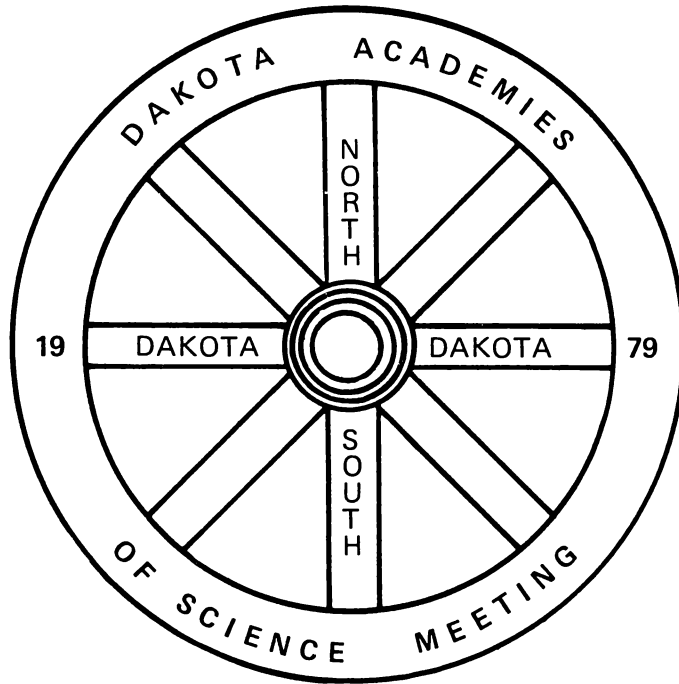


# Proceedings of the NORTH DAKOTA Academy of Science



April, 1979

Volume 33

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PROCEEDINGS  
of the  
NORTH DAKOTA  
ACADEMY OF SCIENCE

Volume 33

April 1979

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NORTH DAKOTA ACADEMY OF SCIENCE  
(Official State Academy; founded December, 1908)  
1978-79

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71st ANNUAL MEETING  
April 20-21, 1979  
Joint Meeting of the  
Dakota Academies of Science  
held at  
Aberdeen, South Dakota

### Editor's Notice

The Proceedings of the North Dakota Academy of Science was first published in 1948, with Volume I reporting the business and scientific papers presented to the fortieth annual meeting, May 2 and 3, 1947. Through Volume XXI, the single yearly issue of the Proceedings included both Abstracts and Full Papers. Commencing with Volume XXII the Proceedings were published in two Parts. Part I, published before the annual meeting, contained an Abstract of each paper to be presented at the annual meeting. Part II, published later, contained full papers by some of the authors.

Commencing with Volume XXXIII of the Proceedings of the North Dakota Academy of Science, a new and functional format will appear. The Proceedings will change to an 8½ x 11 format, it will be produced from camera-ready copy, and it will be issued in a single part prior to the annual meeting (*i.e.* in mid-April).

Each presentation at the annual meeting will be represented by a full page "Communication" which will be more than an abstract, but less than a full paper. The communications will contain results and conclusions, and permit data presentation. The communication will convey much more to the reader than did an abstract, but still provide the advantage of timeliness and ease of production. The communications normally will be reviewed by the Editorial Board prior to acceptance and publication.

The Communications included in this volume also represent another "first." The 1979 meeting, labelled the Dakota Academies of Science meeting and represented by the logo on the cover, is the first joint meeting held by the North Dakota Academy of Science and the South Dakota Academy of Science. The Communications published in this volume therefore include those from professional and collegiate members of both Academies.

A. William Johnson  
Editor

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# NORTH DAKOTA ACADEMY OF SCIENCE

## I. Rules for Preparation of Proceedings Communication

1. Each paper presented at the annual meeting of the Academy must be represented by a communication in the Proceedings, including A. Rodger Denison student research competition papers.
2. Only communications intended for presentation at the annual meeting will be considered for publication. They must present original research in as concise a form as possible. Quantitative data should be presented with statistical analysis (i.e., means with standard errors). Papers which merely summarize conclusions or ideas without supporting data are discouraged and will not normally be accepted. The communication should include the purpose of the research, the methodology, results, and conclusions.
3. Authors are encouraged to utilize the full space available in order to provide sufficient information to fully describe the research reported.
4. Communications must be prepared on the special blue-line form and sent, with two legible xerox copies, by first class mail to the Secretary, North Dakota Academy of Science, University Station, Grand Forks, ND 58202. The form must not be folded; a cardboard backing should be used to avoid damage. The Proceedings will be published by direct photo-offset of the submitted communication. No proofs will be prepared.
5. All typing, drawing and secured art or photographic materials must be within the boundaries of the blue-line form. Consult the example on the reverse side of the special form for proper style (i.e., titles, authors, address, tables, figures, references, indentations, headings, and punctuation). *Indicate the author to present the communication by an asterisk (\*) after that person's name.*
6. Tables, diagrams, and photographs are acceptable provided they are secured to the special form and do not occupy a total area of more than 100 square centimeters.
7. Only essential references should be cited, and should be indicated in the text by numerals and quoted at the end of the communication. Up to three authors' names may be cited in full; with four or more authors only the first should be cited. The following form of citation should be used:
  - Journals: Neary, D., Thurston, H. and Pohl, J.E.F. (1973) *Brit. Med. J.* 3., 474-475. (Abbreviate titles.)
  - Books: Batsone, G.F., Blair, A.W. and Slater, J.M. (1971) *A Handbook of Pre-natal Paediatrics*, pp. 83-90. Medical and Technical Publishing, Lancaster
  - Individual chapters in books: Farah, A.E. and Moe, G.K. (1970) in *The Pharmacological Basis of Therapeutics*, 4th edition (Goodman, L.S. and Gilman, A., eds.), pp. 677-708. Macmillan, New York
  - Conferences and symposia: Rajewsky, M.F. (1973) Abstr. 2nd Meeting European Association for Cancer Research, Heidelberg, Oct. 2-5, pp. 164-5
8. Use a typewriter with elite type and with a carbon or good quality black silk ribbon. Single space and begin paragraphs with a 3 space indentation. Special symbols, not on the typewriter, must be hand lettered in black ink.
9. Abbreviations: Only standard abbreviations should be used, and should be written out the first time used with the abbreviation following in parentheses.
10. Titles: It is suggested that authors select a sufficient number of keywords to describe the full content of their paper, and then construct a title using as many of these as practicable. Title normally should not exceed 140 characters in length. In particular, they should be free from unnecessary phrases such as "a preliminary investigation of" or "some notes on" which add little or nothing to their meaning.
11. Session Assignment: In order to assist the program committee in organizing the presentations, please indicate on the reverse side of the blue-line form your 1st, 2nd, and 3rd preferences for the topical classification of your paper.
12. The authors' permission for the North Dakota Academy of Science to publish is implied by a submission. The Academy does not restrict the right of authors to include data presented in a communication in full papers submitted at a later date to other publishers.

## II. Rules for Oral Presentation of Paper

1. All papers are limited to 15 minutes total time, for presentation and discussion. It is suggested that the presentation be limited to 10 minutes with an allowance of 5 minutes for discussion. It is also suggested that major emphasis be placed on the significance of the results and the general principles involved rather than on the details of methods and procedures.
2. Academy members represent a variety of scientific disciplines; therefore, speakers should avoid "jargon" and briefly explain or define such specialized terminology as may be judged to be indispensable to the presentation.
3. Projectors for 2" x 2" slides only will be available in all session rooms. Opaque projectors will NOT be provided. Only slides which can be read easily on projection should be used. Authors who desire suggestions for preparation of slides are referred to Smith, Henry W. 1957. "Presenting information with 2 x 2 slides." *Argron. J.* 49, pp. 109-113.
4. Timed rehearsals with slides are highly recommended. There is usually time for a *maximum* of 6 or 7 slides for a presentation of this kind.

## PROFESSIONAL COMMUNICATIONS

### PROFESSIONAL COMMUNICATIONS

In this section of the Proceedings appear ninety-eight Communications representing presentations in the Professional Section of the 1979 Annual Meeting of the Dakota Academies of Science, the special joint meeting of the North Dakota Academy of Science and the South Dakota Academy of Science. Student Communications appear in the Collegiate Section of this volume of the Proceedings.

#### ALIGNMENT SENSING AND DETERMINATION OF LARGE X-RAY OPTICAL SYSTEMS

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The discovery of stellar X-ray sources and the desire to correlate the X-ray observations with radio, infra-red, and visible observations has led to the development of orbiting X-ray observations using Wolter Type I optics.<sup>1</sup> In order to correlate this X-ray data with other observations, the attitude of X-ray sources with respect to known visible sources must be achieved to accuracies on the order of 1 arc second or better.

The proposed Advanced X-ray Astrophysics Facility (AXAF) is an orbiting X-ray observatory scheduled to be shuttle-launched in the mid-1980's. Unlike the currently operational HEAO-II satellite, AXAF is to be a long-lived facility with replaceable components and support systems. The projected lifetime of the observatory is ten years.

Pointing accuracies of approximately 1.5 arc seconds are currently being achieved with HEAO-II. The design goal for AXAF is to improve this by at least a factor of three to .5 arc seconds. This study investigated the advantages of a proposed alignment scheme for AXAF which utilizes the focusing properties of the X-ray mirror assembly. A ray tracing program written by D. Korsch<sup>2</sup> was used to investigate the positioning of fiducial light sources in the focal plane of the X-ray telescope and the possible effects on system alignment.

The results of the study showed that there is an optimum positioning of the fiducial sources and that the X-ray mirror assembly can be used to determine system alignment. An analysis of the alignment errors showed that it should be possible to meet the design specifications of AXAF using this method of alignment if new image sensors are developed for star trackers.

1. Weisskopf, M. C., "Design of Grazing Incidence X-Ray Telescopes", Applied Optics, Vol. 12, No. 7, July 1973.
2. Korsch, D., "Analysis and Design Optimization of the 1.2 m X-Ray Telescope, Final Report, Vol. I", Contract NA8-32829 for Marshall Space Flight Center, TAI Corporation, Huntsville, Alabama, July 1978.

\*Work performed while author was a NASA/ASEE Summer Faculty Research Fellow at Marshall Space Flight Center, Huntsville, Alabama.

Biological Control Attempts Using Five Species  
of Bacillus as Seed-Treatments of Wheat

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Garden City, Kansas 67846 and Plant Science Department,  
South Dakota State University, Brookings,  
South Dakota 57007, respectively.

Several spore-forming bacteria of the genus Bacillus (Fisher) have been studied as biological control agents for reducing plant diseases (1). Their success in biological control depends primarily upon their production of an antibiotic inhibitory to certain plant pathogens (2). Several species of Bacillus were studied as potential biological seed-treatments on spring wheat.

Spring wheat seed (Triticum aestivum 'Protor') treated with one of several species of the genus Bacillus (B. globigii, B. polymyxa, B. subtilis, B. thuringiensis subsp. sotto and B. uniflagellatus) was planted in hill plots or in plots inoculated with one of six organisms pathogenic to wheat (Fusarium roseum f. cerealis, Gleosporium bolleyi, Helminthosporium sativum, Pythium graminicola, Rhizoctonia solani and Pseudomonas syringae). Yields and root lesion counts were not affected by the seed treatments.

Since growth of the Bacillus spp. precedes antibiotic production, the effect of low temperatures on the growth of the bio-control agents was investigated. Bacillus spp. growth and antibiotic production and growth of the pathogenic organisms was tested at 10, 12, 14 and 16 C. On glucose yeast-extract agar the minimum temperature for both growth and antibiotic production of the Bacillus spp. was 12 C. On soil extract agar the minimum temperature for growth was 14 C, and 16 C was necessary for antibiotic production. The fungal pathogens grew at lower temperatures than the Bacillus spp. A temperature of 14 C was necessary for growth of the Bacillus spp. in a soil medium.

Since spring wheat is planted in soils that average 7-10 C at planting depth, these soil temperatures may be too low to allow growth and antibiotic production by the Bacillus spp. tested in this study. The low temperatures of the soil at spring wheat planting time may account for the absence of response to seed treatments in the field experiments.

1. Alexander, M. (1961) Introduction to Soil Microbiology, pp. 19-44. John Wiley & Sons, Inc.
2. Kommedahl, T., and Mew, I. C. (1975) Phytopathology 65, pp. 296-300.

ATTACHMENT SITES OF A STALKED CIRRIPEID IN THE  
CRETACEOUS NIOBRARA SEA

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The uppermost beds of the Smoky Hill member of the late Cretaceous Niobrara formation contain a sparse suite of megafossils. A member of this group is the stalked cirriped Stramentum harworthi (Williston). This report explores the possible attachment sites and habitats of this barnacle. Although the holotype was attached to a small oyster, Pseudoperna congesta (Conrad), the bulk of the known fossils have been found attached to the flanks of a straight-shelled ammonite, Baculites (smooth) sp. There is no evidence yet available to indicate whether the Baculites sp. was living or dead.

The logical conclusion therefore is that the flanks of Baculites sp. (living or dead?) represent the preferred attachment site.

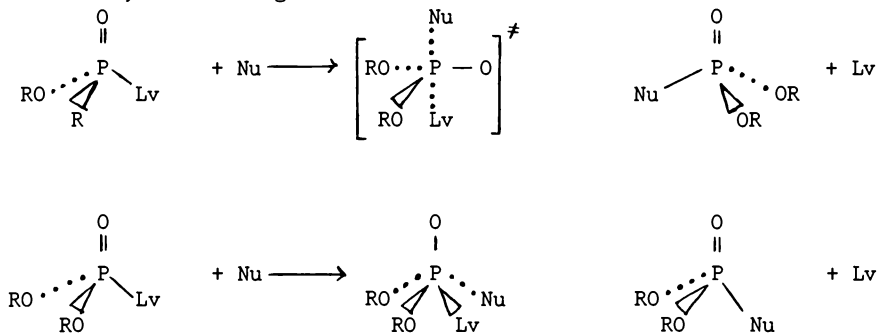


## SUBSTITUTIONS AT PHOSPHORUS: THE NUCLEOPHILE

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For a number of years we have been actively engaged in a study of phosphate chemistry. The phosphates are found in such diverse areas as plastics, insecticides, herbicides, plant hormones. The bio-phosphorus compounds which occur naturally are important to our body structure and function. Our present interest lies primarily in phosphate triesters,  $(RO)_3P(O)$ , and in the mechanisms by which alkoxy groups (RO-) are replaced, i.e., phosphorylation of alcohols. It is the most important reaction of phosphates and one which is the least understood.

Our work has led to the conclusion that phosphate esters undergo substitution by two separate mechanisms, one leading to inversion the other to retention.



We will describe our efforts towards the separation of these two mechanisms such that each can be studied independently. The parameters which influence each, especially with regards to the nucleophile, will be discussed as will our efforts toward the isolation of a plausible intermediate.

SARCOCYSTIS OF DEER IN SOUTH DAKOTA

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The incidence of Sarcocystis, a protozoan parasite, in white-tailed deer (Odocoileus virginianus) and mule deer (O. hemionus) in South Dakota was determined through histologic preparation and microscopic examination of tongue samples obtained throughout the state. The percentage of Sarcocystis infection for both species of deer was determined for prairies west of the Missouri River, east of the Missouri River, and the Black Hills of western South Dakota. Seventy-one percent of the mule deer tongues from West River, 87.5 percent from East River (counties bordering the Missouri River), and 100 percent from the Black Hills were positive. Tongue samples from East River white-tailed deer were negative; tongues from West River and Black Hills white-tails have not yet been examined.

Experimental coyotes (Canis latrans) and dogs were fed naturally infected Sarcocystis tissue from white-tailed and mule deer obtained from the Black Hills. Fecal samples were recovered and examined for sporocysts. Two red foxes (Vulpes vulpes), one gray fox (Urocyon cinereoargenteus), one bobcat (Felis rufus), and one raccoon (Procyon lotor) were also fed infected meat to determine their role if any as definitive hosts of Sarcocystis. All coyotes, dogs and the gray fox shed sporocysts, while none were recovered from the other animals. Sporocysts from coyotes fed white-tailed deer meat were counted and concentrated into an inoculum for oral administration to an experimental white-tailed deer fawn. No signs of weakness were observed. The deer was euthanized on post-inoculation day 85. Sections of heart, tongue, esophagus, diaphragm and skeletal muscle were found to be heavily infected with sarcocysts, while a control fawn's tissues were negative.

This research has demonstrated a high incidence of Sarcocystis infection in deer of western South Dakota and has implicated the coyote as the major definitive host.

(Supported by South Dakota Agricultural Experiment Station Project H-887).

Decomposition of Organic Matter in Two Midgrass  
Prairies in Western South Dakota

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Decomposition of organic matter was observed in an alpine midgrass prairie and in a midgrass prairie in western South Dakota by measuring absorption of evolved  $\text{CO}_2$  in a KOH solution. In the midgrass prairie an ungrazed area was also compared to a grazed area.

