITICAL VALUES FOR THE MANN-WHITNEY U-TEST

1. Move across the top horizontal row N1 until you reach the correct column.
2. Proceed down this column until you reach the correct horizontal row corresponding to the N2 group number.
3. There may be up to 4 U values corresponding to the p values listed in the probability column. To be significant at the p value listed, the U value must be equal to or less than the number printed.

Example. N1 = 12, N2 = 9 and U = 3

Since the U value of 3 is less than 18, there is a p value of <0.01.

Therefore, the mean rank values are significantly different .

MANN WHITNEY PROBLEMS (SET 2 page 1)

Use the procedural steps described on the preceding page to determine if significant differences exists between the mean rank values of the following groups.

1. Chris Scanlan, an ex-John Adams research student who then graduated from the U. of Chicago maintained some Planaria flatworms in Aspartame solution and others in pond water. Initial conditioning to mild electrical shock required the following # of trials. Determine between which groups there were significant differences.

| Control | Rank Value | Aspartame | Rank Value |
|---------|---------------|-----------|---------------|
| 73 | | 82 | |
| 61 | | 99 | |
| 84 | | 89 | |
| 83 | | 87 | |
| 71 | | 83 | |
| 89 | | 92 | |

| Control | Rank Value | Aspartame | Rank Value |
|---------|---------------|-----------|---------------|
| 89 | | 110 | |
| 73 | | 115 | |
| 61 | | 83 | |
| 84 | | 120 | |
| 83 | | 140 | |
| 71 | | | |

| Aspartame | Rank Value | Aspartame | Rank Value |
|-----------|---------------|-----------|---------------|
| 92 | | 110 | |
| 82 | | 115 | |
| 99 | | 120 | |
| 87 | | 140 | |
| 89 | | 83 | |
| 83 | | | |

INTERNET-MANN-WHITNEY U TEST

FOR BEHAVIORAL STUDIES

The Mann-Whitney U-Test is an alternative to the Student t-Test for behavioral studies. It is commonly used where the experimenter draws two random samples from the same parent population, subjects each to a different experimental treatment, and compares the two on a single criterion. The U Test may also be used in the situation where independent random samples are drawn from two different parent populations and compared on a single criterion to determine whether the two populations differ.

1. Access the Internet and enter the following address

www.socseistatistics.com/tests/mannwhitney/

2. Select – TAKE ME TO THE CALCULATOR

3. Enter your data points in the vertical boxes.

4. After entering the data, under significant level, SELECT – 0.05
SELECT 0.05

5. Under # of tails – SELECT ONE –TAIL unless informed differently.

6. Next SELECT – CALCULATE U

7. In your lab notebook,(or worksheet) record the NAMES OF GROUPS COMPARED,
THE SMALLER U VALUE, THE P VALUE AND DEGREE OF SIGNIFICANCE IF ANY.

8. Explain mathematically the meaning of the p values.

9. At the bottom of the screen – Select – RUN CALCULATION DETAILS – scan to bottom of next screen.

10. Open Microsoft Excel and generate a bar graph of Mean Rank values (*Pop 1. & Pop 2.*) to represent the Mann Whitney results for one comparison. Include the p values on the graph. Print out your graph.

11. Record Mean Rank Values in your Notebook.

12. If more than two groups, continue comparing the groups in pairs.

EX. POP. 1

26

16

4

12

98

40

EX. POP. 2

128

146

56

46

128

65

LINEAR REGRESSION

References Experimental Biology (Ch. 16), Biology Science (Ch. 5), Fundamental Statistics For Psychology (Ch. 5)

The most common relationship found in research work is of a linear nature. In experimentation it is often found that a high independent variable value is associated with a high dependent variable value and that, a low independent variable value is associated with a low dependent variable value. However, the relationship may be linear, but inversely related. In these cases, a high value on one axis will correspond to a low value on the other axis. In still other cases, there may be no linear relationship between the two factors.

A straight line can represent a true linear relationship if the data are plotted on a graph. Due to individual differences in the experimental organisms or to error, the tabulated results may not reflect a perfectly straight line. Thus, it becomes necessary to calculate the best straight line for representing the data.

Mathematically, it is found that the extreme values tend to move or regress gradually toward the mean value for that group and do not tend to become more and more extreme. The measurement of this trend to regress toward the mean is termed linear regression.

The line equation which results from the linear regression equation provides the best straight line for the entered data. It should be emphasized that the linear regression equation should only be used where there would seem to be a logical relationship or effect between the two variable being considered.

Two formals are combined to produce the formula of the best straight line. The first formula calculates the slope value of the line (b) and the second one (a) includes the slope value with the Y-intercept. On the last line is listed the slope of the best fit formula in the common form.

$$b = \frac{N(\sum XY) - (\sum X)(\sum Y)}{N\sum x^2 - (\sum x)^2}$$

$$a = Y - bx$$

$$Y = bx + a$$

N = number of data points

$\sum X$ = sum of X's

$\sum Y$ = sum of Y's

$\sum XY$ = sum of the products of XY's

\bar{X} = mean value of X

\bar{Y} = mean value of Y

$(\sum X)^2$ = squared value of the sum of the X's

$\sum X^2$ = sum of the squared individual X values

LINEAR AGGRESSION (Page 2)

Thiourea when administered to lab mice is known to decrease thyroxin output, which in turn, reduce basic metabolic rate (BMR). The following results were obtained by Jenny Lackman in her work funded by the American Heart Association.

| Mice Group | O ₂ Consumption cc/10g/10 min | Time of Readings (wks) |
|----------------------------------|---|---------------------------|
| Control | 7.2 | Zero |
| | 7.4 | +3 |
| | 9.2 | 11 |
| | 9.5 | 14 |
| Experimental (0.25% Thiourea) | 9.4 | Zero |
| | 7.6 | +3 |
| | 6.1 | 11 |
| | 5.7 | 14 |

- a. Using standard graph paper, plot these two lines on the same piece of paper. Utilize the entire sheet of graph paper so as to spread out the data.
- b. List the values for the following factors for each group.

| Equation Factor | Control Group | Experimental Group |
|--------------------|------------------|-----------------------|
| N | | |
| $\sum X$ | | |
| $\sum Y$ | | |
| $\sum XY$ | | |
| $(\sum x)^2$ | | |
| \bar{X} | | |
| \bar{Y} | | |
| $\sum x^2$ | | |

- c. Using the linear regression formulas, calculate the formula for the best straight line representing the data for each group of mice. Add these two additional lines to your graph.

CORRELATION COEFFICIENT

REFERENCE: Experimental Biology (Ch. 16), Biology Science (Ch. 5),
Fundamental Statistics For Psychology (Ch.6)

In the previous exercise you used the formulas for linear regression to determine the formula for the best straight line through a given set of data points. More importantly than just determining the best straight line, is comparing how closely the collected data parallels the best straight line. For example, the best straight line may fall directly on top of each of the plotted points, or the points may be widely scattered above and below the line with not even one point on the line.

The term, correlation coefficient, reflects the strength of the relationship between the collected data and the calculated trend. Correlation coefficient values will range from +1, which represents a perfect linear relationship to -1, which indicates a perfect inverse linear relationship between the two variables.

A coefficient of 0 indicates no relationship between the two variables.