The alpine prairie evolved  $11.87 \text{ g CO}_2/24\text{hr/m}^2$ , significantly more than the  $4.19 \text{ g CO}_2/24\text{hr/m}^2$  for the ungrazed area or  $5.77 \text{ g CO}_2/24\text{hr/m}^2$  for the grazed area of the midgrass prairie. The alpine meadow evolved more carbon during the growing season ( $389 \text{ g C/m}^2/\text{season}$ ) than either the ungrazed area ( $211 \text{ g C/m}^2/\text{season}$ ) or grazed area ( $290 \text{ g C/m}^2/\text{season}$ ) of the midgrass prairie. Theoretical net primary production, (TNPP), as determined from  $\text{CO}_2$  evolution, was  $864 \text{ g C/m}^2/\text{season}$  for the alpine prairie,  $469 \text{ g C/m}^2/\text{season}$  for the ungrazed area and  $643 \text{ g C/m}^2/\text{season}$  for the grazed area of the midgrass prairie. Theoretical NPP was from 25-56% higher than actual net primary production (ANPP) in both the midgrass prairie and alpine midgrass prairie.

The difference between theoretical net primary production and actual net primary production may represent root and microbial respiration; but more likely represents errors in the methods for observing organic matter decomposition since the difference between TNPP and ANPP could not be correlated with a difference in root biomass. By increasing sampling frequency, increasing sampling area, and eliminating interpolation of data, the use of  $\text{CO}_2$  evolution as a determination of decomposition may be more quantitative.

Photoinitiated Cationic Polymerization by Electron-Transfer from  
Photosensitizers to Diaryliodonium and Triarylsulfonium Salts

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Diaryliodonium and triarylsulfonium salts of complex metal halides have been shown to be effective photoinitiators for cationic polymerization.<sup>1</sup> Due to the low absorptivity of these salts in the near ultraviolet spectral region (300-400 nm), we have undertaken photosensitization studies to enhance their spectral response and quantum efficiency.<sup>2</sup> The effectiveness of each photosensitizer/onium salt combination was evaluated by monitoring gelation of a diepoxide monomer as a function of time of irradiation. Three mechanisms are considered to explain the photosensitization results: (1) classical energy transfer, (2) homolytic bond cleavage of the onium salt within a photosensitizer-onium salt excited state complex, and (3) electron transfer from the photosensitizer to the onium salt. Each mechanism was evaluated on the basis of energetics and it was concluded that the results are best explained in terms of the electron transfer mechanism. Photogeneration of the cationic initiator by photosensitization as well as by direct irradiation of the onium salts will be discussed.

1. Crivello, J. V. (1978) in UV Curing: Science and Technology, S. P. Pappas, Ed., Technology Marketing Corporation, Stamford, Conn., Ch. 2 and references cited therein.
2. Pappas, S. P. and Jilek, J. H. (1979) Photogr. Sci. Eng., in press.

## UPLAND SANDPIPER NESTING IN SOUTHEASTERN SOUTH DAKOTA

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Waterfowl, pheasant (Phasianus colchicus), and upland sandpipers (Bartramia longicauda) were the predominant upland nesting species on U.S. Fish and Wildlife service waterfowl production areas (WPA's) in southeastern South Dakota during 1974-75. Habitat types and characteristics important for nest site selection of these species were determined. The effect of grazing by cattle on nesting was studied to provide managers with habitat management guidelines. This report is restricted to the upland sandpiper.

Nests were identified as being in either native prairie grasslands or tame grass and/or legume seedings. Plant species composition, range condition, range site, vegetation height, and mulch condition of the residual vegetation were determined at nest sites. Nest success and nesting densities were calculated.

All 33 upland sandpiper nests that were found during this study were in native prairie grasslands and none were in the tame grass or legume seedings. Native prairie in good or excellent range condition contained 97 percent of the nests. Of the 775 HA of native prairie searched during the study, 64 percent was in good or excellent range condition.

Nest density on tracts of native prairie rested during the year of nesting and on spring grazed (May 1-31) areas was the same at 1.7 nests per 40.5 HA. In general, vegetation taller than 60 cm was not used for nesting. Overall nest success was 80 percent.

Because native prairie grasslands are preferred nesting habitat for upland sandpipers, native prairie should be managed to attain excellent range condition. Periodic manipulation (2-3 years) by management techniques such as burning, grazing, or haying can be used to attain or maintain native prairie in excellent condition. A well planned habitat management system can provide areas for nesting as well as for other wildlife uses.

<sup>1</sup>Present address: Wisconsin Department of Natural Resources, State Game Farm, Poynette, Wisconsin 53955

## A PHYSIOLOGICAL FUNCTION FOR METALLOTHIONEIN

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Metallothionein (MT) is a metal binding protein, inducible by Zn(II), Cu(II), and Cd(II), which, because of its metal binding properties, has been implicated in heavy metal metabolism. To show that MT participates in zinc homeostasis, the ability of ZnMT to reactivate apoenzymes, prepared from zinc enzymes (carbonic anhydrase [CA], alkaline phosphatase [AP], alcohol dehydrogenase [AD], thermolysin [TL], yeast aldolase [YA]), was compared to that of various zinc salts. Ten  $\mu\text{g}$  of an apoenzyme was incubated with saturating amounts of  $\text{ZnSO}_4$ ,  $\text{ZnNO}_3$ , ZnAcetate, or ZnMT for up to 2 hr at 37°, and the extent of reactivation was monitored by appropriate assays. With apoYA ZnMT gave 100% reactivation within 30 min. Reactivation by  $\text{ZnSO}_4$  and ZnAcetate was complete and instantaneous. With apoCA ZnMT was better than any of the zinc salts with a maximal reactivation of 54%. At a 10:1 molar ratio of Zn:TL ZnMT and  $\text{ZnNO}_3$  completely reactivated apoTL. With apoAP 43% reactivation was obtained with ZnAcetate and 18% with ZnMT. Reactivation of AD failed. That metal transfer by ZnMT was an ongoing process was shown by the fact that dipicolinic acid and 1,10-phenanthroline had minimal effects on reactivation of apoCA when added after incubation, (apoCA + ZnMT) + chelator, but inhibited reactivation when added before incubation, apoCA + (ZnMT + chelator). Thus, ZnMT could function in zinc homeostasis as a reservoir of zinc for zinc apoenzymes. [Supported by NIH grant ES 01288, with funds from the EPA (IAG-DS-E772), and an RCDA, ES 00022.]

## 2,4-D INDUCED ALTERATIONS IN MAMMALIAN CHROMOSOMES - PRELIMINARY RESULTS

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The herbicide 2,4-D (dichlorophenoxyacetic acid) a chlorinated hydrocarbon, is widely used as a broad leaf herbicide. Several compounds of this group used for biological control are known to resist biodegradation and are potential mutagens. However, there is essentially no such research conducted on 2,4-D. The purpose of this study was to determine if 2,4-D is mutagenic, particularly to mammals.

Various concentrations of 2,4-D were introduced to human leukocytes in culture, and intra-peritoneal injections in several species of small mammals were made, to assess the effect on the chromosomes. Cells from human whole blood were cultured under standard procedures. After 20 hours of incubation of the lymphocytes, 2,4-D dissolved in ethyl alcohol was introduced. One lambda of various dilutions of 2,4-D was used for each culture so that the final concentration of the treatments in  $\mu\text{g/ml}$  were 0.2, 2.0, 10, 20, 30, 40, 50 and 60. Two groups of controls were used, one with ethyl alcohol and the other without any treatment. Air dried slides were analyzed for chromosomal damages.

In vivo studies are currently underway on Mus musculus and Rattus norvegicus. The treatment ranges from  $1.25 \times 10^4$  ppm to  $1.5 \times 10^3$  ppm of 2,4-D in safflower oil. Each animal was injected with 0.01 ml of the various 2,4-D concentrations per gram body weight. Controls received either oil or no treatment. Forty-eight hours after the exposure, bone marrow was used for chromosomal analysis following standard techniques. Chromosomal breaks, gaps and other changes are being analyzed at each level of treatment and control.

Preliminary results indicate that besides breaks and gaps, chromosomes display varying levels of coiling and "G" banding. Partially banded chromosomes were particularly frequent at lower concentrations of 2,4-D. Though the levels of gaps and breaks seem similar between controls and lower concentrations, the difference is rather sharp at 50  $\mu\text{g/ml}$  in human lymphocytes. Further data on both in vitro and in vivo studies will be conducted before arriving at a conclusion if the compound is mutagenic to mammals at certain levels. It does seem that at the 50  $\mu\text{g/ml}$  level or higher 2,4-D is a potential mutagen to human cells in culture.

## WHOLE ASPEN TREE SILAGE AS WINTERING FEED FOR BRED STOCK COWS

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South Dakota State University, Brookings, South Dakota

Thirty bred Hereford stock cows were used in a wintering experiment to maintain the breeding herd of the Western Dakota Vo-Tech Institute herd located at Sturgis, South Dakota. Fifteen animals were fed a traditional wintering ration of alfalfa-brome hay with free choice minerals and water. Fifteen animals were fed an experimental whole aspen tree silage (87% aspen, 3% pro-sil nitrogen source, 10% corn grain with alfalfa added at feeding), with free choice minerals and water. During the 123 day feeding period which began in December and terminated in April, alfalfa supplementation was varied to regulate animal condition. For example, after the first 17 days animals had increased in condition on a ration of 75% aspen silage and 25% alfalfa with a daily gain averaging 0.68 kg. It appeared that the stock cows would have difficulty in calving if the increase in condition was allowed to continue until spring. An adjustment was made such that animals were allowed a diet of 87.5% aspen silage and only 12.5% alfalfa for a period of 56 days. The initial ration was resumed prior to calving to allow for better milk production and promote increases in calf birth weights. Vigorous, healthy calves were born in each group with no death loss of cows, although one animal was removed in both the control and experimental group prior to calving. One additional animal was removed in the experimental group when the calf was aborted for causes not related to the experiment. All animals were in excellent condition at calving time. The experimental aspen silage was readily consumed throughout the experiment. Little, if any, spoilage losses were observed and silage quality parameters (pH, temperature, lactic acid level, odor) were similar to traditional silages prepared from corn.

# PROFESSIONAL COMMUNICATIONS

## ENERGY EFFICIENCIES OF ELECTRIC IRRIGATION PUMPING PLANTS

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A field study was conducted to evaluate the energy efficiencies of electric powered irrigation pumping plants in eastern South Dakota. The purpose of the study was to evaluate the energy status of irrigation technology in South Dakota and to obtain data which will serve as bench mark data for future efficiency studies.

Evaluations were performed on 33 pumping plants. Energy inputs, in the form of electrical energy to the motor, and energy outputs, in the form of water horsepower, were measured. The ratio of energy output to energy input was used as a measure of pumping plant energy efficiency.

Table 1 summarizes the results of the study. The efficiency values ranged from 47 to 73 percent with an average value of 63 percent. These data indicate that the pumping plants were operating at a relatively high level of efficiency when compared with the results of other reported studies. This is to be expected since the majority of the pumping plants were in service less than four years.

TABLE 1

### Pumping Plant Efficiencies

	Percent					Total	
	45-49	50-54	55-59	60-64	65-69		70-75
Number of Plants	1	1	7	10	9	5	33
Percent of Total	3	3	21	30	27	16	100

## INTERACTIONS BETWEEN SOIL-BORNE MICROORGANISMS AND INOCULUM IN DOSAGE-RESPONSE STUDIES WITH PHYTOPATHOGENIC FUNGI.

G. W. Buchenau\* and C. W. Wirth  
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Knowledge of the relationship between inoculum dose and plant response is basic to research on soil-borne plant diseases. The traditional inoculum bases, a cornmeal-sand mixture or wheat-oats-barley mixtures, have certain disadvantages: the former is difficult to quantify as propagules, the latter poses problems due to its relatively large propagule size and its tendency to become mush during autoclaving. To alleviate these problems, we have been using autoclaved Proso millet (Panicum miliaceum) seed as a growth substrate for Cochliobolus sativus, Gibberella zeae and Fusarium roseum "culmorum". In initial tests, these inocula effectively established dosage response curves for growth reduction of spring wheat seedlings. However, it was also discovered that sterile millet alone produced lesser but significant reductions in growth. Treatment of the sterile millet with arasan, captan or mancozeb fungicides eliminated the toxic effect, but terraclor treatment did not. Isolations from untreated and terraclor treated millet revealed an extensive colonization by Fusarium spp, predominantly F. solani, F. oxysporum and F. roseum. It appeared that a substantial proportion of the dosage-response curves for the common roots rot pathogens were confounded with effects of other fusaria.

Later tests, however, showed that when millet was colonized thoroughly by any of the common root-rot fungi prior to sterilization, this "killed inoculum" was not detrimental to wheat growth. We conclude that sterilized millet is a valid inoculum base for determining dose-response curves for these pathogens, without compensating for the effects of sterile millet. This conclusion assumes that the precolonized millet is not readily invaded by the confounding fusaria, either because of its low nutrient level or because of toxic metabolites.

## CHARACTERIZATION AND QUANTIFICATION OF WATER SOLUBLE FRACTIONS OF OUTBOARD MOTOR EXHAUST

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Operation of two-cycle outboard motors results in a wide variety of combusted and non-combusted hydrocarbons which are forcefully expelled into water. A portion of the hydrocarbons are water soluble (water soluble fraction or WSF) while others are insoluble and form an emulsion. The lighter hydrocarbons probably volatilize and are lost rapidly.

Laboratory experimentation consisted of running a 7.0 HP outboard motor in a 50 gal. galvanized tank containing Fargo City tap water. The tank was equipped with a spigot drain at the bottom and samples were removed from the tank via this drain. Samples were placed in separatory funnels and allowed to stand for 5 days. An emulsion was noticed during this time, and there was no apparent partitioning of the WSF's from the aqueous phase. The emulsions were not removed by either centrifugation at 4°C at 27,000 x g for 30 min., or passing them through a silica support column, or solvent extraction.

One hundred ml of emulsified liquid was passed through a short reverse phase column containing support material coated with a C18 stationary phase (Sep-paks; Waters Associates, Milford, MA). The sample hydrocarbons adsorbed on the Sep-pak were eluted from the column using 2 ml of acetonitrile or hexane. This method extracts the WSF's and probably a portion of the emulsion with at least 85% efficiency. Two to three µl of the eluted sample were injected on an 8' 1. x 1/8" O.D. x 2 mm I.D. stainless steel gas chromatographic column. The column support was 100/120 Chromosorb coated with 1,2,3-tris (2-cyanoethoxy) Propane (TCEP) available from Supelco, Inc., Bellefonte, PA. The gas chromatograph was operated under the following conditions: carrier gas (He) = 60 cc/min., H<sub>2</sub> pressure = 44 cc/min., compressed air = 240 cc/min., column pressure = 20 psi, column temp. = 80°C, and detector temp. = approximately 180°C. The detector was a flame ionization detector (FID). Peaks were tentatively identified in two ways: (1) by comparing retention times with those of known analytical standards and (2) by a peak enhancement method. Among the known carcinogenic and/or toxic compounds found thus far are benzene, toluene, and o-, m-, and p- xylenes.

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A CHEMICAL BASIS FOR THE ANTI-TUMOR ACTIVITY OF PLATINUM (II) COMPLEXES<sup>1</sup>

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In 1969 Rosenberg, et.al., first reported that various platinum complexes were able to inhibit sarcoma 180 and leukemia L 1210 in mice (1). Since this historic publication there has been an enormous amount of research performed that has been related to the anti-tumor activity of various platinum (II) and platinum IV species (2,3). In an effort to understand the implications of this research, a chemically consistent model has been developed in this laboratory which accounts for the variations in the activity of the platinum (II) complexes. Specific attention is directed toward the influence of *cis*-substituted amines, the apparent specificity of the platinum (II) complexes for rapidly dividing tumor cells, and the growth cycle of a cell. Special attention will be given to the predictions associated with the model and the necessary experiments that must be performed to evaluate the model.

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EFFECTS OF MERCURY AND SELENIUM ON EMBRYOS AND LARVAE OF THE JAPANESE MEDAKA  
(Oryzias Latipes)

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The protective action of selenium against mercury toxicity is well documented. However, a recent study (1) has demonstrated that during embryonic development the adverse effects of the two elements may be more than additive. These conflicting data raise the question of whether a vertebrate may be protected from Hg by Se at some parts of its life cycle but not at others. To investigate this phenomenon we utilized inorganic Hg (as mercuric chloride) and Se (as selenium dioxide). We examined the toxic effects of these elements when administered separately and when given as mixtures.

Based on LC<sub>50</sub> determinations of embryos the toxicity for mercury (LC<sub>50</sub>=0.047 mg/L and 95 confidence limits = 0.04-0.055) was greater than that for selenium (1.66; 1.27-2.16). The ratio of molar concentrations of the LC<sub>50</sub> values was 1:90. Larval 96 hr. LC<sub>50</sub> values for Se (3.0; 2.3-3.92) and Hg (0.737; 0.665-0.818) indicate that Hg is also more toxic to larvae.

Teratogenic effects were characteristic for each toxicant. Mercury caused a tube heart and prevented the formation of red blood pigments. Selenium caused poor differentiation of the heart into chambers resulting in a "knobby tube". Other defects were noted.

When one mg/L selenium was given simultaneously with mercury, or as a pretreatment, antagonistic interactions were observed; the embryos were protected. A hypothesis is advanced which might explain contradictory results concerning the interactions of these elements during embryonic development.

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STUDIES ON SARCOCYSTIS OF WILD UNGULATES IN SOUTH DAKOTA

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Research was initiated to determine the incidence of Sarcocystis in wild ungulates and to ascertain the transmissibility of the protozoan parasite to domestic livestock. Tongue samples were collected from elk, pronghorn antelope, mountain goats, bighorn sheep, and bison from western South Dakota. Histological examinations were made for sarcocysts in muscle. All elk samples examined showed heavy infection with Sarcocystis. Most of the bison and mountain goats were lightly infected in comparison with elk, although they occupy approximately the same range in the Black Hills. None of the antelope examined showed infection.

Elk meat infected with Sarcocystis was fed to 1 red fox, 2 coyotes, and 2 dogs. Fecal samples were collected daily for 45 days and examined for sporocysts using a sugar flotation technique. The number of sporocysts shed per gram of feces was calculated. Both the coyotes and the dogs were good definitive hosts, shedding large numbers of sporocysts, while the red fox shed very few sporocysts. To test for transmissibility of Sarcocystis from wild to domestic ungulates, cervid sporocysts from canid feces were concentrated in an inoculum and administered per os to calves, sheep, and pigs. These animals will be euthanized approximately 60 days post inoculation. Various tissue samples will be examined histologically for presence of sarcocysts.

In conclusion, this research has shown that elk in the Black Hills are heavily infected with Sarcocystis and that coyotes are highly susceptible definitive hosts. Other aspects of sarcocystosis are still under investigation.

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DIAGNOSIS AND PATHOLOGIC EFFECTS OF  
PENTACHLOROPHENOL TOXICOSIS IN YOUNG PIGS

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During the last few years an increasing number of cases of suspected pentachlorophenol (PCP - wood preservative) poisoning of farm animals have been brought to the attention of veterinarians and extension agents. The diagnosis of PCP poisoning has been largely based on the statements of the farmers that the animals were subjected to PCP's. To improve methods of diagnosis and to study pathologic effects, a study was initiated in October, 1978. Three treatment groups each consisting of six young (about six weeks old) pigs were administered 5, 10 and 15 mg/kg/day of purified PCP in capsule form for 30 days. One group of six pigs served as a control and received a daily capsule containing pure lactose. Levels of PCP in the blood, kidneys and livers of the 5, 10 and 15 mg groups were not significantly different with an average of 74, 25 and 28 ppm, respectively. Levels of PCP in controls were below 1.0 ppm in all three types of samples.

Significant differences ( $P < .05$ ) were found between the PCP treated groups and the controls with an increase in the weight of livers on g/kg body weight basis, an increase in blood urea nitrogen and a decrease in white blood cells. During the course of the study the gamma globulin levels in the controls increased while those in the PCP groups did not. The conclusions of this study were that at levels above 5 mg/kg/day no further increases of PCP occur in blood, kidneys or livers of young pigs. The increase in blood urea nitrogen and g/kg body weight of liver could indicate damage to the kidney and liver. The lack of increase in gamma globulin and the decrease of white blood cells of PCP treated pigs may indicate a weakening of the immune systems.

## EFFECTS OF TILLAGE PRACTICES ON NEMATODE POPULATIONS ASSOCIATED WITH CORN AND SPRING WHEAT

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Recent studies indicate several species of plant parasitic nematodes are important factors in corn and spring wheat production in South Dakota. At present the principal method of nematode control is the use of nematicides. Information concerning effects of tillage practices may provide supplemental or alternative control procedures. Studies conducted over a two year period in continuous corn near Madison indicate lesion nematode populations increase under minimum tillage. In 1976 the highest populations occurred in no till plots and yields in these plots were only slightly higher than in conventional tillage. In 1977 populations were highest in minimum (Buffalo) till plots and yields in these plots were less than conventional till. These initial studies indicate that higher lesion nematode populations under minimum tillage may partially offset the advantages of additional moisture conserved with these operations.

Studies in spring wheat have been conducted in three locations. Results obtained at two locations have been rather inconclusive except that populations of lesion nematodes have declined under continuous spring wheat culture. In the third location populations of dagger, pin and spiral nematodes are highest under minimum tillage while numbers of stunt nematodes are lowest. The above four nematode groups have all been previously associated with damage to spring wheat. Whether the decline in stunt nematode populations offsets the increase in the remaining groups in relation to spring wheat injury requires further investigation.



## CLONING OF SMOOTH BROMEGRASS THROUGH CALLUS CULTURE

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 57007

Cloning of smooth brome grass (Bromus inermis Leyss.) through callus culture has been achieved following the methods described previously (1).

Calluses were initiated from segments of unemerged young inflorescences explanted on RM medium supplemented with 4 possible combinations of 5 and 10 mg 2, 4-dichlorophenoxyacetic acid (2,4-D) and 0 and 0.2 mg kinetin/liter in the dark at 25°C. These calluses were then propagated on the same basal medium containing 5 mg 2,4-D/liter under the same environmental condition. Shoots differentiated from calluses subcultured on RM medium which contained either no hormones or kinetin at 0.2 to 1.5 mg/liter. No apparent enhancement of shoot formation was observed in the presence of kinetin. Shoot formation was the only type of organogenesis noted in the morphogenetic induction media. No aerial roots as seen in Indiangrass cultures (2) appeared. Plantlets developed after the shoots were transferred onto the RM basal medium under 16 hr light cycles (cool white fluorescent at 25  $\mu$ E/sec m<sup>2</sup>) at 25°C.

The callus appeared to be maintained indefinitely on RM medium supplemented with 5 mg 2,4-D/liter, but albino plants began to emerge from the callus previously subcultured on the 2,4-D medium for 3 passages (1 month/passage). The proportion of the abnormal to green plantlets increased as subcultivation of callus prolonged. By 11th passage, all plantlets differentiated from the subcultures were albino. In addition to 11 albinos, which did not survive at the time of transplanting, 3 variants bearing narrow leaves were noted among 86 plants established from plantlets in the field. These plantlets were initiated from the calluses which had been subcultured for 5 passages. The variants remained at the rosette stage while the rest of the plants produced panicles in the first year of establishment.

Gamborg et al (3) reported that instead of shoot formation, embryogenesis occurred in a brome-grass suspension culture, but all the plantlets developed from the embryoids were albino and establishment of plants was not possible. Embryoids were also isolated from a protoplast culture of brome grass by Kao et al (4), but again no plants were established in the field.

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IDENTIFICATION AND QUANTIFICATION OF METALS IN THE HOG ROUNDWORM,  
ASCARIS LUMBRICOIDES SUUM

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The hog roundworm, Ascaris lumbricoides suum, was analyzed in an attempt to identify and quantify the metals found in this animal. A comparison was made of metals found in the males and females of this species of helminth parasite with the metals found in certain tissues of the host. The two hog tissues involved were skeletal muscle and kidney. Several methods of analysis were used for comparison purposes which included the ash method, wet digestion method, and the Olson method (for selenium). These preparatory methods were followed by either flame or flameless Atomic Absorption Spectrophotometric analysis.

The metals analyzed included Cd, Ca, Cu, Fe, Pb, Mg, Mn, Mo, K, Se, Zn, and As. There were no apparent differences in concentrations (ppm/dry wt.) of As, Ca, Pb, Mg, and Mo between the host tissues and the worms or between the sexes of the worms. Cu, Mg, and Fe concentrations appeared to be higher in the hog tissues than in the worms and the male worms were higher than in the females. K, Se, and Zn levels appeared to be higher in the hog tissues but there were no differences noted between male and female ascarids for these metals.

ENHANCEMENT OF FLAMELESS ATOMIC ABSORPTION DETERMINATION OF TELLURIUM IN  
BIOLOGICAL MATERIALS BY MEANS OF SOLVENT EXTRACTION

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The physiological and toxicological significance of tellurium in biological and botanical systems is unknown. This absence of information is primarily due to the lack of an analytical method for determining tellurium at the concentrations that are inherently present in living organisms or that are present in systems that have been exposed to reasonable elevated levels of tellurium in the environment. Interest in the role of tellurium in biological systems is stimulated by its relationship to selenium in the periodic table, an element that is well known for its essential role and toxic effects.

This paper reports a flameless atomic absorption procedure for the determination of tellurium in biological systems. The procedure incorporates a solvent extraction step for preconcentration of the sample and removal of interfering species. In testing the procedure, ten gram corn samples were digested with a tertiary acid mixture of nitric, perchloric, and sulfuric acids. The tellurium was extracted from the wet digestion solution by formation of an iodate complex according to the method of Hanson (1). The organic solvent of choice is n-pentanol. The extract is evaporated to near dryness then oxidized with hydrogen peroxide. Evaporation of the solution is completed and the residue is redissolved in dilute nitric acid. The tellurium content of the nitric acid solution is determined by heated graphite furnace atomic absorption spectrophotometry. A deuterium arc background compensator is used to correct for broad band absorption in the furnace.

The sensitivity of the method is  $2 \times 10^{-2}$  ng of tellurium for 1% absorption. This corresponds to a minimum detectable concentration of 0.2 parts per billion in the original corn sample. The recovery of tellurium added as a spike prior to digestion of the sample is  $98 \pm 16\%$ . The imprecision of the method appears to be related to the intense broad band absorption present in the vicinity of the 2143 Å tellurium line in spite of the preliminary extraction. This absorption is not perfectly corrected by the deuterium background compensation system. Current efforts are attempting to determine the cause of this intense absorption and reduce its magnitude.

This reported procedure for the analysis of tellurium represents a significant improvement in sensitivity compared to any other method reported in the literature. It should, therefore, assist in the enhancement of our understanding of the role of tellurium in living organisms.

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INTERACTION OF DIETARY CARBOHYDRATE, FAT AND  
PHOSPHATE IN LIPID METABOLISM

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Dietary carbohydrate (cornstarch or sucrose, approximately 65% by wt.), fat (corn oil or butterfat, 10%) and phosphate level (negligible, 0.46 or 1.85%), when fed for 7, 14 or 28 days to young adult male rats interacted in their effects on liver lipids, including phospholipid fractions. Sucrose usually contributed to higher levels of total liver lipids, phospholipid, triglyceride and, to a lesser extent, cholesterol. It also tended to increase levels of the larger fractions of phospholipid, whereas lysolecithin was more likely to be increased by butter or interaction of the two. Both high and low phosphate levels tended to increase liver lipids. Serum lipid levels were also affected most frequently by carbohydrate. Effects of phosphate level were as often noted as those of fat source.

It is difficult to interpret the many kinds of changes seen because not enough is known about the functions of the components measured. This is especially true of phospholipids which occur as structural components of membranes; changes could affect their permeability, function and/or integrity. Sugar saturated fat and high phosphate in diets, which could co-occur in the U.S., could be jointly affecting lipid metabolism adversely.

The results emphasize the importance of studying complex changes in whole animals under widely-varying dietary conditions.

IMPACT OF ZOOPLANKTON GRAZING ON PRAIRIE  
LAKE ALGAL STANDING CROPS AND WATER TRANSPARENCY

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The effect of zooplankton grazing on the populations of algal species was investigated in four prairie lakes. The change in standing crop of algae over a two-day period was measured by 16 in situ experiments by using three different concentrations of grazing zooplankton in large polyethylene containers (1). Also, gut contents of 289 zooplankters were analyzed (2). Correlation and regression analyses of changes in standing crop of algal species, changes of chemical and physical parameters of lake water, and changes in standing crop and calculated filtration rate of zooplankton (3) were performed.

Six common bluegreen algal bloom-formers, Anacystis incerta, A. cyanea, Aphanizomenon holsatica, Anabaena spp., Coccochloris penicystis, and Gomphosphaeria spp. were sometimes grazed by the zooplankton. Other species which were grazed included the diatoms Cyclotella spp., Stephanodiscus spp., Melosira spp., Asterionella formosa, Fragilaria spp., the green algae Oocystis spp., and Spondylomorom sp., the chrysophyte, Dinobryon spp. Concentrations of some of the species listed were increased by grazing, but more were depleted.

Calculated zooplankton filtration rates were significantly correlated with water transparency in all four lakes, and predictive formulas are developed for water transparency. Calculated zooplankton filtration rate showed significant correlations with chlorophyll a concentrations only in the deeper, less eutrophic lakes.

The grazing of abundant algal bloom species by the zooplankton suggests that they may be partially subject to control by zooplankton.

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SOCIAL HIERARCHIES  
AMONG PENNED PHEASANTS  
AND EFFECTS OF DIELDRIN  
ON INTERACTIONS

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Social interactions were observed among groups of pheasant chicks and adult cock and hen pheasants. To determine the effects of dieldrin on the pecking behavior of individual birds, pairs of pheasants and pairs of chickens were placed in a neutral cage. When patterns of dominance and subordination were consistent, dieldrin in capsules (4 mg to pheasants and 6 and 10 mg to chickens twice weekly) was given to one member of each pair and birds were observed for changes in pecking behavior. Toward the end of the study, one bird from each of the pheasant chick groups was given 4 mg of dieldrin twice a week.

Fights were observed among pheasant chicks at 3 weeks of age, and it was concluded that aggressive behavior and peck-order development began at that time. Only in smaller groups (two to five birds) could a rank be determined based on the total number of each individual's interactions. Groups of adult pheasants also displayed peck-orders, usually not linear.

Weight, previous dieldrin treatment, parental dieldrin treatment and ear tuft length had no effect on the peck-orders. There were no correlations between sex and rank in the pheasant chick groups. Dominance-subordination patterns of pheasants were not affected by dieldrin administration.

Examination of Low Rank Coals and Ash Deposits  
Using Thermal Analytical Procedures

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Thermal analytical procedures are becoming an important tool in characterizing coal and coal ash deposits. Thermal analysis is defined as the techniques in which some physical parameter is measured as a function of temperature.

The thermogravimetric technique (TGA) simply measures loss of mass as a function of temperature. The application to coal is the proximate analysis which is a traditional method of characterizing coal in terms of moisture, volatile matter, ash, and so-called fixed carbon. The TGA method shows good agreement within 7% of the standard methods. The results of a TGA proximate analysis on a Montana subbituminous coal are 23.90% moisture, 28.17% volatile matter, 37.39% fixed carbon, and 10.54% ash. The results of the standard method on the same coal are 23.08% moisture, 27.84% volatile matter, 37.73% fixed carbon, and 11.35% ash.

Pressure differential scanning calorimetry (PDSC) measures heat flow into or out of a sample under pressure. The heating value of coal can be determined by this method. A comparison of the data from the PDSC and standard bomb calorimetry shows agreement within 6%. The heat content of a lignite coal was determined by PDSC and bomb calorimetry to give values of 10921 and 10528 Btu/lb, respectively. The resulting thermograms indicate one or two large exothermic peaks in which the number of peaks and the intensity of the peaks vary with the rank of coal.

Differential thermal analysis (DTA) is a thermal technique where the temperature of the sample is compared to that of a thermally inert material. This difference in temperature is recorded as a function of furnace temperature.

At the Grand Forks Energy Technology Center a pilot scale combustor is used to study the fouling characteristics of low rank coals. Air cooled probes are placed into the combustor to simulate the heat exchange tubes in power plant boilers. Ash deposits form on these probes and are studied thermally by DTA techniques. The DTA characterizes the probe deposits as to phase changes, decomposition, and melting points. An endothermic peak between 900-960°C is characteristic of a large number of probe deposits. This peak correlates with the volatilization of  $\text{Na}_2\text{SO}_4$  observed at 850-950°C in the heated stage microscope.

Thermal analysis, because of its ability to do the proximate analysis and heat content, proves to be a useful tool in coal analysis. It also can be used as a research tool in characterizing coals by PDSC and thermally characterizing probe deposits with the DTA.

NUMERICAL SIMULATION OF QUANTUM MECHANICAL WAVE PACKET PROPAGATION

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A computer program has been developed to propagate wave packets past potential wells. The wave packet was generated by first finding the Stationary State Wavefunctions (SSW). Each SSW was multiplied by an appropriate weighting factor and its specific, time dependent, phase function. Summing these SSW's allowed the wave packet to be observed at arbitrary times. By plotting the wave packet at various times, reflection, transmission and resonance was observed.