The formula to determine correlation coefficient is listed below.

$$r = \frac{\sum(X - \bar{X})(Y - \bar{Y})}{\sqrt{\sum(X - \bar{X})^2 \sum(Y - \bar{Y})^2}}$$

X = x value of each point

\bar{X} = mean value of the X 's

Y = y value of each point

\bar{Y} = mean value of the Y 's

Sample Problem

| | | | | | |
|-------|----------|----|---|---|---|
| Given | Point | A | B | C | D |
| | X values | -2 | 2 | 3 | 5 |
| | Y values | -1 | 0 | 2 | 3 |

Step 1 Determine the mean values of X and Y

$$\bar{X} = 8/4 = 2 \qquad \bar{Y} = 4/4 = 1$$

Step 2 Determine the following values

| $(X - \bar{X})$ | $(X - \bar{X})^2$ | $(Y - \bar{Y})$ | $(Y - \bar{Y})^2$ |
|-----------------|-------------------|-----------------|-------------------|
| $-2 - 2 = -4$ | 16 | $-1 - 1 = -2$ | 4 |
| $2 - 2 = 0$ | 0 | $0 - 1 = -1$ | 1 |
| $3 - 2 = 1$ | 1 | $2 - 1 = 1$ | 1 |
| $5 - 2 = 3$ | 9 | $3 - 1 = 2$ | 4 |
| | <u>26</u> | | <u>10</u> |

Correlation Coefficient Page 2

Step 3 Determine the following values

| $(X - \bar{X})$ | | $(Y - \bar{Y})$ | |
|-----------------|---|-----------------|-----------|
| 4 | * | 2 | = 8 |
| 0 | * | 1 | = 0 |
| 1 | * | 1 | = 1 |
| 3 | * | 2 | = 6 |
| | | | <u>15</u> |

Step 4 Determine the r values

$$r = \frac{\sum(X - \bar{X})(Y - \bar{Y})}{\sqrt{\sum(X - \bar{X})^2 \sum(Y - \bar{Y})^2}} = \frac{15}{\sqrt{(26)(10)}} = \frac{15}{\sqrt{260}} = \frac{15}{16.1} = 0.93$$

Assignment --Determine the Correlation Coefficient values for Jenny's data listed on the Linear Regression Page.

LINEAR REGRESSION AND CORRELATION COEFFICIENT (Using Excel 2011)

1. Select Excel and enter data on a blank spreadsheet. First, add a title to each column and then enter associated values under the appropriate title.
(Save data after it is entered)
2. Using the mouse, HIGHLIGHT the data you wish to represent in the graph. DON'T INCLUDE THE TITLE OF THE COLUMN.
3. Now select CHART tab — Select SCATTER — Select MARKED
4. Single click the mouse on the chart you have just generated. Select Chart Layout — Select Axis Titles — For *horizontal axis*, select TITLE BELOW AXIS and for the *vertical axis* select HORIZONTAL TITLE.
5. Select CHART LAYOUT TAB — Select CHART TITLE — Add a Title
6. Select CHART LAYOUT TAB — LEGEND — Select NO LEGEND
7. Select CHART LAYOUT TAB — click on Trendline (linear)
8. Now double click the mouse on the Trendline that was just generated and this should open the FORMAT TRENDLINE WINDOW — select options in the menu and check “DISPLAY EQUATION ON CHART” and “DISPLAY R-SQUARED VALUE ON CHART”. Close the menu and move the R-squared value & equation away from Trendline.
9. For Error Bars, Select CHART LAYOUT TAB — select ERROR BARS — *Error Bars with Standard Deviation*
10. Now using the same data as the chart you have just generated, generate different styles of graphs. Print out 3 differently formatted graphs.
11. Double click on a data point to open a menu and select marker fill to change the color of the data points (*if you so desire*)
12. Save all of the graphs that have been generated with and without data tables.

Linear Regression and Correlation Coefficient Using Excel (2011)

Indiana State Standards- Statistical Inference--Students use confidence intervals and hypothesis tests, fit curves to data and calculate correlation coefficients.

P.A. 3.1 Compute and use confidence intervals to make estimates.

P.A. 3.2 Understand hypothesis tests of means and differences between means and use them to reach conclusions.

P.A.3.3 Use the principle of least squares to find the curve of best fit for a set of data.

P.A.3.4 Calculate and interpret the correlation of a set of data.

1. Select Excel and enter data on blank spreadsheet. **FIRST** add a Title on each column then
enter associated values under the appropriate title. **(SAVE data after it is entered)**.
2. Using the mouse, **Highlight** (box in) the data you wish represented on the graph.
DON'T include the title of the column.
3. From the green menu bar select **CHART** icon at the top of the screen.
4. Select the actual graph style you want to use. Most of the time you will use a scatter graph or a bar graph. **(Select your choice.)**
5. Click the purple **CHART LAYOUT**.
6. You should add a graph **Title** and the axes **Titles**.
7. Other options can be added at this time i.e. Trend line with formula.
Select **Trendline then select Trendline options** .Select **display equation** and **R²**
8. Click on the graph to make it active .
Type in a value or word on the formula line, such as **Con vs Exp 2 p<0.001**.
Click on return- then drag this box around outside and inside the graph.
9. **Select Chart options-** click legend. **Select hide legend**.
10. Enlarge graph by dragging from the corner.

11. Double click on the graph and change the background color of the graph.
12. Double click any object on the graph and change its color.
13. Double click on a graph axis- change the font size.
14. **SAVE** graph both with and without data table.
15. Use the supplied data and construct different styles of graphs. Print out 3 different formatted graphs.

PIE CHARTS IN EXCEL

PIE CHARTS are limited to a single data series (one row or column of data from the worksheet) and cannot display more complex series of data. The value of each element in the data series is assigned a slice of the pie and all of the slices add up to the total of the data series. However, they are visually appealing and simple to understand.

State Standards—Probability and Statistics—Descriptive Statistics

P.S. 1.1 Create, compare and evaluate different graphic displays of the same data, using histograms, frequency, frequency polygons, cumulative distribution functions, pie graphs, scatterplots, stem and leaf plots and whisker plots. Draw these by hand or use a computer spreadsheet program.

Pie Chart #1 List the data series below on your spreadsheet. On step 1 of the Chart Wizard, select pie chart. and Highlight the data to be included and follow the Wizard to complete a graph. Print out the data table and its graph.

Death rate in the U.S. due to the following causes

Tornados 45.1, Floods 95, Hurricanes 24.7, Lightning 64.2

Pie Chart #2 List the data series below on your spreadsheet.

On step 1 of the Chart Wizard select pie chart. and Highlight the data to be included and follow the Wizard to complete a graph. Print out the data table and the pie chart.

Second PIE CHART Estimated Population of the following countries in 2005 (in millions)

China 1256, India 982, Indonesia 206, United States 315, Brazil 165, Iran 66

Possible Topics to Explore

Behavioral Studies

Effects of Astragalus Root Extract on Memory Retention of Planarian Flatworms 2016

A Study of the Effects of Ellagic Acid on the Rate of Regeneration and Behavior of Planarian Flatworms 2016

A Study of the Effects of Extended Periods of Time and the Regeneration Process on the Retention of a Conditioned Behavior in Planarian Flatworms 2016

The Effects of Ingestion of Coenzyme Q10 on Cognitive Process as administered with Cholesterol to Mimic Alzheimer's Disease 2014

A Study of the Effects of Transplantation of Tissue from Planarian Flatworms Conditioned with Light-Shock Therapy into Naive Planarian Flatworms 2014

Study on the Properties of Electromagnetic Fields and Their Possible Effects on Biological Materials and the Light-shock Memory of Planarian Flatworms 2013

A Study of the Effects of Quercetin Supplementation on the Cognitive Function of Lab Mice 2012

Effect of Light-shock Conditioning on Neurotransmitter Concentrations in Planarian Flatworms. 2013

A Study of the Possible Role of Long Term Ingestion of Supplemental Antioxidants on Cognitive Processes 2012

A Study of the Possible Role of Long Term Ingestion of Juice Plus on Cognitive Function 2012

Investigation of Faunal Diets at Pokagon Village Site to Determine Farming Practices of the Pokagon Band 2010 Intel Finalist

The Effects of Flavonoids on Cognitive Functions in Lab Mice 2011

The Possible Effects of Ginseng (Panax Ginseng) on Reducing Behavioral Responses to Instantaneous Stress in Laboratory Mice... 2005

The Effects of Bioflavonoids on Cognitive Dysfunction in Aged Mice... 2005

The Locomotor Effects of Caffeine Administered at Varying Dosages in Laboratory Mice 2005

Possible Effects on Behavior Due to Elevated Intakes of Aspartame 2006

Possible Effects on Behavioral Changes 55 due to Elevated Intakes of Aspartame, Sucrose and Fructose 1995

**Examining the Relationships Between Circadian Rhythms and Behavioral Changes 2010
Investigating the Possible Correlation Between Sleep Habits, Lifestyle, and Socio-Economic Status in a Representative Population--- 2000**

The Possible Effects of Symbol Perception On Learning and Memory

The Effect of Computer-Aided Instruction on Application of Learned Material 1984

Examining the Relationship Between Melatonin Production in Light/Dark Cycles and Behavior Changes 2000

The Effects of Sidestream Smoke on Behavioral and Learning Patterns in Lab Mice 2001
Investigating the Possible Relationships Between Strenuous Exercise and Learning, Memory, and Behavior in Lab Mice

The Effects of Sidestream Smoke on Behavioral and Learning Patterns in Lab Mice 2001

The Possible Enhancement of Symbol Association and Learning in Laboratory Mice with a Vitamin A Supplemented Diet 1989

The Effectiveness of Computer-Bases Instruction with Emphasis on the Effectiveness of Graphic Details and Animation in Educational Software 1985

The Effects of Aspartame on Learning in Planaria--- 1986

The Role of Long Term Ingestion of the Antioxidant Turmeric on Cognitive Function 2009

The Possible Effects of Coenzyme Q Supplementation on Cognitive Function in Mice
Cierra Strawder 2009

The Effects of Varying Dosages of Caffeine on Cognitive Function in Laboratory Rats 2008

A Comparative Study of the Possible Role of Ingestion of Supplemental Antioxidants on the Physiological and Behavioral Processes of Laboratory Mice 2008

Possible Behavioral Changes Due to 56 Intakes of Valium 2000

Biochemistry and Chemistry Studies

Coupled Advanced Oxidation Processes and Surface-Modified Photocatalysts for the Decolorization of Reactive Azo Dyes 2007

The Relationships Between Light and the Structure of Organic Molecules in Solution 2000

Identifying a Possible Mode of Action for Dietary Fiber on Enzyme Deactivation and Sugar, Starch, and Bile Salt Affinity 2003

An Evaluation of the Antioxidant Properties of Various Vitamin C Solutions And Fruit Juices Containing Vitamin 2001

An Evaluation of the Changes in the Antioxidant Potential of Vitamin C Due to Exposure to Cigarette Smoke 2002

An Analysis of the Ability of Ferrofluids to Attract and Transfer Albumin 2002

An Evaluation of the Antioxidant Properties of Various Vitamin C Anomers

Examining Relationships Between Light, the Nature of Color, and Chemical Molecules 2002

Analyzing the Antioxidant Properties of Astragalus Membranaceus Using the Ferric Reducing Ability of Plasma (FRAP) Assay Related to Aging and Long-Term Efficacy

Construction of a Prototype Microbial Fuel Cell and its Performance with Various Andoes and Biofilms 2005

Measuring the Antioxidant Potential of Flavonoids and Other Compounds Using the FRAP Assay 2008

Directed Evolution of Human Alpha Crystallin for Increased Stability 2012

Engineering

Evaluating the Effects of Modifying Building Design and Arrangement on Pollutant Dispersion in Urban Areas 2016

Utilizing Ducted Turbines to Maximize Energy Generation from the Wind 2016

| | | |
|---|----|---|
| Electricity Generation through and Wetland Sediment in a Fuel Cell | 57 | Bioremediation using Wetland Water |
| | | 2016 |

Utilizing Altered Building Designs and new City planning to produce Improved Building and pedestrian Safety during Times of Extreme Weather Conditions. 2013

The Effects on the Stability of Tall Buildings with Major and Minor Geometric Modifications High Velocity Winds 2014

Using Ultra-capacitors to Increase the Efficiency and Power Output of the Direct Methanol Fuel Cell 2014

A Study of the Effects of Utilizing Heat Generated from Computer Processing, Supplemental Phosphate and Supplemental Nitrate to Enhance Hydrogen Gas Production in Heterocystous, Filamentous Cyanobacteria *Anabaena cylindrica*. 2014

Improving Electrode Kinetics and Operational Lifetime of a Prototype Membrane-less Glucose-Oxygen Physiological Fuel Cell. 2013

Studies of Algae Oil Enhancement Project through Mineral Supplementation with Nitrogen and Phosphate Compounds 2011

The Effects of Hydrophobicity and Microporous Layers in the Membrane Electrode Assembly of a Direct Methanol Fuel Cell to Enable Efficient Operation Using High Concentration Fuel. 2012

Minimizing Methanol Crossover in Direct Methanol fuel Cell Through the Application of Polyaniline Films to the Proton Exchange Membrane. 2012

Efficiency Studies and Design in Wind Energy Generators 2012

Evaluating Factors to Optimize Energy Output From a Microbial Fuel Cell 2000

Characterizing Engineered TiO₂ Nanoparticle Adhesion : Implications for Environmental Transport and Remediation Eric Fein Intel Finalist

The Effects of Different Mediators and Electrode Configurations on the Efficiency and Longevity of a Membrane-less, Enzymatic Biofuel Cell Operating Under Physiological Conditions 2012

Comparing the Effects of Gradient Wind Velocity and Building Pressure Due to Building and Street Structures 2012

Improving Solar Efficiency Through Retrofitting a Flinn Solar Cell with Carbon Nanotube Fiber 2012

| | | |
|--|-----------|-------------------------------------|
| The Effectiveness of Light Diodes as an | 58 | Alternative Light Source for |
| Subwavelength Aperture Array-Based | | Testing 2011 |

Evaluating Pressure, Velocity, and Distance as Factors Affecting Particulate Fallout Rate in an Indraft Wind Tunnel 1999

Construction of a Prototype Microbial Fuel Cell and its Performance with Various Andoes and Biofilms 2005

The Empirical Effect of Various Conjugated Double Bond Systems on the Light Absorption Maimas of Azo Dyes 2006

Managing Gas Evolution and Reduction and Other Factors in the Direst Methanol Fuel Cell to Increase Power Output 2006

Studies in Small-Scale Horizontal Axis Wind Turbine Generators: Their Operations and Efficiencies 2004

Evaluating Factors to Optimize Energy Output From An Aluminum-Air Fuel Cell 2000

An Evaluation of the Relative Effectiveness of Different Types of Passive Air Filters: Implications for Respiratory Health 1986

An Investigation of the Energy Possibilities of Simulated Freshwater Lake Sediment Power Systems 2003

Investigation of Methane Production by Methanogenic Bacteria for Possible Use in an Ecologically Closed Systems

Energy Conservation Associated With Using Rotating Polarized Glass Windows in Houses 1995

Preliminary Studies of Retrofitted Solar Cells 2012

Investigation of the Aerodynamic Characteristics and Airflow Patterns of Blended-Wing-Body Aircraft with Changes of the Inboard and Outboard Wing Panel Sweep Angles 2003

Investigation of Methane Production by Methogenic Bacteria for the Possible Use in an Ecologically Closed System

A Study Of Variables Affecting the Peak Power Output of a Proton Exchange Membrane (PEM) Fuel Cell

The Improvement of Passive Air Filters with the Imbedding of Different Types of Polymer Beads 59

An Evaluation of the Relative Effectiveness of an Electrostatic Precipitator in Comparison to Fiber Filter Media Phase II 1994