## THE EFFECT OF SELENIUM AND LEAD ON THYROID FUNCTION IN YOUNG PEKIN DUCKLINGS

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Young Pekin ducklings were divided into four groups: one group served as controls; a second group received 3 lead shot dropped down the throat of each bird; a third group received sodium selenite at a concentration of 1 ppm in their drinking water twice daily; and a fourth group received both the 3 lead shot and sodium selenite in their drinking water.

Two weeks and three weeks later, 20  $\mu\text{Ci}$  of carrier-free  $^{125}\text{I}$  was injected into each bird and several indices of thyroid function were examined.

When necropsies were performed at 2 and 3 weeks after initiation of the investigation, thyroid weight was increased in all treated groups of birds at 2 weeks. The increase in thyroid weight at 3 weeks in the lead-selenium treated group of birds was not significantly different from that observed in the control group.

Thyroidal uptake of  $^{125}\text{I}$  24 hours after injection of radiiodine was increased in the lead-treated groups of birds at both 2 and 3 weeks although the serum protein-bound  $^{125}\text{I}$  was not significantly different from that of the control group at 2 weeks. A significant increase in serum protein-bound  $^{125}\text{I}$  in these lead-treated birds was observed at 3 weeks. Thyroidal uptake of  $^{125}\text{I}$  was decreased in the selenium-treated group of birds both at 2 and 3 weeks with associated decreases in serum protein-bound  $^{125}\text{I}$  levels. While thyroidal uptake of  $^{125}\text{I}$  in the lead-selenium treated groups of birds was not significantly different from that observed in the control groups at 2 and 3 weeks, serum protein-bound  $^{125}\text{I}$  was reduced at both 2 and 3 weeks.

Chromatographic analyses of thyroid hydrolysates revealed a significant depression in labeling of the iodothyronines, throxine and triiodothyronine, in birds that had been treated with lead alone or with selenium alone. There was some indication that lead-selenium treatment in accordance with the reputed antagonism between lead and selenium interacted to moderate the depression in labeling of thyroid hormones resulting from either lead or selenium treatment alone. The impairment in synthesis of thyroid hormones was associated with reduced serum protein-bound  $^{125}\text{I}$  levels and is probably reflected in a decrease in secretion of thyroid hormones. These results indicate that lead or selenium ingestion by Pekin ducklings alter thyroid function.

TOLERANCE OF FARMERS  
FOR A LOCAL CANADA GOOSE FLOCK

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Four hundred farmers were interviewed in northeastern South Dakota in 1974 and 1975. Eighty-six percent of the interviewed farmers indicated that they wanted the local goose flock to increase. Only 6 percent of the farmers had complaints about the geese, despite 23 percent having received goose related crop damage. Percent occurrence of complaints and crop damage was influenced by distance of the farm from the goose concentration areas. Farmers adjacent to goose concentration areas had a lower tolerance for geese than those farther away, but still retained relatively positive attitudes toward the geese and goose flock expansion.

SYMPTOM REMISSION ON GARDEN AND LANDSCAPE PLANTS AFTER REMOVAL OF A SULPHUR  
DIOXIDE SOURCE AT MOBRIDGE, SOUTH DAKOTA

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Symptoms resembling sulphur dioxide (SO<sub>2</sub>) injury were found in 1977 on cultivars of garden and landscape plants growing at Mobridge, South Dakota (1). In 1978, about a year after a suspected SO<sub>2</sub> source ceased operation, the same area and cultivars were observed for possible symptom remission.

A majority of the plants reexamined were free from symptoms. The 16 exceptions exhibited milder but similar symptoms one season's growth after the SO<sub>2</sub> source was removed. Malus sylvestris showed a light amount of brown patch leaf necrosis. Juglans nigra leaflets had a light amount of brown necrotic spots, streaks and chlorosis. Acer negundo leaves showed moderate marginal and interveinal tan necrotic spots and patches. Catalpa bignonioides showed severe chlorotic mottle and necrosis on leaf margins and between major veins with youngest leaves nearly normal. Prunus virginiana leaves had a moderate number of dark brown necrotic spots and shot-hole leaf symptoms. Chrysanthemum sp. showed moderate dark brown necrosis of leaf margins. Ulmus americana leaves showed a light amount of dark brown marginal serration necrosis. U. pumila leaflets had margins with brown necrosis and interveinal spotting. Sambucus canadensis leaflets had moderate brown necrotic margins and severe chlorotic mottle. Gladiolus sp. 'Glacier' leaf tips showed uniform tan necrosis. Acer platanoides 'Schwedler' had a few leaves showing dark brown patch necrosis. Acer sp. leaves had a trace of brown marginal serration necrosis. Morus rubra leaves had moderate marginal necrosis. Paeonia lactifolia had a light amount of leaf tip necrosis. Erianthus ravennae leaves had mild marginal and apical banded necrosis. Rhus copellina showed light amounts of brown necrotic spots on leaflet margins.

Twenty-two cultivars were entirely free from symptoms that were present in 1977 (1). In general the boulevard and landscape trees and shrubs had made remarkable recovery. There was evidence of the severe effect of SO<sub>2</sub> on limbs that had been killed prior to 1978. Striking evidence of partial recovery was shown by black hills spruce (1). The needles produced in 1978 were normal in contrast to the necrotic and chlorotic needles injured by SO<sub>2</sub> in 1976 and 1977. Observations will be continued to determine the cause of the symptoms that developed on the 16 cultivars following removal of the SO<sub>2</sub> emission. Residual effects of SO<sub>2</sub> pollution may be one explanation. Also, the symptoms may be caused wholly or in part by other factors or agents. It was apparent that removal of the emission source caused 22 cultivars to produce normal leaves and 16 cultivars to produce leaves showing much less pollution-like injury.

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NEMATODE POPULATION DYNAMICS IN FIRST YEAR WESTERN WHEATGRASS

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Nematode populations associated with a recent western wheatgrass planting were monitored throughout the growing season. Prior to the grass seeding the field had been used primarily for winter wheat production for approximately 40 years. Population changes were measured by removing 10 soil cores to a depth of 10 cm in each of 10 locations along a 100 meter transect, and nematodes were then extracted, identified and counted according to major taxonomic groups. Results indicated that members of the Tylenchidae increased significantly during the sampling period as did Quinisulcius, Xiphinema and the Dorylaimida other than Xiphinema. Numbers of Rhabditida showed an initial significant increase, but had decreased at the last sample date. The data indicate a response to the newly developing habitat. Populations of plant feeding nematodes increased, many of which are found in association with native prairie ecosystems. The number of different species encountered also changed during the study period. The May sample yielded 11 species, the June sample 15 and the November 14 species. Plant feeding nematode populations that increased were Quinisulcius acutus, Xiphinema americanum, and the Tylenchinae. Helicotylenchus sp. also increased, but to a lesser degree. Among the microbial feeders Acrobeles sp. and Eucephalobus sp. were major genera encountered. An Axonchium sp. was the primary dorylaim recovered.

THE CARBONIC ANHYDRASE OF *RHODOSPIRILLUM RUBRUM*

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The carbonic anhydrase (CA) enzymes of plant and animal tissues have received considerable attention (1). Microbial CA's have been rarely investigated, although the CA of *Neisseria sicca* has been purified to homogeneity and its properties described (2). We have investigated the CA of the facultatively photosynthetic bacterium *Rhodospirillum rubrum* and described some of its properties.

Large batches (50 L) of *R. rubrum* were grown photosynthetically with yields of about 100 g wet weight of cell paste. Cell-free enzyme preparations were made by passage of thick suspensions through a French pressure cell. CA activity ( $\text{CO}_2 + \text{H}_2\text{O} \rightleftharpoons \text{HCO}_3^- + \text{H}^+$ ) was assayed in MOPS-NaOH buffer by determining the time required for the pH to drop from 7.5 to 7.3. Crude CA preparations were purified by first discarding the 60%  $(\text{NH}_4)_2\text{SO}_4$  precipitate. The supernatant was desalted on Biogel P-4 and chromatographed on hydroxylapatite in 0.01 M  $\text{PO}_4$  buffer pH 7.3, with  $\text{PO}_4$  gradient elution (0.01 to 0.2 M). The active fractions were then chromatographed on Sephacryl that had been equilibrated with 0.1 M  $\text{PO}_4$ , pH 7.3. Hydrophobic interaction chromatography on phenyl sepharose was also a useful purification procedure.

The Sephacryl chromatography, as well as similar experiments with Biogel P-200 and Sephadex G-200, indicated a molecular weight of the order of 150,000 to 200,000. The enzyme was stable at 22°C and also to freezing and thawing. Kinetic experiments have been performed in which the substrate ( $\text{CO}_2$ ) concentrations were varied. From double reciprocal plots of initial velocity as a function of substrate concentration, the  $K_m$  of the reaction was calculated to be 139  $\mu$  molar  $\text{CO}_2$  and the  $V_{\text{max}}$  was 0.87  $\mu$  moles/min.

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EFFECTS OF MICRO-SLOPES CREATED DURING  
CULTIVATION ON RUNOFF WATER QUALITY <sup>1</sup>

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Effects small ridges created during corn cultivation have on runoff volume and quality were studied to test the practicability of meter-square runoff plots. The small inexpensive plots were found to be feasible to study cultural practices in large fields because the results were similar to those published from larger permanent plots receiving natural precipitation or small plots treated with artificial rainfall. Mean runoff per event was 11.6, 6.7, and 3.1  $\text{m}^{-2}$  from plots, respectively, with ridges oriented on the contour. The main sources of runoff nutrient were:  $\text{NH}_4$ -N, the precipitation;  $\text{NO}_3$ -N, the precipitation and soil; soluble and total  $\text{PO}_4$ -P and soluble K, the soil. Nutrient losses were related to runoff volume independent of the soil surface configuration. Losses are less if runoff is less.

<sup>1</sup>Approved for publication by the Director, Agricultural Experiment Station, South Dakota State University, Brookings, as Journal series No. 1544.

<sup>2</sup>Professor, Plant Science (Soils).

## HYDROGEOLOGY OF THE BELLE FOURCHE, SOUTH DAKOTA

## WATER INFILTRATION GALLERY AREA

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The Belle Fourche, South Dakota water infiltration gallery is situated in alluvial sands and gravels of the Spearfish Creek valley, Lawrence County, South Dakota. Water of generally good quality was obtained until the summer of 1977, when samples from the infiltration gallery began to show levels of contamination of 50-150 total coliforms per 100 ml., and 0-2 fecal coliforms per 100 ml. The water is chlorinated before reaching Belle Fourche users.

Belle Fourche city officials were understandably concerned about the decline in the quality of their drinking water. In an attempt to determine the source of this coliform contamination, forty-nine observation wells were monitored for water table levels, bacteriological and chemical water samples were taken from surface and ground waters, an aquifer test and a dye test were performed, and aerial photography was examined.

Possible sources of contamination were considered to be: (a) Nearby residential sewage from septic tanks situated about 1/4 mile upgradient from the infiltration gallery (b) induced infiltration from Spearfish Creek, which shows high levels of fecal and total coliform contamination at certain times of the year (c) the Spearfish sewage lagoon, which has been leaking since 1973, and (d) distant residential septic tanks in the upper reaches of the alluvial valley.

High rates of ground-water movement are indicated by an average aquifer transmissibility of approximately 1,000,000 gallons per day per square foot, as determined by the Theis method (1), and approximately 700,000 gallons per day per square foot as determined by flow net analysis (2). Spearfish Creek and nearby residential septic tanks are considered the most likely sources of contamination. Water could migrate from the creek to the infiltration gallery in two to three days; water could migrate from the residential area 1/4 mile away to the infiltration gallery in about five days. The Spearfish sewage lagoon cannot be ruled out as a possible contamination source but would require movement of contaminated ground water through shaly beds of the Spearfish Formation, then migration up dip, probably through partially dissolved gypsum layers.

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## SURFACE POTENTIAL CORRECTIONS IN THE CALCULATION OF MADELUNG SUMS

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Madelung sums are conditionally convergent infinite series which correspond to perfect, but otherwise physically realizable, crystal. Although these sums are most commonly evaluated using the method of Ewald, the conceptual simplicity of direct summation methods and the advent of the modern computer has led to a renewed interest in direct summation methods.

A necessary condition for the sequence of finite sums to approach the Madelung sum is an ordering of the terms to correspond to some "space filling" scheme such as an ordering in terms of the distance from the reference point. However, such an ordering is not a sufficient condition. In physical terms, the potential at the reference point is dependent upon the configuration of the ions at the surface of the corresponding crystallite.

Correction for any surface potential can prove troublesome because of the difficulty in assigning unambiguous meaning to the term surface. Previous solutions to this problem will be reviewed, and original results for several additional special cases will be presented.



## EDUCATIONAL USES OF THE ORDWAY PRAIRIE

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The Samuel H. Ordway, Jr. Memorial Prairie was purchased in 1975 by The Nature Conservancy, a national, non-profit, conservation organization. The Ordway Prairie was established for educational uses and scientific study, and to insure the existence of a portion of the native prairie ecosystem managed to the most natural conditions possible. The Ordway Prairie, 20 miles north and 29 miles west of Aberdeen, South Dakota, lies on the eastern edge of the Coteau du Missouri. The 7,600 acre prairie supports the natural vegetative community often referred to as mixed grass prairie. The topography consists of rolling hills, and an estimated 400 temporary and permanent wetland basins.

A full time manager lives at the prairie headquarters. A bunkhouse and lab-conference room is available for use by researchers and visitors. The lab-conference room is equipped with projection equipment and lab basics.

Six research projects have already been completed on the Ordway Prairie through cooperation with colleges and universities. An additional three projects are in progress. Research to date has generally involved work with the flora of the preserve. Studies remain to be done on the fauna of the preserve including the gathering of baseline information and the effect present land management has on wildlife populations. Ecological inventories, research and other educational uses are expected to enhance the Conservancy's ability to properly manage the preserve as well as be valuable to nearby landowners which include wildlife agencies.

Present management of the Ordway Prairie involves warm season deferment grazing by cattle and selected-rotation, yearlong grazing by a herd of buffalo. Controlled burning is also being considered as a management tool. Management plans are subject to change as natural history documentation, plant succession and research progress.

The Ordway Prairie is steadily being recognized as a community and regional educational resource. Field trips, work shops and short courses are encouraged on the preserve. Classes of students, local citizens groups and conservation organizations are among those making regular use of the area.

In the spring of 1979, a permanent Prairie Heritage Diorama will open at the Dacotah Prairie Museum in Aberdeen depicting the Ordway Prairie. This display will provide a means of further educating South Dakotans to their prairie heritage and will be helpful to individuals and groups planning to visit the Ordway Prairie.

THE DETERMINATION OF THE DISTRIBUTION OF PARTICLE SIZES  
IN FLOWERS OF SULFUR: AN EXPERIMENT FOR UNDERGRADUATE  
PHYSICAL CHEMISTRY LABORATORY

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One of the better techniques for the determination of particle size distributions involves the determination of the rate of sedimentation onto a balance pan from an initially uniform suspension. This technique has been adapted for use in undergraduate physical chemistry laboratory.

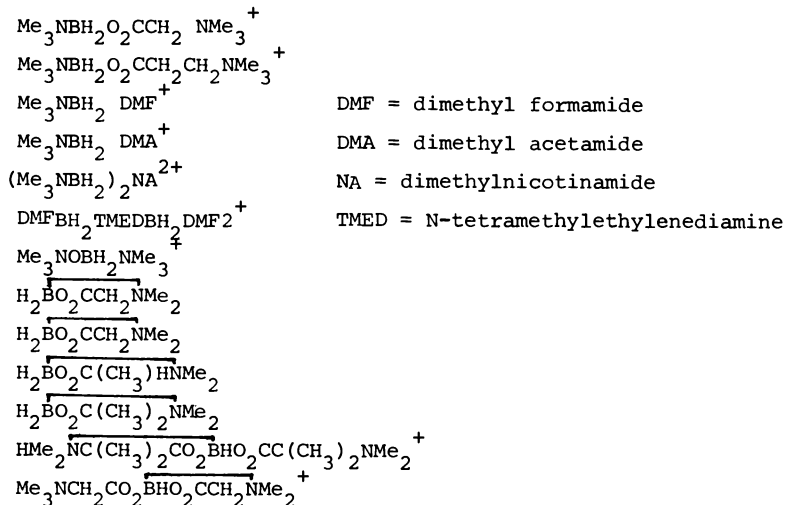
Sulfur particles are allowed to settle from a uniform aqueous suspension of flowers of sulfur onto a large planchet cup suspended beneath a top leading balance of 1 mg sensitivity. The weight of the sediment is determined as a function of time. The determination of the weight distribution function requires considerable data manipulation involving averaging and two separate numerical differentiations. The calculations are performed using an HP 9825A calculator and the distribution function is plotted using an HP 9872A plotter. The theoretical basis for the experiment, the experimental procedure, and the algorithms for the calculation will be presented.

## HYDROLYSIS OF BORANE CATIONS

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The hydrolysis of borane cations of the type  $\text{Me}_3\text{NBH}_2\text{OL}^+$  have been studied, and certain features of mechanism are becoming apparent. 1) There is second order dependence upon hydroxide; 2) A tetrahedral carbonyl site is implicated; 3) Activation enthalpy is about 8-12 kcal and activation entropy is strongly negative 20-40 eu; 4) There is only minor involvement of BH in the slow step; 5) Trace amounts of inhibitors can slow rate. Chromate and/or chromium inhibits; 6)  $\text{Me}_3\text{NBH}_2\text{O}^-$  or  $\text{Me}_3\text{NBH}_2\text{OH}$  are not likely early intermediates.

Cations and other related species examined are:



THE STRUCTURE OF BIS(2,2'-BIPYRIDYLAMINE-N,N')CHLOROCOPPER(1+)  
CHLORIDE TETRAHYDRATE

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The structure of bis(2,2'-bipyridylamine-N,N')chlorocopper(1+) chloride tetrahydrate has been determined by three dimensional x-ray analysis and refined by least squares methods to conventional residuals of  $R=0.081$  and  $R_w=0.096$ . The compound was precipitated from solution containing copper(II) chloride, and dipyridylamine in 1:2 stoichiometric amounts. Dark green rectangular crystals formed after two weeks as the solvent was allowed to slowly evaporate. The lattice parameters are  $a = 12.556(6)$ ,  $b = 28.002(7)$ ,  $c = 6.997(4)$  and  $\beta = 99.98(6)$ . The space group is  $P2_1/a$  with four molecular units per unit cell. A total of 5049 unique data with  $F_o \geq 3\sigma_f$  were used in the structure determination. The copper ion is pentacoordinate with a chloride ion and two pyridyl nitrogen atoms from different bipyridylamine ligands occupying the equatorial positions of a distorted trigonal bipyramid. The remaining two pyridyl nitrogen atoms occupy the axial positions. The copper-nitrogen distances range from 2.001 - 2.172 Å and the copper-chlorine distance is 2.334(4) Å. Carbon-carbon distances in the bipyridylamine complex average 1.38 Å. Carbon-nitrogen (pyridyl) distances average 1.34 Å. The carbon-nitrogen (aliphatic) average distance is 1.39 Å. The dihedral angles between pyridyl ring least squares planes within a bipyridylamine molecule are 23° and 37.5° indicating a high degree of flexibility of the bipyridylamine ligand molecule.

HEAT LOAD PREDICTION FOR POWER PLANT COOLING SYSTEMS

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Heat rejection from a power plant cooling system is always of interest to the regulatory agencies and environmentalists. The paper is intended to present a methodology by which the heat rejection rate under various conditions can be determined with the system component performance information. This methodology is different from that of the traditional prediction model which is generally based upon the theoretical principles.

The power plant cooling system consists of three components: condenser, cooling tower, and turbine exhaust end. Each component has its own performance characteristics. In case of a condenser, the condenser pressure is always expressed in terms of cold water temperature and condenser heat load. For a mechanical-draft cooling tower the performance curves are always prepared with the cold water temperature as the dependent variable and the cooling range, water flow rate and ambient wet bulb temperature as the independent variables. To indicate the steam turbine performance, the turbine net heat rate is frequently used. It is generally expressed as a function of condenser pressure and turbine output.

The methodology proposed in the paper is first to develop an empirical equation for each component in the cooling system by using the multiple linear regression and then, to solve the resultant nonlinear equations simultaneously. The calculation is straight forward but time consuming. Because of this, the generalized computer programs have been prepared for this purpose [1]. In general the error in this approach is well within the acceptable range. Table 1 indicates the maximum percentage of error.

To illustrate this proposed methodology, a case study is used. The inputs of this case study are:

Turbine - General Electric/TC2F/3600 RPM/2400PSIG/1000/1000F/252500 KWE/BEPT ;  
 Condenser - Foster Wheeler Energy Corporation/195813 Sq. Ft./Titanium 22 BWG/0.875  
 OD/92F/140342 GPM/40 Ft/ 2 passes; and

Mechanical Draft Cooling Tower - Marley Cooling Tower Company/ Model 6615-3-09/145000GPM. Some of the predicted results are indicated in Table 2. It is seen that for a given water flow rate the heat rejection from the power plant cooling system is strongly dependent with the turbine output. Since the mechanical-draft evaporative cooling tower is selected in this operation, the heat load is not sensitive to ambient air temperature as indicated.

In summary the proposed methodology directly makes use of the actual component performance data and the heat rejection thus predicted should be more accurate than that by the traditional mathematical model.

Table 1 Error Estimates

	Condenser	Cooling Tower	Turbine
Max. Error Percentage	1.8%	0.6%	0.9%

Table 2 Heat Load Estimation for Circulating Water Flow Rate 130500 GPM  
 Ambient Wet Bulb Temp. Percent Load of Generator Rating (252500 KW)

°F	Heat Load MBTU/h				
	20%	40%	60%	80%	100%
80	314.07	511.88	709.09	942.90	1172.91
75	309.30	506.29	705.04	939.96	1169.65
70	304.84	501.22	701.76	938.02	1166.89
65	300.74	496.66	699.13	936.88	1164.58
60	297.04	492.61	697.04	936.37	1162.65

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Canine Dental Annulation, Enamel Line, and  
Pulp Cavity Analysis of the American Badger  
with Reference to Aging

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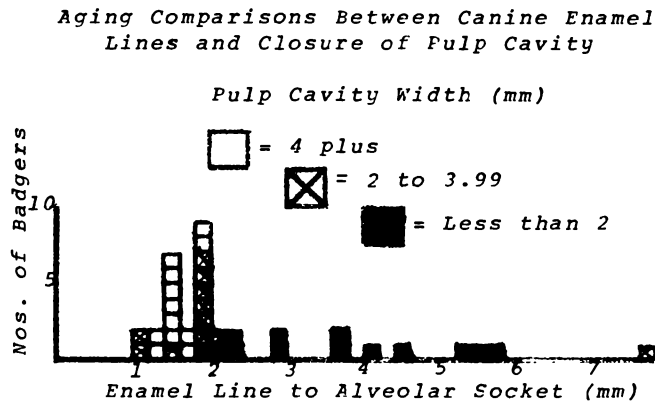
Relatively little has been done using dental characteristics to age American Badgers. Crowe and Strickland (1) suggested dental annulations as valid indicators of age in the American badger in south-east Wyoming. In order to compare and enlarge upon dental aging techniques in badgers particularly in reference to enamel lines; pulp cavity closure, and cementum annuli, skulls and canine teeth of 36 animals of unknown ages are presently being studied.

Upper canines were extracted and prepared following Allen (2) and Fogl and Mosby (4). Prior to extraction, alveolar socket to enamel line distance was measured (mm). Cross sections were cut with razors at the gum line. These were measured for width (longest axis). Sauer, et al. (3) studied black bears of known ages by canine analysis. They found that the pith (or pulp) cavity was less than 2.2 mm in 13 animals over three years old, that it was between 2 and 3.4 mm in four bears 2 to 3 year old and from 3½ to 8 mm in width for five animals between one and two years of age. Grue and Jensen (5) were able to separate only foxes of less than one year of age by pulp cavity size. Of 10 South Dakota badgers with pulp cavity diameters of less than 2 mm, all but one showed enamel line measurements to be greater than 2 mm.

MacPherson (6) used enamel live measurements to assign with confidence all classes in arctic foxes. Allen (2) found 88% of all juveniles of known ages had enamel lines of less than 2 mm. He found no adults with enamel lines of less than 2 mm.

Twenty-two of 26 juvenile badgers from South Dakota were found to have enamel line measurements of less than 2 mm.

In the American badger, root canal width appears inversely proportional to longevity, and, used with enamel line measurements, should prove to be another useful tool in partitioning age classes.



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LARGER LAKE THAN STREAM MUSSELS (BIVALVIA: UNIONACEA): AN  
EXCEPTION TO THE RULE IN NORTHWESTERN MINNESOTA

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It has been realized for some time that lake forms of mussels (*Bivalvia: Unionacea*) are generally smaller than river forms of the same species. Lake forms, too, are generally shorter, more inflated, thinner-shelled, and possess more distinct rest rings (1). This report documents an exception to the general rule of smaller lake forms, and points out other differences in two species of mussels from a stream and two lakes in northwestern Minnesota.

Collecting sites (August, 1977), all in Clearwater County, are on Lost River (SW $\frac{1}{4}$ NE $\frac{1}{4}$ NW $\frac{1}{4}$  sec. 32, T. 149 N., R. 38 W.; channel width averages 4m); Pine Lake (SW $\frac{1}{4}$ SW $\frac{1}{4}$ SW $\frac{1}{4}$  sec. 27, T. 149 N., R. 38 W.; east, exposed side of lake), and Lone Lake (SW $\frac{1}{4}$ NW $\frac{1}{4}$ NE $\frac{1}{4}$  sec. 1, T. 147 N., R. 38 W.; protected embayment). Average values of total alkalinity, pH, and specific conductance at the Lost River, Pine Lake, and Lone Lake sites for three analyses during September-October, 1977 are: 354, 162, and 110 mg/l; 7.8, 8.8, and 8.0; and 733, 358, and 238 micromhos/cm. Shells, with four or more rest rings, were measured for length (greatest distance parallel to the hinge line), height (greatest dorsoventral distance normal to the hinge line), and width (greatest distance across both valves normal to a plane passing between them).

Shells of *A. grandis* from Lost River are significantly ( $P=0.05$ ) shorter, lower (but not with respect to length), narrower (less wide) (Table 1), and less inflated (lower width/height and width/length ratios) than those from Pine Lake. Shells from Lost River are significantly ( $P=0.05$ ) shorter, lower (but higher in respect to length), narrower, and less inflated than those from Lone Lake. Shells from Pine Lake are significantly ( $P=0.05$ ) shorter, lower (but higher in respect to length), narrower, and less inflated (lower width/height, but not significantly different width/length ratios) than those from Lone Lake. Shells of *L. radiata* from Lost River are significantly ( $P=0.05$ ) shorter, lower (but higher in respect to length), and narrower than those from Pine Lake (Table 2), but may be more inflated (only width/length ratios are higher). Shell weights are significantly ( $P=0.05$ ) less for Lost River vs. Pine Lake, Lost River vs. Lone Lake, and Pine Lake vs. Lone Lake shells of *A. grandis* and Lost River vs. Pine Lake shells of *L. radiata*. The number of rest rings for shells of both species from the three water bodies are not significantly ( $P=0.05$ ) different.

Table 1

Statistical data of shell measurements for  
*Anodonta grandis* Say

Variable	N.	Mean ± S.E.	Range
<u>Lost River</u>			
Length(mm)	6	74±4.7	55-89
Height(mm)	6	43±2.5	35-53
Width(mm)	6	27±2.0	20-35
<u>Pine Lake</u>			
Length(mm)	50	91±1.0	80-115
Height(mm)	50	52±0.6	46-66
Width(mm)	50	35±0.4	29-45
<u>Lone Lake</u>			
Length(mm)	31	129±2.4	99-155
Height(mm)	31	71±1.3	54-86
Width(mm)	31	50±1.1	35-62

Table 2

Statistical data of shell measurements for  
*Lampsilis radiata* (Gmelin)

Variable	N.	Mean ± S.E.	Range
<u>Lost River</u>			
Length(mm)	50	67±0.9	52-81
Height(mm)	50	41±0.6	31-51
Width(mm)	50	23±0.4	16-29
<u>Pine Lake</u>			
Length(mm)	50	78±1.0	66-95
Height(mm)	50	46±0.6	39-54
Width(mm)	50	26±0.4	20-37

Smaller shells of *A. grandis* and *L. radiata* in Lost River (actually a creek or brook) may be because its small water volume, and perhaps fluctuating water level, results in a restricted food supply and lesser growth. Smaller shells of *A. grandis* in Pine Lake than in Lone Lake may be the result, in part, of greater wave exposure (2), and less time for feeding.

Caution should be exercised in extrapolating general habitat from relative mussel shell size in paleoecological studies.

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## KEPONE INHIBITION OF SUCCINIC ACID DEHYDROGENASE IN MOUSE LIVER MITOCHONDRIAL PREPARATIONS

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The chlorinated hydrocarbon insecticide Kepone<sup>R</sup> (decachlorooctahydro-1,3,4-metheno-2H-cyclobuta (cd) pentalen-2-one) is an inhibitor of mitochondrial ATPase (1), lactate dehydrogenase (2), and malate dehydrogenase (3). Our study was undertaken to determine the effect of Kepone on succinic acid dehydrogenase from albino Swiss-Webster mouse (Mus musculus) livers.

Two percent mitochondrial suspensions were prepared (4), and succinic acid dehydrogenase activity was assayed spectrophotometrically (5) at  $37.0 \pm 0.5^\circ\text{C}$  (pH 7.6) in the presence and absence of Kepone at various substrate (succinic acid) concentrations (0.66 to 1.3 mM).

Table 1

Kinetic Parameters for Succinic Acid Dehydrogenase at  $37^\circ\text{C}$   
 as Determined from Double Reciprocal Plots (by Linear Regression)  
 of Substrate Concentration Versus Reaction Velocity

Kepone Concentration ( $\times 10^{-5}$ M)	$K_m$ ( $\times 10^{-3}$ M)	$V_{max}$ (Absorbance Change/ Minute/mg Protein)	Correlation Coefficient
none	1.06	0.029	0.70
0.2	1.20	0.026	0.83
1.0	1.38	0.022	0.82
2.0	1.44	0.023	0.74

Succinic acid dehydrogenase activity was inhibited by micromolar concentrations of Kepone and the Michaelis constant ( $K_m$ ) increased with addition of Kepone suggesting competitive inhibition. However, the variability in maximum velocity ( $V_{max}$ ) suggested mixed inhibition with respect to substrate such as was reported for malate dehydrogenase in the presence of Kepone (3).

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The Role of Allelopathy on the Vegetational Composition of Disturbed Sites on the Samuel H. Ordway Memorial Prairie

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The Samuel H. Ordway Memorial Prairie is a twelve square mile tract of prairie in a near native state, located in McPherson County in north-central South Dakota. The Nature Conservancy owns and manages the prairie and has designated it as a model preserve. The goal of the Nature Conservancy is to preserve Ordway Prairie in its native state. However, scattered throughout the prairie are disturbed sites on which the native vegetation has been destroyed. The major sources of disturbance are cattle and human activity. Most of the disturbed sites are characterized by alien and weedy plant species, some of which are known to be allelopathic. The ability of allelopathic species to slow the rate of succession of prairie sites can be significant.<sup>1</sup>

The goals of this research are: to determine the vegetational composition of select disturbed sites on Ordway Prairie, and the relative importance of native versus weedy, possibly allelopathic species; to construct a disturbance scale based on the vegetational composition, and physical parameters; and to make recommendations regarding treatment methods to minimize effects of allelopathic species and promote the re-establishment of native species on the disturbed sites.

Corners of pastures were selected as representative of disturbed sites on the prairie. Twenty-five corners were randomly selected for study, during the period of June 12 to July 12, 1978. Sampling involved use of a 0.1 square meter quadrat device. Species presence and density were recorded for each quadrat. At each selected corner, a square plot was established by measuring 30 meters from the corner post along each fenceline. Total plot size for corners was 900 square meters. Within each plot, four random transect lines were established perpendicular to a randomly designated baseline fence. Along each transect, ten random points were established at which quadrat sampling was conducted. A total of 40, 0.1 square meter quadrat samplings, were taken for each corner.

Based on observations of plant species present and physical disturbance levels, a five-level disturbance scale was produced. Level one represents an undisturbed site with only native vegetation. Level five was a site with only alien or weedy species. Levels two, three and four represent gradations between these two extremes. Table 1 shows the predicted disturbance levels of the 25 study plots. As seen in Table 1, the accuracy of prediction for all plots by discriminate analysis was 74.4%.

Table 1 A Proposed Disturbance Scale

Disturbance Scale Level--description	Predicted Plot Membership	Accuracy of Prediction
1 no disturbance	0	-
2 slight dist.	10	74.8%
3 moderate dist.	11	78.0%
4 major dist.	4	65.0%
5 total dist.	0	-

Table 2 Mean Plant Number/Quadrat

Dist. level	Alien Plants	Native Plants
2	11.75	20.17
3	18.91	25.12
4	28.11	13.65

A total of 108 species of plants were found on the 25 plots sampled.<sup>2</sup> Of this total, 26 species were alien or introduced species. Of the 26 alien species, 6 are allelopathic. As seen in Table 2, level two plots exhibited the lowest number of alien species, 36.8%, while level three plots had 45.3% alien plants. Level four sites, considered the most seriously disturbed, were 68.5% alien in character. These findings support and clarify the proposed disturbance levels shown in Table 1.