The Effects of UV and Photoreactivation when Combined with Various Sunscreens on Intact Bacterial Enzyme Systems in *E.coli* 2001

Studies in Liquid Electrolyte and Polymer Electrolyte Membrane Direct Methanol Fuel Cells 2004

Improving the Design and Efficiency of a novel, Prototype Enzymatic Fuel Cell Using an Enzyme Biocatalyst Embedded on the anode 2006

Investigations of Electrical Power Production From Aquatic Sediment Power Systems

Throwing Away Energy: Evaluating the Efficiency of Saccharification and Fermentation of Cellulose Prior to Usage as a Substrate in an Enzymatic Fuel Cell 2006

Evaluating the Usefulness of Carbon Nanofoam Electrodes and Carbon Nanotube Catalyst Support in the Direct Methanol Fuel Cell 2006

Investigations of Efficiency and Operation of *in situ* Sediment Fuel Cells with Modified Anode Electrodes 2007

Maximizing Voltage and Total Power Output in Both Liquid-Electrolyte and Polymer-Electrolyte Direct-Methanol Fuel Cell 2002

Methanol Concentration and It's Effects on Direct Methanol Fuel Cells Power Output 2005

Investigating the Effects of Various Factors Including Carbon Nanotube Supported Catalysts on the Performance of the Direct Methanol Fuel Cell 2006

Examining Factors to Increase the Energy Output for Aluminum/Air Batteries 2008

The Efficiencies of Various Alcohols in a Direct Methanol Fuel Cell 2008

Ameliorating Obstacles to the Commercial Production of the Direct Methanol Fuel Cell 2006

Minimizing the Effect of Catalyst Poisoning on the Performance of the Direct Methanol Fuel Cell via the Addition of H₂O₂ to the Fuel Stream and Other Factors 2008

Utilizing Electrical Energy as Thermal Energy to Provide a Grid Heating Environment for

***Spirulinato* Increase its Carbohydrate Productivity 2008**

Developing Prototype Models to Increase the Effectiveness of Using Wind Energy to Provide Electrical Power 2008

Construction of a Prototype Enzymatic Fuel cell for Purposes of Oxidizing and Generating Power from Wastewater 2007

Ameliorating Glass Deformation During High Wind Velocities Through Creative Engineering 2008

A Study of Variables Affecting the Peak Energy Output of a Proton Exchange Membrane (PEM) Fuel Cell

Nanostructure Architectures for Photoelectrochemical Solar Cells 2009

Investigation of the Aerodynamic Characteristics and Airflow Patterns of Blended-Wing-Body Aircraft with Changes of the Inboard and Outboard Wing Panel Sweep Angles

Improving Electrode Kinetics and Operational Lifetime of a Prototype Membrane-less Glucose-Oxygen Physiological Fuel Cell 2011

Evaluating the Effectiveness of Retrofitting Direct Methanol Fuel Cells with Carbon Nanotube Fibers 2012

The Effect of High Currents on Ultracapacitor Properties 2011

Environmental Studies

Evaluating the Effects of Modifying Building Design and Arrangement on Pollutant Dispersion in Urban Areas 2016

Examining the Role of *Chlorella vulgaris* in Removing Elevated Concentrations of Nitrogen and Phosphorus 2016

An Evaluation of the Effects of Nutrient Supplementation on the Bioremediation of Hexane-Contaminated Lake Water 2014

In vitro Nanotoxicity Evaluation of Titanium Dioxide on *Dugesia tigrina* Planarian Flatworm 2014

A Study of Xylene Bioremediation Nitrogen and Phosphorus Compounds 61 Potential of Carbon Fiber Biofilms with and Dissolved Oxygen 2012

A Comparative Study of Two different Methods of Removing Xylene Contamination from Ground Water Through Bioremediation 2010

A Study of Intrinsic Biodegradation of Benzene Contaminated Soil with Artificial Enhancement 2010

Evaluating the Effect of Nutrient Supplementation on Xylene Contaminated Groundwater Containing a Permeable Reactive Barrier and Determining DNA Sequences of the Microorganisms most Greatly Effecting the Xylene Concentration 2012

Impact of Rusty Crayfish (*Orconectes Rusticus*) Invasion and Sediment Depth on Seed Bank Germination from Lake Sediments. 2012

Possible Effects of Graphene Oxide Ingestion on Behavior and Physiological Processes in *Daphnia Magna* Triops. 2012

Studies in the Development of Enhancing Bioremediation Processes of Carbon Tetrachloride Contaminated Soil 2001

Characterizing Engineered TiO₂ Nanoparticle Adhesion : Implications for Environmental Transport and Remediation 2011-2012 Intel Finalist

The Effects of Altering the Nitrogen and Phosphorus Concentrations In Simulated Lake Communities 2001

An Investigation of the Energy Possibilities of Simulated Freshwater Lake Sediment Power Systems 2003

Investigation of Methane Production by Methanogenic Bacteria for Possible Use in an Ecologically Closed System

Energy Conservation Associated With Using Rotating Polarized Glass Windows in Houses

A Mathematical Comparison of the Toxicity of Environmental Pollutants Using Brine Shrimp (*Artemia Salina*), 1998

Ecotoxicological Effects of “ Green Chemistry” Solvents on *Daphnia Magna* 2006

A Study of Xylene Bioremediation Potential of Carbon Fiber Biofilms with Nitrogen and Phosphorus Compounds and Dissolved Oxygen 2012

An Evaluation of Possible Compensatory Mechanisms of *Myriophyllum Spicatum* Compared to *Elodea Canadensis* and *Cabomba Carolinianna*

Developing Techniques and Procedures for Using Brine Shrimp, *Artemia Salina*, as a Test Organism to Measure the Ecotoxic 62 Effects of Groundwater Pollution 2001

Hydrogen-Based, Hollow Fiber Membrane Biofilm Reactor for Reduction of Bromate 2005

Studies of a Simulated Reclamation Project for the Removal of Excess Nitrogen and Phosphorous Compounds From Wetland Water 2005

Studies of Enhancing the Remediation of Benzene Contaminated Ground Water 2005

Ecotoxicological Effects of Ionic Liquids on Aquatic Life 2006

Examining the Role of Different Aquatic Plants in Removing Elevated Concentrations of Nitrogen and Phosphorus Simulated Lakes 2006

The Efficiency of the Bioremediation Process in Xylene Contaminated Soil 2008

The Effects of Different Mediators and Electrode Configurations on the Efficiency and Longevity of a Membrane-less, Enzymatic Biofuel Cell Operating Under Physiological Conditions 2012

Measuring and Optimizing the Antioxidant Potential of administered Flavonoids and of Other Compounds 2005

Possible Effects of Creatine Phosphate Supplementation on the Process of Strenuous exercise and Physical Fitness 2011

The Possible Effects of Elevated Lipid Diets on the Colon Contents with Implication for Atherosclerosis 1995

The Possible Effects of Elevated Lipid Diets on the Biochemistry of the Colon Contents with Implications for Atherosclerosis 1995

Analyzing the Antioxidant Properties of Astragalus membranaceus Using the Ferric Reducing Ability of Plasma (FRAP) Assay Related to Aging and Long-Term Efficacy 2002

An Evaluation of the Relative Effectiveness of Different Types of Passive Air Filters: Implications for Respiratory Health 1986

The Effects of Swimming and Treadmill Exercise on the Physiological Response of Strenuously Exercised Young Mice 2000
64

A possible Mode of Action of Voltaren Ingestion on Gastrointestinal Responses and Behavioral Changes 1993

The Development and Refinement of Invertebrate Assays for Identification of Possible Teratogens

Development of a Computer Model to Predict Potential Hazards Associated with Accidental Pollutant Explosions: Phase 2