The presence of alien species is a significant factor on disturbed sites. Several of the prominent alien species are known to be allelopathic, and others may also be allelopathic. The presence of these alien, allelopathic species can slow the growth and reintroduction of native plant species onto disturbed sites. Our recommendation involves the removal of known allelopathics in order to allow for regrowth of natives. This removal must be compatible to other efforts to maintain the native character of the prairie. Possible methods: controlled burning, selective fencing, herbicides, reseeding, and rest from grazing. Recommendations will be made on a plot by plot basis, depending on the problem species present.

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STUDIES ON THE DETERMINATION OF  
2,4-DICHLOROPHENOXYACETIC ACID AND  
2,4-DICHLOROPHENOL RESIDUES IN PLANT TISSUES

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The analysis of pesticide residues in plant samples poses two special problems to the analytical chemist. One problem is that of developing an analytical method which will remove 100% of the pesticide from the plant tissue. This is complicated by the presence of pesticide residues which are chemically bound (conjugated) to the plant. A second problem is the storage of the plant samples until the time of analysis. Ideally, no loss or chemical change of the pesticide should occur during storage.

Potato tubers and millet seeds obtained from plants which were field treated with 2,4-dichlorophenoxyacetic acid (2,4-D) were analyzed for residues of 2,4-D and 2,4-dichlorophenol (2,4-DCP). Two procedures were used; one in which the sample was extracted with an organic solvent, and a second in which the sample was subjected to an acid hydrolysis before extraction. Residue levels of 2,4-D and 2,4-DCP were determined by each procedure and compared. Untreated samples of potato tubers and millet seeds were fortified with 2,4-D and 2,4-DCP standards and stored at  $-20^{\circ}\text{C}$  for periods of up to 73 weeks. The potato tubers were stored in plastic bags and the millet seeds were stored in glass jars sealed with lids lined with aluminum foil.

An increase in residues of both 2,4-D and 2,4-DCP resulted when the samples were subjected to a one hour acid hydrolysis before extraction. Residue levels of 2,4-D in potato tubers increased from 0.083 ppm without hydrolysis to 0.108 ppm with hydrolysis. In millet seed, the change in 2,4-D residue levels was more dramatic, increasing from 0.042 ppm without hydrolysis to 0.262 ppm with hydrolysis. Residue levels of 2,4-DCP increased from 0.001 ppm to 0.003 ppm in potato tubers and from  $<0.020$  ppm to 0.026 ppm in millet seeds. Analysis of the samples stored at  $-20^{\circ}\text{C}$  showed essentially no loss of 2,4-D. At storage times of up to 73 weeks, 96.8% of the added 2,4-D was recovered from potato tubers. At up to 24 weeks storage, 101.2% of the added 2,4-D was recovered from millet seeds. Only 56.4% of the added 2,4-DCP was recovered from potato tubers stored 73 weeks at  $-20^{\circ}\text{C}$ . At up to 24 weeks storage, 104.0% of the added 2,4-DCP was recovered from millet seeds.

The increase in residues of 2,4-D and 2,4-DCP found by employing a hydrolysis step confirms the presence of conjugated pesticides in the plant and demonstrates the importance of developing an analytical method capable of removing all of the pesticide from the plant matrix. The loss of 2,4-DCP from the potato tubers was later determined to be due to the 2,4-DCP vaporizing out through the plastic bags (1). No loss of 2,4-DCP occurred from the millet seeds which were stored in glass jars. This demonstrates the importance of using proper containers for storage and of testing the storage conditions for each pesticide.

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TIPS On Improving Large Lecture Sections  
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Most courses are planned to fulfill several broad objectives. The measure of student success in reaching these objectives is normally demonstrated by performance on hour exams and final exams. While this testing method will indicate student progress, it often comes too late to allow the professor to concentrate on problem areas that arise for individuals or for large segments of the class.

The progress-as-you-learn format, the Keller Plan (1), can take care of problem areas but can become unwieldy and very time consuming. Computer assisted instruction (CAI) can be used for course individualization, but it can also become very time consuming. In addition CAI requires the availability of sufficient terminals to allow computer access to large numbers of students at any given time.

An alternate approach to using a computer to aid in the learning process is computer managed instruction (CMI). With the CMI system, only the professor, or an assistant, interacts directly with the computer. This eliminates much of the student vs. computer problem. TIPS or Teaching Information Processing System (2) is one approach to CMI.

TIPS works best in those subject matter areas where class objectives can be tested with objective-type questions. It has proven useful in the sciences where the subject matter is cumulative and sequential in nature, and the ability to assess student deficiencies early in the semester is particularly critical.

The use of TIPS requires the professor to prepare and give a series of short multiple choice surveys throughout the semester. The data processing equipment is used to print individual reports for each student based on his or her performance on TIPS surveys and to provide the professor with summary reports which indicate individual and class performance.

The reports that the professor receives allows rapid identification of problem areas for the class and for individual students. High-achieving students can also be identified before examinations and they may be provided with alternate projects or assignments.

To date, TIPS has been used in ten disciplines including accounting, chemistry, economics, engineering, environmental sciences, geology, literature, pathology, philosophy and psychology (3).

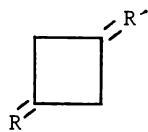
Through a grant from EXXon Education Foundation, the TIPS system has been purchased and is now available throughout the HECS computer network in South Dakota. On the University of South Dakota campus it is currently being tested in general chemistry and in a chemistry course for the health sciences.

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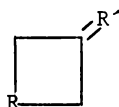
## ORBITAL INTERACTIONS IN SMALL RING SYSTEMS

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Ab Initio Gaussian 70 calculations (STO-3G) were carried out on 1,3-Cyclobutanedione (1) ( $R=R'=O$ ) and on 3-azetidinone (2) ( $R=NH, R'=O$ ) and related systems. The purpose in the cyclobutanedione series was to determine the extent of orbital interactions of the lone electron pairs on the

(1)

$R=R'=O, N, S, CH_2$

(2)

$R=NH, O, S \quad R'=O, N, S, CH_2$

respective heteroatoms and to compare these results with the observed ionization potentials obtained from He(I) source photoelectron spectroscopy. The calculated and observed PES data are summarized in Table 1. The calculated weighting factors for the non-bonding orbitals in the highest occupied molecular orbitals indicates a strong circumannular (through bond) interaction. The difference in energy between the two highest occupied molecular orbitals is about 0.5 eV.

In an attempt to understand the potential transannular (through space) interactions calculations were carried out on heterocyclobutanones. In the case of 3-azetidinone (2) ( $R=O, R'=O$ ) the highest occupied molecular orbital shows a large orbital weighting factor for the non-bonding atomic orbital on the cyclic heteroatom indicating very little transannular interaction. The observed carbonyl stretching frequency in the infrared supports the concept of minimal transannular participation in these systems. The stretching frequency does not change appreciably from that observed for cyclobutanone. One would expect the frequency to shift to lower energy as the transannular participation increased or as otherwise stated as the carbonyl chromophore became conjugated. (Table 2)

Table 1  
 Observed and Calculated IP's

Comp.	Observed IP(eV) ( $n_1+n_2-n_1-n_2$ )	Calc. IP(eV)
DIONE	0.63	0.56
bisimine	0.31	0.21
dithione	0.44	0.48

Table 2

Compound	Orbital Weighting Factor	C = OIR Freq.
cyclobutanone		1800
3-azetidinone	.95	1820
3-oxetanone	.902	1847
3-thietanone	.62	1768, 1731

All data indicate that circumannular (through bond) interactions are as large and perhaps as significant as the transannular (through space) interactions in cyclobutanedione and related systems.

THE INFLUENCE OF HIGH NEST SITE TEMPERATURE  
ON PARENTAL BEHAVIOR IN THE RED-WINGED BLACKBIRDG. W. Blankespoor\* and M. Ahrendt  
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In recent years there has been a great deal of interest in the manner in which polygynous mating systems have evolved. The usual conclusion has been that such a system confers a selective advantage when suitable breeding habitats are in short supply. This is thought to be so because it prevents the relegation of a certain number of females to inferior habitat. In this context, an adequate food supply and the availability of suitable nesting sites are most often cited as characterizing acceptable habitat. Suitable nest sites have typically been thought of as those which provide concealment from predators and protection from cold and rainy weather. In this study, we tested the idea that for red-winged blackbirds, breeding in mid-continental areas where extremes of temperature often occur, an additional attribute of suitable nesting sites is that they provide ameliorated mid-day microclimates.

Our approach was to observe parental behavior at nests whose nest temperatures were being simultaneously measured. The following were carefully noted during each observation period: duration (to the nearest minute) of each attentive period, duration of each inattentive period, behavior of female during attentive period and number of feeding visits made.

Under conditions of clear or partly cloudy sky, mean hourly ambient air temperature in degrees C was  $28.6 \pm 0.44$  SE (N = 44) while the corresponding value for mean hourly nest site temperature was  $36.2 \pm 0.73$  SE (N = 44). Females responded to increased nest site temperatures by increasing time spent at the nest in shading behavior. For all hourly periods during which the average nest temperature was 35°C or lower, the mean percentage attentive time was  $4.6 \pm 2.51$  SE while the comparable mean for nest temperatures 35°C and above was  $33.1 \pm 6.30$  SE. The most important possible consequence of protracted nest attentiveness is that nestlings are fed less often. Such was the case for the red-wing population in this study. The mean number of feeding trips per hour for all hours when females were attentive less than 25% of the time was  $10.6 \pm 0.86$  SE and the comparable mean for all hours when females were attentive for more than 25% of the time was  $7.8 \pm 1.08$  SE.

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## MOISTURE CONTENT ESTIMATION FOR LANDFILL REFUSE FROM SIOUX FALLS, S. D.

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The City of Sioux Falls is considering conversion from disposal of solid refuse in a landfill site to a process in which the refuse would be sorted into products suitable for compost and fuel production. Basically, the nonburnables and heavier fractions from the solid waste would be utilized for compost production while the burnables and lighter fractions would be fabricated into a refuse derived fuel (RDF). At the present, this technology is in its infancy. A number of problem areas have been defined and must be further investigated before the city can implement this new concept in waste management. One possible problem area is the moisture content of the RDF fraction of the solid waste. To respond to this need the SPM Group of Denver Colorado funded a preliminary investigation of this problem.

Fifteen samples from the solid waste being delivered to the Sioux Falls, S.D. landfill site were collected on November 27, 1978 and each sample was sorted into the following seven (7) categories: paper and cardboard; plastic; table scraps; ferrous and nonferrous metals; glass; and miscellaneous items not fitting these categories. Once sorted, the volume of each category was estimated and its actual weight determined. These weight-volume relationships for each category were used to translate the moisture content determined in this work into an estimated total moisture content for the total solid waste stream as inventoried earlier. Moisture determinations were made on a total of thirty-eight samples taken from the paper-cardboard, and table scrap categories. Moisture content of the samples was determined by weight loss after drying at 100 - 105°C, until no further weight loss was observed.

Prior to this study, the SPM Group completed an inventory of the solid refuse being delivered to the Sioux Falls Landfill Site. In this study, the weight, volume, and estimated composition by volume of the solid refuse stream was measured. The percentages of the waste categories of interest appear in Table I. Using the survey data, the weight-volume relationship, the moisture analysis (Table I), and literature moisture values (1-3), for the waste categories not determined experimentally, an estimated moisture content of 15.8 - 27.2%, including leaves, and 13.3 - 22.7%, excluding leaves, was made for the solid waste being delivered to the Sioux Falls landfill site. This estimate falls within the range found in the limited literature available (1-3) which indicates mean moisture content ranges of 15 - 40% for municipal solid wastes.

Because the mixture of solid waste is so heterogeneous, chief errors in moisture estimation will be made in the selection of representative samples. It is not possible at this time to place an error term on our moisture estimated, until a large number of samples can be collected from landfill sites under varying seasonal conditions. Our estimate of the error involved in reducing samples for drying (3%) could also be improved by further investigation. We are currently initiating further research to more accurately analyze solid waste moisture content, and to quantify the errors involved in the measurement.

Table 1

## Percentage Waste Composition and Percent Moisture Content

<u>Category</u>	<u>% Composition</u>	<u>% Moisture</u>	
Paper and Cardboard	26.9-29.6%	6.6-21.8%	1. United States Environmental Protection Agency (USEPA) Publication. (1972) EP 1.17:305 p. 31. 2. USEPA. (1972). High pressure compaction and baling of refuse. Final Rpt. pp. 33-35. 3. Bowerman, F. R. (1969) <u>Introduction to the Principles and Practices of Incineration</u> . Wiley Interscience, New York, p.7.
Wood	8.4%	10.0-20.0%*	
Rubble	15.1%	3.2% (5)	
Leaves	16.9%	28.6-50.0% (5)	
Logs	11.4%	40.0-60.0%*	
Food and Table Scraps	9.0-18.0%	37.2%	
Plastic	0-1.3%	2.0% (5)	
Tires	0.4%	1.2%	
Glass	0-17.0%	2.0% (5)	
Other Misc. (Metal, Paunch Manure, Sawdust, Stone, Park & Yard Burnable Stuff)	13.0%	0-10.0%	

\*Pers. comm. PFI Inc.

## A GENERAL HISTOLOGICAL STAIN FOR AXIS CYLINDERS AND CELL BODIES OF THE CENTRAL NERVOUS SYSTEM

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The Fink and Heimer (1967) stain for degenerating axons has been modified for normal axis cylinders and cell bodies. A single adaptation of the reducer solution coupled with an appropriate counterstain demonstrates the normal course of axis cylinders against a well defined background morphology. This technique avoids the undesirable traits of specific fixation methods, laborious embedding regimes or long, complex staining methods generally associated with this type of central nervous system procedure. This short communication represents an overview of the general technique. Specific protocols are available upon request to the authors.

**FIXATION:** A wide range of fixation methods and fixatives can be employed. Fixative solutions applied by means of vascular perfusion, preceded by washes of physiological saline which have been buffered and isotonicly balanced are recommended. Solutions containing aldehydes work well, however, there is a decrease in stain efficiency as glutaraldehyde concentrations are increased. Less than one percent glutaraldehyde in a total aldehyde concentration of three to five percent appears satisfactory. This laboratory routinely uses buffered ten percent formalin solutions.

**SECTIONING:** Two methods of sectioning can be applied in this technique. The first is frozen sectioning following cryoprotection. Dissected tissue is kept in a fixative solution containing 30% sucrose four to five days, frozen with dry ice and sectioned at 40-100  $\mu\text{m}$  with a sliding microtome. A method of even greater convenience employs the use of a tissue chopper or vibratome. Freshly fixed tissues are sectioned at 40-100  $\mu\text{m}$  immediately after primary fixation. Tissues are therefore not subjected to the freezing and cryoprotection and are handled more efficiently.

**STAINING:** This technique, as with nearly all silver impregnation methods, is subject to variability from many sources. Standardization is accomplished by utilizing a "trial run". Twelve sections are placed in a 3 x 4 factorial design thus allowing the determination of the optimum concentrations of chemical variables. The staining procedure involves mordanting with potassium permanganate and subsequent bleaching with hydroquinone-oxalic acid. Sections are stained in a uranyl-silver nitrate solution, then with alkaline buffered silver nitrate. The silver is reduced and developed in an acidic solution and fixed with sodium thiosulfate. The background color is then bleached with potassium ferricyanide and the sections are counterstained with azure-eosin. Consistent results are easily attainable using acid cleaned glassware and solutions made with deionized distilled water.

The stain as outlined exhibits the course of axis cylinders through the tissue. Axis cylinders are dark brown upon a nearly clear background. The counterstain reveals background morphology. Cell bodies and cell nuclei range from red to violet depending upon the pH of the stain. Electron microscopy of stained tissue demonstrates silver grains most commonly localized just exterior or interior to the myelin sheaths and aligned with microtubules within the axoplasm. Silver grains are also associated with nissl bodies within the cytoplasm which explains the occasional appearance of stained clumps in the soma cytoplasm. The time course of the complete protocol from live animal to stained tissue using frozen sections is less than 36 hours. This could be reduced to approximately 18 hours by utilizing a vibratome for sectioning.

This stain therefore provides a method for exhibiting axis cylinders in the central nervous system by a protocol which is efficient, easy, and readily adapted to perhaps any fixation method.

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The authors wish to thank Drs. Bruce Albright and John Oberpriller for their helpful comments and Debra Beck and Joy Brew for help with the typing of this communication.

## EVIDENCE FOR LATE WISCONSINAN CATASTROPHIC FLOODING IN THE SOURIS RIVER AREA, NORTH CENTRAL NORTH DAKOTA

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During the Late Wisconsinan deglaciation, glacial Lake Regina in Saskatchewan drained through the Souris melt-water channel (1). Reconnaissance field mapping suggests that the drainage was a single catastrophic event. Topographic evidence along the path of the flood between Lake Regina and glacial Lake Souris in North Dakota (channel bifurcations, divide crossings, and spillovers) indicates that the flood followed and overwhelmed previously existing drainage paths. Because both the erosional and depositional effects of the flood have not been subjected to later erosion or deposition, the event must have been rapid and non-repetitive.

In Saskatchewan, the Souris melt-water channel consists of an inner trench about 1200 m wide and as much as 60 m deep flanked by a high-level eroded surface up to 6 km wide on each side of the trench. In places, the inner trench bifurcates, becoming two parallel linear trenches of the same depth separated by a narrow ridge. The broad upper eroded surface is dissected by channels oriented parallel to the inner trench and is mantled by scattered thin lag deposits. The upper surface merges with the floor of Lake Regina in a zone about 8 km wide in the outlet area of the lake. The inner trench starts abruptly as a small gully in the outlet area, indicating that the drainage of the lake must have been accomplished by water flowing across the broad eroded surface at the outlet. Therefore, both the inner trench and upper surface had to have been eroded during the same event, and post-flood drainage from the lake could not have occurred or the inner trench would have been eroded headward into the lake basin.

Southeast of Estevan, Saskatchewan, the melt-water channel bifurcates into northeast and southeast trending branches. The southeast branch spilled across a divide into the headwaters of the drainage of the Des Lacs River. The upper Des Lacs channel consists of a broad flat channel which is a continuation of the upper surface of the Souris channel. An inner trench, which is 50 m deep at the mouth of the Des Lacs, starts abruptly in the broad shallow channel, indicating that water flowed over the surface for only a short period of time. The northeast branch of the floodwater flowed around a bend and then south into North Dakota along the course of a previously existing melt-water channel. At several places, floodwater spilled out of the flood channel and cut shallow channels in the till plain.

From the international border south to Minot, the floodwater eroded the deep, steep-walled Souris and Des Lacs melt-water channels. Just east of Minot, floodwater spilled out of the Souris channel at one point and flowed eastward toward glacial Lake Souris. After breaching the Souris channel, the floodwater eroded a plexus of anastomosing shallow channels east of Minot (Fig. 1). If water had spilled out of the Souris channel periodically in this area, a single channel to the Lake Souris basin would have developed. During the flood, the breached point of the Souris channel eroded to only 15 m above the present valley floor. Therefore, after the spillover, discharge must have dropped rapidly or subsequent spillovers at the same point would have occurred.

Channel deposits from the flood consist of huge constructional bars of coarse gravel located mainly at inside bends of valley meanders. These giant point bars range in height up to 40 m. They consist of coarse gravel and sand with generally obscure plane bedding and boulders up to 3 m in diameter. The bars extend to the floor of the present valley and possibly into the subsurface, indicating that the entire present channel was occupied by water during the event. Another type of deposit consists of streamlined, longitudinal ridges of gravel 5 to 10 m high in mid-valley positions. In at least one place, a longitudinal ridge has a bedrock or till core, indicating that the bar is in part an erosional remnant left during rapid deepening and widening of the valley. The longitudinal bars show no evidence of post-depositional erosion or modification; however, some point bars have two levels. The lower level may have been produced by declining discharge as the flood ended.

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Fig. 1. Air photo of Souris melt-water channel east of Minot (left) showing location of spillover (hachured lines). Arrow shows direction of water movement. Network of anastomosing channels in upper right corner of photo. North at top.

0 1 mile (1.6 km)

SEROLOGICAL SURVEY OF THE INCIDENCE OF OVINE  
PROGRESSIVE PNEUMONIA IN SIX NORTH DAKOTA FLOCKS

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Ovine progressive pneumonia (OPP), a chronic insidious, pulmonary disease of adult sheep, has a relentless course leading to progressive loss of weight, increasingly severe respiratory distress and death. The etiologic agent of OPP, progressive pneumonia virus (PPV), is an enveloped, spherical single stranded RNA virus classified in the family *Retroviridae*. This virus stimulates the production of antibodies in the host animal and antibodies are indicative of PPV-infection but apparently provide no immunity against infection.

The prevention and control of a disease is an important consideration in any livestock industry. The only means of controlling OPP is by isolation and eradication of infected animals. As yet there is no effective treatment. The purpose of this investigation was to characterize the extent of OPP infection in 6 separate sheep flocks in North Dakota. One flock will be additionally analyzed for the incidence of OPP according to age and breed. Identification of PPV-infected animals was by agar-gel immunodiffusion (AGID), a reliable simple serological method for the detection of precipitating antibodies against PPV (1).

A problem with AGID test is the production of a strong yet specific antigen. Concentrated antigen was prepared as previously described with the following exceptions; ovine trachea cells were employed for propagation of PPV instead of ovine lung cells and PPV antigen was concentrated by means of Amicon diafiltration rather than dialysis against polyethylene glycol. Using diafiltration, the concentrated antigen was easily quantitated, and less viscous than the dialysed antigen.

The entire sheep population of North Dakota State University (NDSU) was surveyed for precipitating antibodies against PPV by AGID. Of the 265 serums collected from sheep at the NDSU sheep barn 88 were positive and 177 were negative. These results include animals from five different breeds which ranged from 1 to 7 years in age (Table 1). Newborn lambs were not tested. Some breeds had higher proportions of positive serum than others; positive serums were most frequent in Cheviots (57%) and least frequent in Hampshires (21%). Considering the entire flock, irrespective of breed 33% of the animals had positive serums. In general, the proportion of sheep with positive serums increased with the age of the sheep. Only 20% of the yearling had positive serums, whereas 67% of the 7 year-olds were positive. An additional 150 serums were randomly collected from 5 commercial flocks throughout the state of North Dakota. The overall percentage of positive serums ranged from 18% to 85% in the different flocks. The ages and breeds were not known for the 150 serums.

The high incidence of OPP in sheep flocks is documented by researchers in Idaho (2), who reported an incidence of 58% in all ages and 90% in cull ewes with similar breed and age susceptibilities as reported in this study. The high incidence of OPP throughout the United States presents a major problem to the sheep producer. Additional knowledge into breed and age susceptibilities to OPP may lead to a better understanding of this disease and possible means of control.

TABLE 1. AGAR-GEL IMMUNODIFFUSION ANALYSIS OF SERUM SAMPLES. FOUNDATION FLOCK

Breed	Total No. tested	Age (years)							Total
		1	2	3	4	5	6	7	
Suffolk	90	10/37*	3/13	3/15	3/8	2/5	4/11	1/1	26/90
Hampshire	62	1/19	1/10	3/14	1/3	6/12	1/4	--	13/62
Rambouillet	43	3/11	3/12	4/9	3/4	2/3	0/2	1/2	16/43
North Country									
Cheviot	30	2/7	2/8	5/6	3/3	3/4	2/2	--	17/30
Columbia	40	1/11	2/9	4/6	1/3	3/5	5/6	--	16/40
	265	17/85	11/52	19/50	11/21	16/29	12/25	2/3	88/265

\* Number positive/number tested.

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POLYHALOPHENOLS: OXIDATION. SILVER SALTS OF POLYHALOPHENOLS;  
THERMAL DECOMPOSITION TO POLYPHENYLENE ETHER OXIDE POLYMERS

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As found by Hantzsch (1), and later in more detail by Torrey and Hunter (2), the silver and other metal salts (such as mercury and thallium) of polyhalophenols often exist in both white and colored modifications. They are, like other silver salts, very sensitive to light, especially while wet, and must therefore be prepared in the dark.

When heated in benzene or toluene suspension, or when treated with iodine, or slightly pinkish ethyl iodide, they decompose to yield silver halide and highly colored polyhalophenylene ether oxide polymers, very soluble in chloroform, but precipitated out in powder form on addition of benzene or alcohol. Deep blue colors appearing during the reaction are presumably due to free-radical intermediates. The alkali salts (Na and K) are colorless.

On oxidation by chromic acid or ferricyanide solutions the polyhalophenols lose an ortho or a para halogen atom to form ortho and para-polyhalodiphenoquinones. These reactions were studied in detail by Hunter and Wollett (3).

When several different halogen atoms are present, iodine splits out more readily than does bromine, which in turn is lost more readily than chlorine. Hunter and Joyce (4) in their studies also found that the para-halogen is lost in preference to the ortho-halogen.

The preparation of the 2,6-dibromo- and diiodo-4-methyl-, and of the 4,6-dibromo- and di-iodo-2-methyl-phenols, and of their alkali and silver salts, was described by Hunter and Rathmann (5). The 2,6-dibromo-, di-iodo- and bromo-iodo-4-fluorophenols, as well as the corresponding 2,6-dibromo-, diiodo-, and bromo-iodo-4-chloro-3,5-dimethyl-phenols were prepared in a similar manner from the 4-fluoro- and the 3,5-dimethyl-4-chlorophenols.

Thermal decompositions of the dibromo- and the diiodo-ortho- and para-cresol silver salts are similar to those of the trihalogen phenol silver salts, but yield besides the amorphous powdery polymers also considerable amounts of oily varnish-like substances, as well as alkali-soluble organic compounds other than simple phenols. The iodo-cresol silver salts are, however, more stable than the bromo salts. The para-cresol salts are more stable than the ortho-cresol salts. The methyl groups appear to activate the halogen atoms. While the methyl groups do by and large remain on the ring the amounts of silver oxide and of free silver formed in the decompositions of these silver dihalo-cresol salts are much greater than in the case of the silver salts of the trihalophenols. The silver salt of dibromo-para-cresol in particular is stable enough so that it reacts with silver iodide to give chiefly the phenetole,- 2,6-dibromo-4-methylphenyl ethyl ether rather than the amorphous polymeric polyphenylene oxides.

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TOTAL ECLIPSE OF THE SUN  
FEBRUARY 26, 1979

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The total eclipse of the sun on February 26, 1979 covered a path some 300 km. wide, starting at local sunrise 1200 km. out in the Pacific, and passing along the Washington-Oregon border, across Idaho, Montana, northwestern North Dakota, southern Manitoba, part of Hudson Bay, Baffin Island, ending at local sunset in northern Greenland.

Groups from the Moorhead-Fargo Astronomical Society, Concordia College, Moorhead State University, North Dakota State University, as well as some local high school and junior high schools, travelled to several points more or less along or near a line passing through Brandon and Riverton (on Lake Winnipeg), Manitoba. Equipped in various degrees with telescopes, electronic time recorders, photographic equipment, including movie, sun filters, etc. they set up observing stations. The principal ones, along with the relevant data, were:

Station	Concordia				Morris M.S.U.	Fargo
	A	B	C	D		
Event, or						
Longitude	96°59'	97°38'	99°50'	99°48'	97°21'	96°47'
Latitude	50°58'	51°06'	50°14'	49°44'	49°22'	46°52'
Distance from Center Line	3 km.	29 km.	34 km.	13 km.	128 km.	395 km.
1st Contact	9:38	9:37	9:33	9:34	9:35	9:32
2nd Contact	10:48:27	10:48:23	10:42:54	10:42:58	10:46:29	----
Center, or Max.	10:49:52	10:49:47	10:44:18	10:44:22	10:47:06	10:45
3rd Contact	10:51:18	10:50:11	10:45:41	10:45:47	10:47:43	----
4th Contact	12:05	12:04	11:59	11:59	12:02:35	12:02
Duration of Totality	2' 51"	2' 48"	2' 47"	2' 49"	1' 14.5"	----
						96.5%

A = Riverton. B = Fisher Branch. C = Minnedosa. D = Pepper (Brandon).  
Data taken, or extrapolated, from U.S. Naval Observatory Circular 157.

Other North Dakota groups from the colleges and schools in Bottineau (2' 29") and Minot (2' 03"), as well as other places were well-organized for eclipse observations.

ASYMBIOTIC NITROGEN FIXATION AND PHOSPHATE SOLUBILIZATION  
ON RECLAIMED STRIP-MINED LANDS IN WESTERN NORTH DAKOTA

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An investigation was conducted to determine the relative numbers of asymbiotic nitrogen fixing bacteria and phosphate solubilizing bacteria in the rhizosphere of a certain grass and in the soil adjacent to the plants as affected by the depth of the topsoil and subsoil over the strip-mine spoil.

Rhizosphere and soil samples were collected from a United States Department of Agriculture-Agricultural Research Service test plot (project No. M-74-7) located on the Glenharold Coal Mine near Stanton, North Dakota. The plot consisted of a wedge of subsoil material (B and C horizons), ranging in depth from 0 cm to 230 cm, spread over the impermeable sodic spoils. On top of the subsoil wedge was either 0, 20, or 60 cm of topsoil material (A horizon). Plant and soil samples were collected at subsoil depths of 0 cm and 140 cm for each of the topsoil depths mentioned. Soil samples consisted of soil cores 10 cm deep and plant samples consisted of similar cores trimmed to obtain a dense root mass plus adhering soil. The grass used in this study was crested wheatgrass (*Agropyron desertorum* var. Nordan).

Soil bacteria were isolated from soil and rhizosphere samples at the 6 sites of varying topsoil and subsoil depth mentioned. The bacteria were isolated by plating serial dilutions on a nonselective soil extract agar of Lochhead and Burton (3). Fifty-five isolates from each site were chosen at random and maintained on soil extract slants (660 isolates total). These isolates were tested for acetylene reduction (1) under atmospheric pO<sub>2</sub> in a basal nitrogen-free medium of Hino and Wilson (2) and for the ability to solubilize dicalcium phosphate in the plate test of Louw and Webley (4). Results were recorded as the percentage of isolates capable of reducing acetylene or solubilizing dicalcium phosphate at a particular site (Table 1).

Table 1

Site	Topsoil depth (cm)	Subsoil depth (cm)	% of isolates capable of reducing acetylene		% of isolates capable of solubilizing phosphate	
			Soil	Rhizosphere	Soil	Rhizosphere
1	0	0	12.73	29.09	1.8	3.6
2	0	140	9.09	34.55	5.4	0.0
3	20	0	12.73	29.09	5.4	12.9
4	20	140	29.09	41.82	4.8	21.7
5	60	0	61.82	69.09	14.8	15.7
6	60	140	54.55	63.64	29.6	25.7

A simple regression reveals that a significant functional relationship ( $P < .05$ ) exists between the percentage of acetylene reducers and the depth of the topsoil for the soil isolates. A significant relationship ( $P < .05$ ) was also found to exist between the percentage of isolates capable of solubilizing dicalcium phosphate and the depth of the topsoil for the soil isolates. No significant relationship was found between rhizosphere isolates and either topsoil or subsoil depth.

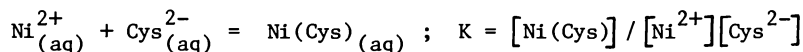
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## CHARACTERIZATION OF COMPLEXES OF CYSTEINE WITH METAL CATIONS

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The amino acid cysteine (Cys) forms complexes (chelates) with certain metal cations. For example with Ni(II) ions:



In this study, the complexes of cysteine with Ni(II), Pb(II), and Hg(II) were studied at 25, 35, and 45°C.

Since the metal ion complexes with the cysteine anion, the release of hydrogen ions is enhanced when the complex is formed, thus perturbing the acid dissociation equilibrium of cysteine. Acid-base titrations were done under nitrogen using 0.027 F cysteine hydrochloride, 0.476 F sodium hydroxide, and a 0.0049 F metal ion solutions. By analyzing the cysteine titration curve with and without the metal ions, the nature and extent of the complexation was elucidated.

The stepwise acid dissociation constants of cysteine, determined by sodium hydroxide titration with no metal ions present, are shown in Table 1 below.

At any time during the titration of cysteine in the presence of metal ions,  $[\text{Cys}^{2-}]$  and the average ligand number ( $\bar{n}$ ) can be calculated knowing the volume of base added, the pH at that point, the formal concentrations of cysteine and metal ion, and the cysteine acid dissociation constants (1). Once  $[\text{Cys}^{2-}]$  and  $\bar{n}$  have been determined for any point in the titration, K can be calculated using,

$$K = \bar{n} / (1 - \bar{n}) [\text{Cys}^{2-}]$$

The formation of  $\text{Ni}(\text{Cys})_2^{2-}$  is possible by reaction of  $\text{Ni}(\text{Cys})$  with a second  $\text{Cys}^{2-}$ ; however, consistent data on this reaction could not be obtained using this technique, perhaps due to the oxidation of cysteine to cystine at higher pH's. The equilibrium constants obtained for the formation of  $\text{Ni}(\text{Cys})$  are shown in Table 2 below.

The results indicate the temperature dependence of the constants. They are consistent with the expected results in that K decreases with increasing temperature, indicating the exothermic nature of the complex formation (2). It should be noted that no detectable cysteine complex was formed with Hg(II).