The Possible Relationship Between Estrogen Replacement Therapy (ERT) and Cardiovascular Diseases

Effects of Ascorbic Acid Supplementation on the Immune Response of Young, Female Mice

The Possible Relationship Between Postmenopausal Hormone Therapy and Cardiovascular Disease

Investigating the Possible Correlation Between Sleep Habits, Lifestyle, and Socio-Economic Status In a Representative Population 2000

Investigating Improvements in Cognitive Function Due to Ingestion of Juice Plus TM 2006

A Comparative Study of the Possible Role of Long term Ingestion of Supplemental Antioxidants on Physiological Behavioral Processes, Longevity 2006-2008

The Antioxidant Properties of Various Vitamin C And Fruit Juices Containing Vitamin C
2001

An Evaluation of the Changes in the Antioxidant Potential of Vitamin C Due to Exposure to Cigarette Smoke 2002

An Analysis of the Ability of Ferrofluids to Attract and Transfer Albumin 2002

An Evaluation of the Antioxidant Properties of Various Vitamin C Anomers

The Roles of Diet and Antibiotic Therapy on the Reproduction and Dissemination of Candida albicans

The Possible Role of a Restricted Diet and an Elevated Cholesterol Diet on Some Atherosclerotic Factors 1984

A Comparative Study on the Effects of Allergic Reactions in Laboratory Mice 65 **Chlortrimeton and Solumedrol On** 1983

The Effects of UV and Photoreactivation When Combined With Various Sunscreens on Intact Bacterial Enzyme Systems in E. coli 2001

Studies in the Possible Enhancement of Immune Response through the Ingestion of Astragalus membranaceus Extracts

Possible Effects of Creatine Phosphate Supplementation on the Process Of Strenuous Exercise and Physical Fitness 2004

Possible Relationships Between Calcium Absorption and Bone Development 2004

The Possible Effects of Passive Cigarette Smoke Exposure on Erythrocyte, Hemoglobin Levels, White Blood Cell Counts, IgG Concentrates, and the Resting Metabolic Rate of Laboratory Mice

The Roles of Diet and Antibiotic Therapy on the Reproduction and Dissemination of Canida albicans

Studies of the Possible Enhancement of Immune Response Through the Ingestion of Coenzyme Q10 and Creatine Phosphate 2002

Possible Relationships Between Weight Reduction Plans: Fat Trapper Plus Carbo-Blaster on Physiological Changes 2002

Analyzing the Antioxidant Properties of Coenzyme Q10 Using the Ferric Reducing Ability of Plasma (FRAP) Assay: Implications for the Enhancement of Immune Response 2003

The Improvement of Passive Air Filters with the Imbedding of Different Types of Polymer Beads

The Antioxidant Efficacy of Ginkgo biloba Compared with Vitamin C, Vitamin E, Vitamin A and B-carotene, Using the Ferric Reducing Ability Plasma (FRAP) Assay

Response Through the Ingestion of

Certain Probiotic Strains in Nature's Biotics 2005

**Investigating the Possible Interaction Between Immune Response and Strenuous Exercise
2000**

The Antioxidant Potential Of Dietary Fiber 2005

**The Possible Effects of Antioxidant Supplementation on the Development of Coronary Heart
Disease 2008**

**A Study of the Role of Resveratrol on the Maturation of Healthy Rats on a Standard Diet
2008**

**The Effects of Creatine Phosphate on the Physical Performance of Physically Active Subjects
2008**

**Possible Effects of Creatine Supplementation on the Process of Strenuous Exercise and
Physical Fitness 2000**

**The Effects of Increased Dietary Protein and Dietary Pyridoxine and the Pathogenesis of
Atherosclerosis 1979-1980**

**A comparison of the Allergy-Lessening Effects of Different Anti-Allergic Drugs on
Laboratory Mice 1986**

**The Possible Concomitant Action of Strenuous Exercise and Ascorbic Acid on Enhancing the
Immune Response of Laboratory Mice 1987**

**An Investigation of the Role of Creatine Supplementation on Strenuous Exercise in
Laboratory Mice**

**Investigating the Possible Effects of Anabolic Steroids on Physiological and Morphological
Changes in Laboratory Mice 1984**

**The Cholesterol- Lowering Properties of Ethanol on Mice Fed a High Saturated Fat Diet
1987**

Possible Mode of Action of Hypothyroidism on Atherogenesis in Laboratory Mice 1978-1979

**An Evaluation of Strenuous Exercise Performance with Recommended Vitamin and Mineral
Supplementation 1986**

**A Comparative Study on the Effects of Chlortrimeton and Solumedrol on Allergic Reactions
in Laboratory Mice 1983**

**The Effects of Amino Acid 68 Supplementation on the regeneration
Rate of Planarian Flatworms 2013**

Micro-organisms (bacteria)

**A Study of the Effects of Indole on the Survival of *Bifidobacteria* and their Viability for Use
in Commercial Probiotic Yogurts 2014**

The Effects of UV Exposure on MIC Values and Enzyme Reaction Rates in Staphylococcus Epidermis

The Development of a Tetracycline-Resistant Strain of Escherichia Coli 1993

The Effects of UV Exposure on MIC Values and Enzyme Reaction Rates in *Staphylococcus epidermis* 2001

The Effect of Hydrogen Peroxide on Microorganisms Known to Inhabit Hydrophilic Contact Lens

The Roles of Diet and Antibiotic Therapy on the Reproduction And Dissemination of Candida albicans

Effects on the Immune Responses of a Diet Supplemented With Food Additive 1992

Investigating the Possible Interaction Between Immune Response and Strenuous Exercise
2000

Identifying a Possible Mode of Action for Dietary Fiber on Enzyme Deactivation and Sugar, Starch, and Bile Salt Affinity 2003

Evaluating Possible Relationships Among Food Knowledge/Food Preference and CHD Risk Factors 1995

Possible Behavioral Changes Due to Intakes of Valium 2000

Possible Anti-Arteriosclerotic Properties of Acetylsalicylic Acid an Analysis of Etiological Factors

Aerobic Exercise: an Aid Toward Improved Health for Diabetics 1988

The Effects of Audiogenic Stress Upon the Immune System of Laboratory Mice

The Effect of Changing the Ditch Angle, Blade Length, and Configuration of Prototype Rotors on Wind Machine Efficiency

Physiological and Biochemical Effects of ⁶⁹ **Bovine Colostrum Supplementation on Laboratory Mice**

The Immune Response of Young Mice To Penicillin Resistant Bacteria 1987

The Improvement of Passive Air Filters With the Imbedding of Charged Polymer Beads: Implications for Respiratory Health

The Effects of Magnetic Fields on *Escherichia Coli* and *Staphylococcus Epidermis* Resistance to Baytril (Enrofloxacin) and Tetracycline 1999

The effects of Ultraviolet Radiation and Photoreaction Repair Mechanisms on the Bacterial Enzymes Pectinase and 2001

Plants and Botany Studies

Optimizing Factors for Hydrogen Evolution in a Bioreactor Containing Vanadium-Nitrogenase *Anabaena Variabilis* ATCC 29413 2007

An Evaluation of Possible Compensatory Mechanisms of *Myriophyllum Spicatum* Compared to *Elodea Canadensis* and *Cabomba Carolinianna*

The Possible Effects of Nutrient Reduction on Plant Compensation After Herbivory

The Effects of Altering the Nitrogen and phosphorus Concentrations In Simulated Lake Communities- 2001

The Possible Effects of Stress on Seed Germination and Seedling Growth

Computer Technology

Using an Artificial Neural network to Classify Handwritten Digits 2016

The Classification of Basal Cell Carcinoma from Benign Tumors Using Promising Machine Learning Models 2016

Identifying Pollen Cells Using Computer Analysis: An Aid for Allergists, Immunologists and Other Health Care Specialists 2014

Examining the Effects of Computer-Controlled Robotics on the Aluminum-Air Fuel Cell 2004

Developing a Computer-Robotic Microscope System to Analyze and Identify Blood Cell Disorders 2005

Developing Software for Image 70 **Recognition of Pollen Samples** 2008

Evaluating the Effectiveness of Using an Artificial Neural Network to Filter Spam Email 2009

Development of a Computer Program to Predict Possible Allergic Reactions for Athletes on Cross Country Courses 1993

The Effect of Computer-Aided Instruction on Application of Learned Material 1985

Development of a Computer Model to Predict Potential Hazards Associated with Accidental Pollutant Explosions

Development of a Computer Program to Evaluate Total Physical Fitness in Lab Mice 2008

References for an Open-ended Inquiry Based Curriculum

It is almost unanimous among science educators that the best Inquiry Based curriculum materials (BSCS, ESCS, Chem Study, Project Physics) were originally developed by the Curriculum Study Committees. Fortunately, some of these books have continued to be updated and printed. It is also most fortunate that a low cost internet site has these used books available in a limited number.

This site, www.bestbooks.com, which is essentially a very low for-profit organization, directs their profits into improving educational opportunities for students in third world countries. We have found their books (used/new) to be in excellent condition and to be sold for a small fraction of their newly published price.

Considerable interest has also developed among teachers using the inquiry approach to science teaching as incorporated in the International Baccalaureate (IB) Curriculum. In brief, the IB Curriculum focuses more on science as a process rather than science as a collection of concepts and facts. However, an understanding of the concepts is required in order for the student to propose reasonable hypotheses and to appropriately discuss the results of the investigation. While the IB Organization does not specially endorse any one textbook, individual teachers and seminar leaders tend to select many activities from the Curriculum Study Committee texts listed below.

In addition to endorsing the Curriculum Study Committee's curricular materials of 30 years ago, IB also strongly recommends the use of interface equipment such as Vernier develops. The easiest method for introducing simulated open-ended lab investigations is with computer-interface equipment using various probes. Here the students can extensively alter most variables. Even with just one demonstration unit, different pairs of students can easily get different data by altering the variable.

Biology/Physiology

BSCS Biology: A Molecular Approach by Jon Greenberg, Revision Editor
Pub. Glencoe McGraw-Hill, New York, New York ISBN 0-538-69039-9
(also available at BetterWorld.com)

BSCS Biology: An Ecological Approach by William J. Csirney
Pub. Kendall Hunt Pub. Co. Dubuque, IA ISBN 0840396597
(also available at BetterWorld.com)

Biology Labs That Work: The Best of How-To-Do-It, Several Volumes edited by
Suzanne Black, Randy Moore and Heidi Haugen ISBN 0-941212-28-9
Pub. National Association of Biology Teachers, Reston, Virginia 2000-2005

Investigating Biology through Inquiry by Robyn Johnson, Scott Holman and Dan
Holmquist Pub. Vernier Software and Technology, Beaverton, OR

Biology with Computers, by David Masterman and Scott Holman
Pub. Vernier Software and Technology, Beaverton, OR

Human Physiology with Vernier by Diana Gordon and Steve Gordon
Pub. Vernier Software and Technology, Beaverton, OR

Water Quality with Computers, by Robyn Johnson, Scott Holman and Dan
Holmquist Pub. Vernier Software and Technology, Beaverton, OR

Water Quality with Computers, by Robyn Johnson, Scott Holman and Dan
Holmquist Pub. Vernier Software and Technology, Beaverton, OR

The American Biology Teacher is a monthly journal published by The National
Association of Biology Teachers, 12030 Sunrise Valley Drive, #110, Reston, VA
20191

Senior Biology 1 by Tracey Greenwood, Lyn Shepherd and Richard Allan
Biozone International, Hamilton, New Zealand 2006 ISBN 1-877329-66-5

Senior Biology 2 by Tracey Greenwood, Lyn Shepherd and Richard Allan
Biozone International, Hamilton, New Zealand 2007 ISBN 1-877329-68-1

Biology Lab Book, by David Masterman and Scott Holman Pub. Vernier Software and
Technology, Beaverton, OR

Agricultural Science Lab Book, by David Masterman and Scott Holman
Pub. Vernier Software and Technology, Beaverton, OR

Environmental Science Lab Book by David Masterman and Scott Holman
Pub. Vernier Software and Technology, Beaverton, OR

Free sample biology labs are available from Vernier at www.vernier.com/labs

Earth Science/Environmental Science

Investigating Earth Science ESCS by ESCS Staff Pub. Kendall Hunt Pub. Co.
Dubuque, IA ISBN 0757501044
(also available at BetterWorld.com)

ESCS Science Tracs Connecting Science and Literacy Investigating Earth Materials by BSCS Staff
Pub. Kendall Hunt Pub. Co. Dubuque, IA ISBN 0757516041
(also available at BetterWorld.com)

Prentice Hall Science Explorer: Earth Science
By Michael Padilla, Ioamis Micoulis and Martha Cyr Pub. Prentice Hall ISBN-
10:0130540714

Earth Science with Computers, by Robyn Johnson, Grethen DeMoss and Richard Sorensen Pub. Vernier Software and Technology, Beaverton, OR

Earth Science Lab Book by Robyn Johnson, Scott Holman and Dan Holmquist Pub.
Vernier Software and Technology, Beaverton, OR

Water Quality with Computers, by Robyn Johnson, Scott Holman and Dan Holmquist Pub. Vernier Software and Technology, Beaverton, OR

Free sample earth science labs are available from Vernier at www.vernier.com/labs

Chemistry

A Portfolio of Investigations for IB Chemistry (paperback), by John Green,
IBID Press, ISBN -10-1876659122 (also available at BetterWorld.com)

Further Investigations in IB Chemistry (paperback) by John Green and D. Greig,
IBID Press, ISBN-10-1876659246 (also available at www.BetterWorld.com)

Water Quality with Computers, by Robyn Johnson, Scott Holman and Dan Holmquist Pub. Vernier Software and Technology, Beaverton, OR

Chemistry Lab Book

by Sally A. Vonderbrink, Don Volz, Dan Holmquist, John Gastineau, Greg Dodd Pub.
Vernier Software and Technology, Beaverton, OR

Chemistry Inquiry Book by Sally A. Vonderbrink, Don Volz, Dan Holmquist, John
Gastineau, Greg Dodd Pub. Vernier Software and Technology, Beaverton, OR

Advanced Chemistry with Vernier: Experiments for AP and IB Chemistry

by Sally A. Vonderbrink, Don Volz, Dan Holmquist, John Gastineau, Greg Dodd Pub.
Vernier Software and Technology, Beaverton, OR

Free sample chemistry labs are available from Vernier at www.vernier.com/labs

Journal of Chemical Education is published by The American Chemical Society,
P.O. Box 1257, Bellmawr, NJ 08099-1267 www.jchemed@egpp.com

Physics

Physics: International Baccalaureate

By G. Paul Ruth and Greg Ketor
Pub IBID Press 2001
ISBN – 10-1876659351

Lab Manual to Accompany Applied Physics: Concepts Into Practice

By Gregory Romine Pub. by Prentice Hall ISBN 9780130870643
(also available at BetterWorld.com)

Physics for Scientists and Engineers IB

By Raymond A. Serway and John W. Jewett Pub. by Brooks Cole Pub.
ISBN 0534408427

Physics with Computers, by Kenneth Appel, John Gastineau, David Vernier, et. al.
Pub. Vernier Software and Technology, Beaverton, OR

Physical Science with Computers, by Donald Volz and Sandy Sapatka
Pub. Vernier Software and Technology, Beaverton, OR

Physics Lab Book by Donald Volz and Sandy Sapatka Pub. Vernier Software and
Technology, Beaverton, OR

Advanced Physics: Mechanics by Donald Volz and Sandy Sapatka
Pub. Vernier Software and Technology, Beaverton, OR

Beyond Mechanics Donald Volz and Sandy Sapatka
Pub. Vernier Software and Technology, Beaverton, OR

Free sample physics labs are available from Vernier at www.vernier.com/labs
The IBID Press publishes primarily for the International Baccalaureate Program
Their address is
IBID Press ,
P.O. Box 396 Washroonga N.S.W.
Sydney, Australia
Email address ib@pronin.com.au

Engineering

Engineering Projects Book by Donald Volz and Sandy Sapatka Pub. Vernier
Software and Technology, Beaverton, OR

STEM 2 Lab Books by Donald Volz and Sandy Sapatka Pub. Vernier Software and
Technology, Beaverton, OR

Techdirections A Prakken Pub. Magazine –Free publication, Ann Arbor, MI
www.napubco.com

Exploring Your Future 4th ed by Oakes, Leone, Gomez Great Lakes Pub
ISBN 188101878-4

Intro to Engineering: Modeling and Problem Solving by Jay Brockman Wiley Pub.
ISBN 978-0471-43160-2

On Line Sources

Selected Online Resources for Teaching about Alternative Energy by Lynn Diener in
Journal of Chemical Education July 2012
Twenty three on line sources.

www.engineergirl.org
www.careercornerstone.org
www.discoverengineering.org
www.asee.org
www.technologystudent.com
[//getea.org/lessons.html](http://getea.org/lessons.html)
pltw.org

APPENDIX OF USEFUL RELATED TOPICS

PowerPoint Evaluation Matrix

Name _____

2 1 0
Good Fair Poor

A. Introduction and Overall Physical Aspects

1. Good contrast of Font and Background Template _____
2. Font SIZE (at least 30 point for Title
and at least 20 point for sub-points) (12 point NOT acceptable) _____
3. State the Title of the Presentation _____
4. State the research title in COMMON language _____
5. Discuss the importance of the investigation _____
6. Steps limited to phrases or bullets (no sentences) _____
7. Use of DIAGRAMS/Flowcharts/Pics in Introduction _____
8. LIMIT of nine lines/bullets per screen _____
9. Brief descriptions of at least TWO other researcher's work _____

B. Methods and Materials

1. Steps limited to phrases or bullets (no sentences) _____
2. Steps interspersed with pictures(2 to 4 steps+pic/screen) _____
(1,2,3+Picture—4,5,6+ picture---7,8,9 +picture)

C. Results and Discussion

1. Repeat associated M and M Step/Picture with graph _____
2. Independent variables (Experimental groups identified)_____
3. Use of Stats and/Tests of significance _____

D. Conclusions

1. Limit of three phrases or bullets/screen _____

E. Possible future investigation which could or should be performed

F. Literature Cited

TWO journal articles with sources.(one screen only)

____ _
____ _

Overall Presentation

1. Did NOT read directly from screen (casual glances are OK) ____ _

2. Could be easily heard

____ _

3. Answered questions in a reasonable and appropriate manner ____ _

(If unsure of answer, state-I did not investigate that area directly,
however, based on the work I did,

I would predict the following would happen_____)

** Never say,"I have no idea what would happen."

Suggestions by evaluator

Siemens Science Talent Search

The Siemens Foundation was launched in 1998 to award scholarships to outstanding science and mathematics students who had scored high in the Advanced Placement Program. In 1999 the program was enlarged to recognize students who had conducted exemplary science and mathematics research.

In recognizing the “best and brightest” students in the sciences, mathematics and technology, the Siemens Foundation seeks to;

- promote excellence in science, mathematics and technology education in American high schools.
- increase the available pool of highly skilled scientists and engineers in order to meet the manufacturing, service, engineering and R&D needs of the globally competitive economy.
- enhance science and technology educational policy through collaborations among Siemens and the nation’s secondary schools, top higher education institutions and other science-rich organizations.
- raise awareness among various U.S. audiences about the need for greater emphasis on mathematics and the hard sciences.

Each year U. S. students, who have conducted investigations, compete for monetary prizes leading up to the top scholarship of \$100,000. There are regional as well as national scholarship winners.

Many of the projects developed by the students each year show great promise for future humanitarian use. The Siemens Competition also indicates that many exemplary U.S. students are starting on the science and math track at an earlier age.

In general, the steps leading up to the final competition in Washington, D.C. are these;

1. Access the Siemens/ Westinghouse Competition website before beginning. www.siemens-foundation.org
2. An individual student or a team completes a science or mathematics investigation, usually before the end of the summer of their junior year.
3. A research paper in normal science journal format is written.
4. The Siemens entry forms and research paper **MUST** be received by Sept. 30, 2013. Siemens Westinghouse Competition Educational Testing Service PO Box 6730 Princeton, NJ 08541
5. Semifinalists will be announced on Oct. 18, 2013.
6. Region Three Competition will be held at Notre Dame on November 8 and 9, 2013

National competition will be held at George Washington University, Washington,DC Dec. 6-10, 2013



**Eric Fein- Properties of Nanoparticles
-Regional Semifinalist**

Indiana Science Talent Search

The Indiana Academy of Science is pleased to invite all **OUTSTANDING high SCHOOL STUDENTS IN THE CLASSES OF 2017 and 2018** to enter the **2017 Indiana Academy of Science Talent Search**, a competition aimed at identifying students with the potential to become creative scientists, and mathematicians. The Indiana Academy of Science Talent Search is sponsored and funded by the IAS Youth Activities Committee.

Applications were due to the Director no later than **May 1, 2017**. (June 1, 2016) The initial screening of applicants' research papers to identify the state finalists is performed by the IAS Talent Search Committee, a group of scientists and mathematicians representing colleges and universities in Indiana. Notification of finalists will occur during a week in September 2017. Twelve finalists and their teacher-sponsor will be guests of the IAS Talent Search Committee at the **IAS Talent Search Honors Weekend** at Indiana University Kokomo on approximately the second Friday and Saturday of **OCTOBER 2017**.

AWARDS:

The **two first place winners** and their teacher-sponsors will be awarded trips to the American Junior Academy of Science, sponsored by the National Association of Academies of Science and the American Junior Academy of Science, scheduled for February 15-19, 2018 in Chicago, IL.

The **second place winner** will receive a \$600 scholarship (to be used at an Indiana college or university)

The **third place winner** will receive a \$300 scholarship (to be used at an Indiana college or university)

Each of the twelve finalists will receive a \$50 stipend to help defray the cost of his/her research project.

Only individual projects will be considered, and all displays must meet IAS display requirements (projects must not exceed a height of 9-ft from the floor, a width of 4-ft, or a depth of 2.5ft).

Student finalists and their teacher-sponsors must be present on both Friday and Saturday in order to compete for awards. Teacher-sponsors must be members of the Indiana Academy of Science.

In general, the steps leading up to the final competition at IN are these;

1. Access the following site for specifics www.indianaacademyofscience.org/ Applications must be received no later than June 1, 2017 by Indiana Science Talent Search Finalists will be announced by approximately Sept. 8, 2017.

Dr. Lynn Thomas, Director
University of Marian
3200 Cold Spring Road
Indianapolis, IN 46222



Kyle Rice-Kevin Tidmarsh-Moshe Friedland-Susan Nace-Eric-Fei

Indiana Science Talent Search Finalists

Regenon Science Talent Search

The Intel (Regenon) Science Talent Search School Award recognizes excellence in teaching and school support of individual student research. It was created in 1942 by Science Service as a means for encouraging talented high school students to pursue a career in science or engineering. Now in its seventh decade, the STS has recognized almost 3,000 finalists with nearly \$3 million in scholarships. In 1999, Intel Corporation was named the title sponsor of the Science Talent Search. Since assuming this role Intel has significantly increased the annual awards and scholarships from \$207,000 to \$1,250,000.

Three hundred Semifinalists and their schools will receive matching awards of \$1000. Every accredited college and university in the United States will receive written notification of the Semifinalists. From these 300 Semifinalists, 40 students will be chosen to attend the six-day Science Talent Institute in Washington, D.C., where students exhibit their research at the National Academy of Science and compete for the prizes ranging from \$5000 to \$100,000.

In addition, the school of each Semifinalist will receive an award of \$1000 to further support research in their school in science, engineering or mathematics. Projects may only be entered as an individual investigation, and there are several restrictions governing the types of investigations which will be accepted for consideration for scholarships. Students are strongly encouraged to access the STS website for instructions prior to beginning the investigation.

www.societyforscience.org/sts

In general, the steps leading up to the final competition in Washington, D.C. are these;

1. Access the Intel Science Talent Search website before beginning your investigation. www.sciserv.org
 2. An Individual student completes a science or mathematics investigation, usually before the end of the summer of their junior year in school.
 3. A research paper in normal science journal format is written.
 4. The Intel entry forms are completed.
 5. The Intel entry forms and research paper MUST be received by early Nov. 9.
- Finalists will be announced by approximately Jan. 22, 2018 deadline.

Society for Science & the Public
Re; Intel STS Transcripts
1719 N Street, NW
Washington, DC 20036-2888

7. Semifinalists will be announced in early Jan. 2017 (see on web site)



8. National competition will be held in Washington, DC, early March 2017

Frieda Fein- John Adams Intel Finalist

Indiana Science and Humanities Symposium

Students who have completed a research project and are enrolled in high school level courses for the current school year may apply for the Indiana Junior Science and Humanities Symposium. **Deadline is approximately Dec. 31, 2017. The competition will be at Indiana State University on approximately March 10, 2017.** Indiana State University and the branches of the military units of the United States share the funding so that a teacher and up to eight students per school may attend for no cost, other than their own transportation. Student observers may be in the eight-student delegation as a learning and preparation activity for future competitions. Students have a specific format required for the papers.

Guidelines are available from the website www.indstate.edu/scied or are posted on the site of the national sponsor, the Academy of Applied Science. See www.jshs.org. Pre-registration and nomination forms are also available on the first web site (Indiana State) and must be completed by the schedule posted on that web page. Students selected for the competition make an oral presentation by slides, transparencies, or PowerPoint. No display board is used. After the oral presentation, the judges and audience may ask questions. Other activities include lab visits, field trips, team building tasks, guest speakers, and formal banquets. The top five students will earn trips to the National Junior Science and Humanities Symposium and the top three students will earn substantial scholarships. The national site rotates each year. The faculty sponsor from Indiana State takes the five students.

Secondary teachers who accompany their students must have their financial considerations covered by their school or other external source.

For further information, please contact Dr. David Grabowski, InJSHS, Department of Life Sciences, 812-237-2395, at Indiana State University, Terre Haute, IN 47809. (This competition may be moved to St Mary's of the Woods-Terre Haute, IN) You may also contact them by email at jshs@indstate.edu or by fax at 812-237-3002. Web address www.instate.edu/jshs



Marian High School

Indiana Junior Academy of Science Fall Meeting

by Tina Gilliland, Director

The Indiana Academy of Science is pleased to invite all Indiana High Schools to participate in the Indiana Junior Academy of Science (IJAS) fall meeting & competition. IJAS will be held on **approximately the first Monday of November 2017** on the Indiana University Bloomington campus. The fall meeting provides several special opportunities for student participation. Teachers may bring students to compete in the Outstanding Scientist, Research Competition, Problem Solving Exam, Issues Presentation (Debate), Science Olympiad Activity, and take part in the various science-based tours and demonstrations.

For more information about these competitions and the tours, please visit our website at:

<http://www.indiana.edu/~college/science/academy>

In addition to the traditional awards, this year we will award cash prizes to the student finalists in the OJS & the Research competition. Based on each school's participation in the various competitions, we will crown the "Most Outstanding School" and the winning team will take home a large trophy for their school display case.

Registration Information - The deadline for registering your school is approximately **October 1, 2017**.

- ☐ Fill-out and submit the online school registration form by October 1, 2017. Note: If you have problems submitting this online, you may print it and mail it in.
- ☐ Mail your \$15 school membership fee and registration fees (\$8.00 per person). Make checks payable to the Indiana Academy of Science.
- ☐ If you have students participating in any of the competitions, be sure to get them registered with the appropriate paperwork. The OJS and the Research application forms are located on our website. They are forms that you print and mail in along with abstracts, etc.

The School Registration Form has fields available for you to register your Issues, Problem Solving, & Olympiad delegates.

Contact Information

Mrs. Tina Gilliland, Director
Indiana Junior Academy of Science
College of Arts & Sciences
Kirkwood Hall, Room 202
Phone: (812) 855-5397;
Email: mgillila@indiana.edu

<http://www.indiana.edu/~college/science/academy>



Indiana Research Grants

The Indiana Academy of Science awards up to **\$300** to each individual student applicant working under the supervision of a secondary school teacher **to support independent projects** demonstrating scientific merit, innovation, ambition, and positive impact on the student's education and future potential. The Academy hopes these awards will encourage students and their teachers to pursue ambitious novel projects and experience the challenges and thrills of "hands on" scientific research. The projects students complete with this support should not only enrich their experience with science but also enhance their competitiveness and aspirations as they evaluate and plan their options for college and beyond.

Program Requirements: For program details and application materials go to the IAS website at www.indianaacademyofscience.org (click on "Research Grants"). Applications must satisfy the following criteria:

- ☐ **eligibility** as a secondary school student (grades 9-12) for the duration of the project with a supervising teacher at a school affiliated with the Indiana Academy of Sciences
- ☐ a **proposal** written by the student and supported by the supervising teacher describing the proposed project; the applicant's background and motivating interest; a description detailing the means of and timetable for implementation, a budget describing expenses related to the project and procedural steps.

Deadline: All applications must be postmarked by late October **1, 2017** and sent to the current chair of the Research Grants Committee of the Indiana Academy of Science. Notification of awards should occur approximately one month later.

Recently supported Projects included;

- ☐ The Effects of Contraceptive Hormones on an Aquatic Food Chain
- ☐ A Comparative Study of Nitrates and Phosphates Due to Blue River Watershed Runoff
- ☐ An Observational Study of the Repellant Effects of Coffee Solutions on *Gastropoda pulmonata*
- ☐ Developing a Computer-Robotic Microscope System to Analyze Blood Cell Disorders
- ☐ Bioremediation of Benzene Contaminated Soil with Artificial Enhancement
- ☐ The Effect of Hym-346 on Head Regeneration in *Hydra vulgaris*
- ☐ Evaluating Usefulness of a Carbon Nano-foam Flow Field Plate in Direct Methanol Fuel Cells

Contacts and Information. Information about this program may be obtained from:

☐ Dr. Alice Heikens, Chair, IAS Research Grants Committee;

Franklin College, Biology Department

101 Branign Blvd.

Franklin, IN 46131

(317)738-8302; aheikens@franklincollege.edu.

Please contact Dr. Heikens with questions, preferably by email.

[www.indianaacademyofscience.org/htmlfiles/Research grants Pages](http://www.indianaacademyofscience.org/htmlfiles/Research%20grants%20Pages)



Kyle Rice- John Adams Grant Recipient

Please refer additional questions about
the syllabus to:

Nevin Longenecker
John Adams High School
808 S. Twyckenham Drive
South Bend, IN 46615

Collin Daniels-Graphics Editor
John Adams High School
808 S. Twyckenham Drive
South Bend, IN 46615