Table 1: Cysteine·HCl pK<sub>a</sub> Values

	25°C	35°C	45°C
pK <sub>a1</sub>	2.11	2.40	2.61
pK <sub>a2</sub>	8.28	8.00	7.83
pK <sub>a3</sub>	10.40	10.02	9.85

Table 2: log K for Cysteine Complexes

	25°C	35°C	45°C
Ni(II)	10.61	9.63	9.49
Pb(II)	14.48	*	*
Hg(II)	no complex	*	*

\*no data collected at these temperatures

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## SOLVENT EFFECTS ON ETHER-HCl ASSOCIATION REACTIONS

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The association of hydrogen chloride (an electron acceptor) with isopropyl ether or methoxybenzene (electron donors) in various organic solvents was studied using a solute isopiestic technique. An approximately 12 F aqueous solution of HCl, equilibrated in a closed chamber with the solvent, was used to establish a constant concentration of uncomplexed HCl in the solvent with and without the donor molecules (1). The formal solubility of HCl ( $f_{\text{HCl}}^0$ ) was linearly enhanced by the presence of ether of known formality ( $f_E$ ). As a first approximation, the increase in HCl solubility ( $\Delta f_{\text{HCl}}$ ) was attributed entirely to the formation of a 1:1 complex.



The 1:1 association constants ( $K_{11}$ ) were evaluated from the slope of plots of the following derived relationship:

$$\Delta f_{\text{HCl}} = K_{11} f_{\text{HCl}}^0 (f_E - \Delta f_{\text{HCl}})$$

Such plots are typically linear with zero intercept, supporting the assumption of the 1:1 complex. Thermodynamic functions were evaluated from the temperature dependence of the association constants using the van't Hoff relationship. Isopropyl ether complexes were characterized in several solvents of varying dielectric constant while methoxybenzene was studied in carbon tetrachloride alone.

The results shown in the table below are consistent with known chemical properties of the systems, agree with literature values for similar complexes (2), and exceptions to generalizations are explicable. Correlation of HCl solubility with dielectric constants of the solvents allows the 1:1 association constant for the benzene-HCl complex to be estimated ( $K = 0.17 \text{ M}^{-1}$ ). However, the formal solubility of HCl in the methylated benzenes was not found to follow the expected pattern (3).

Solvent (ether donor)	$f_{\text{HCl}}^0$ (298K)	$K_{11} (\text{M}^{-1})$ (298K)	$-\Delta H^0$ (kcal/mole)
1,2-dichloroethane (isopropyl)	0.102 $\pm$ 0.001	2.4 $\pm$ 1	5.1 $\pm$ 3
dichloromethane (isopropyl)	0.090 $\pm$ 0.001	3.4 $\pm$ 2	4.9 $\pm$ 2
chloroform (isopropyl)	0.055 $\pm$ 0.001	2.6 $\pm$ 2	5.5 $\pm$ 1
benzene (isopropyl)	0.094 $\pm$ 0.001	2.9 $\pm$ 1	3.8 $\pm$ 1
carbon tetrachloride (isopropyl)	0.037 $\pm$ 0.001	4.6 $\pm$ 1	5.7 $\pm$ 4
cyclohexane (isopropyl)	0.027 $\pm$ 0.001	5.6 $\pm$ 1	5.6 $\pm$ 1
carbon tetrachloride (methoxybenzene)	0.037 $\pm$ 0.001	0.57 $\pm$ 0.05	1.5 $\pm$ 6
toluene (no donor)	0.099 $\pm$ 0.001		
p-xylene (no donor)	0.097 $\pm$ 0.001		
mesitylene (1,3,5,-trimethylbenzene)	0.089 $\pm$ 0.001		

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## CHLOROPLAST DIFFERENTIATION IN LIGHT GROWN WHEAT SEEDLINGS

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Most of the studies on plastid differentiation have utilized dark grown tissue subsequently greened in continuous light. In these studies, all of the plastids are essentially at the same stage of development prior to the greening phase. Plastid differentiation in continuous light has been examined in tissues developing from the apical meristem of dicotyledonous plants (3). The activity of an intercalary meristem in monocotyledonous leaves provides the opportunity of studying plastid development on a rather continuous basis within a single leaf. Wheat was chosen for this cytological study due to crop importance, and to the active photosynthetic research program in our laboratory.

Wheat (*Triticum aestivum* L. cv. 'Waldron') seeds were germinated in a one inch layer of crushed granite and exposed to  $140 \mu$  Einstein  $m^{-2} sec^{-1}$  of continuous light at 25°C and 80% RH. Seedling growth was monitored by the method of Robertson and Laetsch (2). After five days sections of the first leaf were taken at 1 mm, 2 mm, 3 mm, 4 mm, 7 mm, and 15 mm from the seed surface. The tissue was fixed in 2.5% glutaraldehyde in phosphate buffer (pH 7.4), postfixed in 2%  $O_3O_4$ , dehydrated in a graded series of acetone, stained with a saturated solution of uranyl acetate in 70% acetone, and embedded in Epon-Araldite plastic. Samples were sectioned with a diamond knife on a Sorvall MT-2 ultramicrotome. The sections were stained with lead citrate and examined with an AEI Corinth 275 electron microscope at 60KV.

Meristematic cells are characterized by dense cytoplasm with abundant ribosomes, mitochondria, and dictyosomes. Plastids at this stage of development are small ( $0.89 \pm 0.04 \mu m$ ), have few grana ( $1.11 \pm 0.12$  grana per plastid section), and the grana are composed of few thylakoid membranes ( $2.28 \pm 0.06$  thylakoid membranes per granum). Some plastids also contain prolamellar bodies which are generally characteristic of dark grown tissue. Plastids from the oldest tissue examined (15 mm from the seed surface) are larger ( $3.41 \pm 0.12 \mu m$ ), contain more grana ( $11.49 \pm 0.71$  grana per plastid section), and have a greater number of thylakoid membranes per granum ( $3.87 \pm 0.07$ ). The prolamellar bodies previously observed were also not as prominent as they were in the younger tissue. Starch grains were apparent in plastids of all stages of development. Chloroplasts from tissue germinated and developed in continuous light were compared with plastids from tissue which was grown for 6 days in the dark and then greened in 48 hours of light (1). Plastids from the latter were larger ( $7.1 \pm 0.3 \mu m$ ), contained more grana per plastid section ( $32.0 \pm 2.1$ ), and a greater number of thylakoid membranes per granum ( $8.8 \pm 0.4$ ). Chloroplast differentiation in continuous light may involve granal increase due to new membrane formation or reorganization of material in the prolamellar body. In either case there is an increase in plastid size. Storage materials accumulated in dark grown tissue are rapidly transformed on exposure to light and form membranes at a faster rate than membrane synthesis in tissue developing in continuous light.

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## APPLICATION OF AGAR IMMUNE GEL DIFFUSION TO ROTAVIRUS DIAGNOSIS

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In the late 1960's, a Reovirus-like agent was first implicated in neonatal calf diarrhea. Since that time, rotavirus, as it is now known, has been isolated from neonatal calves, pigs, lambs, foals, as well as infants. Difficulty in propagating rotavirus in cell cultures has markedly hindered diagnosis of these viral infections.

The methods of choice presently for rotavirus diagnosis are direct immunofluorescent antibody test (FAT) and electron microscopy (EM) (1). Each of these techniques have their shortcomings. Autolysis of intestinal specimens often make FAT unreliable. Electron microscopy of fecal material allows direct examination, but EM is expensive and not readily available in most laboratories. It is the purpose of this investigation to examine agar-gel immunodiffusion (AGID) as an alternative method for diagnosis of Rotavirus infection.

The problem with AGID is the production of strong, yet specific antigen or antisera that will react with your test samples. For antigen preparation, Nebraska calf rotavirus was inoculated onto rhesus monkey kidney cell cultures propagated with Eagle minimal essential media containing 10 ug trypsin. After appearance of cytopathic effect (CPE), culture fluids were collected and stored at -20°C. After thawing culture fluids were centrifuged; the cellular pellet was first trypsinized and then along with the supernatant fluids was separated into two equal portions. The equal portions were concentrated by the following methods, dialysis against ethylene glycol and amicon ultrafiltration. Concentrations of antigen were approximately 100-fold. Antisera was collected from guinea pigs previously immunized with live virus. Additional serums tested included commercially prepared rotaviral antisera from two separate sources.

Agar gel immunodiffusion was conducted using 1% agarose in 0.05 M Tris buffer and 0.1% NaCl. The four antigen preparations were reacted against the antiseras previously mentioned. Of the four, the cellular concentrates produced better precipitin lines than the concentrated supernatant fluids. The best lines were produced from the Amicon ultrafiltration concentrated cellular antigen. Complete lines of identity were formed between the immunized guinea pig antisera and commercial Rotavirus antiseras.

In the past, rotavirus diagnosis has primarily been based on FAT and EM. Agar-gel immunodiffusion is a simple economical method used for the detection of antibodies and/or antigens. Preliminary results indicate that AGID can be applied not only for the detection of rotavirus antibodies, but also the detection of rotavirus antigen in diagnostic samples. A recent report comparing AGID, FA and EM (2) further suggests the imminent use of AGID as a diagnostic tool for detecting rotavirus infection.

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COUPLING IN ISOLATED WHEAT CHLOROPLASTS DURING GREENING

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Electron transport is coupled to the phosphorylation of adenosine diphosphate (ADP) in photosynthesis (1). Photosynthetic control (PC) is an indicator of coupling in isolated chloroplasts and can be calculated from the ratio of state 3, electron transport rate in the presence of ADP and inorganic phosphate ( $P_i$ ), to state 4, electron transport rate after ADP utilization (4). Plastids isolated from many plants exhibit low rates of electron transport (3) and poor coupling. We report buffering systems that give good electron transport rates and coupling for isolated wheat chloroplasts. Furthermore, wheat plastid and thylakoid disruptions were studied from electron micrographs.

Wheat (*Triticum aestivum* L., cv. 'Waldron') seeds were germinated in the dark in vermiculite at 25°C. After 7 days, the seedlings were exposed to 72 hr of light at 350  $\mu$  Einstein  $m^{-2}sec^{-1}$  for the synchronous development of chloroplasts. A 15 g sample of primary leaves were homogenized in a Sorvall blender at half-speed for 10 sec in 80 ml of solution containing: 0.3 M sorbitol, 10 mM KCl, 50 mM buffer (glycylglycine (GG) or Tricine, pH 7.8 or 8.4), and 1% bovine serum albumin. In some studies, 5 mM of ethylenediaminetetraacetate (EDTA) or ethyleneglycoltetraacetate (EGTA) were added to the isolation medium. The macerate was filtered through miracloth, was centrifuged at 270  $\bar{g}$  for 2 min, and the supernatant was recentrifuged for 5 min at 3000  $\bar{g}$ . The chloroplast pellet was resuspended in a solution containing: 0.3 M sorbitol, 10 mM KCl, and 50 mM buffer (pH 7.8 or 8.4). Plastid electron transport was monitored using a Clark oxygen electrode in a 5 ml reaction vessel exposed to saturating red light. The reaction was measured at 25°C in a medium consisting of 0.3 M sorbitol, 2 mM  $MgCl_2$ , 2.5 mM KCl, 0.1 mM methyl viologen, 0.5 mM  $NaN_3$ , 10 mM  $K_2HPO_4$ , 50 mM buffer (pH 7.8 or 8.4), and 60  $\mu g$  Chl  $ml^{-1}$ . Electron micrographs of isolated plastids were prepared using a previously described technique (2).

The state 2 electron transport rate in absence of ADP, was stimulated upon addition of 1000 n mole of ADP to the 5 ml reaction vessel to give high state 3 rates when wheat plastids were isolated at pH 8.4 in GG with EGTA or in Tricine (Table 1). PC was equally high when wheat plastids were isolated in GG (pH 8.4) with EGTA or in Tricine (pH 8.4). GG (pH 8.4) was not as good a buffer with EDTA or when lacking EGTA in the isolation medium for the PC and 3-2/2 values were low relative to the values obtained with GG and EGTA. When wheat plastids were isolated in GG at pH 8.4, the methylamine uncoupled electron transport rates were reduced compared to the rates of plastids isolated in Tricine. Wheat plastids gave higher PC values when isolated at pH 8.4 than at pH 7.8.

Table 1. Electron transport of isolated wheat chloroplasts, pH 8.4, using different buffer systems. Each value represents the mean of 23 replicates with the standard error of the mean.

Chloroplast isolation medium	State 3-State 2 <sup>1/2</sup> State 2	Photosynthetic control (PC)	Methylamine (uncoupled)
			$\mu eq e^-/mgChl/hr$
Glycylglycine (GG)	<sup>a</sup> 76.1 $\pm$ 3.1	<sup>ab</sup> 2.38 $\pm$ 0.05	311.6 $\pm$ 22.0
GG + EDTA	<sup>a</sup> 81.8 $\pm$ 3.8	<sup>a</sup> 2.33 $\pm$ 0.05	339.6 $\pm$ 9.4
GG + EGTA	<sup>c</sup> 110.0 $\pm$ 3.2	<sup>bc</sup> 2.54 $\pm$ 0.11	290.6 $\pm$ 14.2
Tricine	<sup>c</sup> 121.9 $\pm$ 4.6	<sup>c</sup> 2.62 $\pm$ 0.04	384.4 $\pm$ 11.8
Tricine + EDTA	<sup>b</sup> 107.0 $\pm$ 3.0	<sup>bc</sup> 2.55 $\pm$ 0.04	374.0 $\pm$ 4.8
Tricine + EGTA	<sup>b</sup> 108.0 $\pm$ 3.2	<sup>c</sup> 2.62 $\pm$ 0.06	393.8 $\pm$ 10.8

<sup>1/2</sup> State 2 rates were about 100 u equivalent of electron transported/mgChl/hr.  
<sup>abc</sup> Values with similar letters are not significantly different at the 5% level according to Duncan's Multiple Range Test.

Table 2. Effect of pH on the electron transport activity of isolated wheat and spinach chloroplasts in the presence of EGTA. Each value is the mean of 12 replicates and standard error of the mean.

Plastid type/ isolation medium	Electron transport rate			Ratio of States S3-S2/S2 PC		
	State 3	State 4	Repeat S 3			
	$\mu eq e^-/mgChl/hr$					
Wheat						
Tricine, pH 7.8	74.2 $\pm$ 2.0	163.2 $\pm$ 7.4	91.8 $\pm$ 1.8	149.4 $\pm$ 5.2	107.5 $\pm$ 4.2	2.0 $\pm$ 0.1
Spinach <sup>1/2</sup>						
Tricine, pH 7.8	139.2 $\pm$ 3.4	268.8 $\pm$ 12.0	102.2 $\pm$ 3.8	221.8 $\pm$ 16.6	96.0 $\pm$ 8.8	2.6 $\pm$ 0.1
GG, pH 8.4	142.4 $\pm$ 3.6	218.6 $\pm$ 7.4	100.6 $\pm$ 4.0	217.2 $\pm$ 9.2	52.1 $\pm$ 3.8	2.2 $\pm$ 0.1

<sup>1/2</sup> Spinach was purchased from a local market.

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## CONTAINER-GROWN FORB TRANSPLANTS FOR MINED LAND RECLAMATION

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The restoration of the perennial forb component of grassland vegetation may be an important consideration wherever native grassland vegetation is proposed as the desired cover to be established on previously mined land. In previous studies it was found that the normal forb component of this vegetation was not readily reestablished. This study was undertaken to determine whether the use of container grown transplants might provide an advantageous method of establishing perennial forbs on mine spoil material.

Seeds of 31 native forb species were collected in the summers of 1976 and 1977 in the general vicinity of the Dickinson Experiment Station in southwestern North Dakota. Seedlings for transplant were grown in the greenhouse in each of the two winters of the study, and were transplanted in early summer to artificially constructed spoil piles at the Dickinson Station. Seedlings for transplant were grown in normal potting soil in styrofoam block containers for the 1977 season and in 3-inch peat pots for the 1978 season. At the same time the transplants were made, direct seedings of the forb species were made on small plots on the mine spoil material for comparison with the container-grown plants. Supplemental watering was used in the establishment of both transplants and seedings.

The results of the study showed that the characteristics of the spoil material itself was not a major barrier to the establishment of the native forbs either from transplants or straight seedings. The peat-pot transplants showed somewhat greater vigor than the transplants from the styrofoam blocks, with the transplants of both types generally showing more rapid and vigorous first year growth than the plants from the straight seedings. The relatively late emergence and slow development of plants from direct seedings would place these plants at a competitive disadvantage as compared to the container-grown transplants.

Forb species which showed exceptionally vigorous growth as transplants in both seasons included white prairie clover (Petalostemum candidum), prairie coneflower (Ratibida columnifera), wild licorice (Glycyrrhiza lepidota), stiff goldenrod (Solidago rigida), purple prairie clover (Petalostemum purpureum), and white upland aster (Aster ptarmicoides).

Species which showed exceptionally good emergence from direct seeding and subsequent vigorous growth included white prairie aster, wild licorice, prairie coneflower, blazing star (Liatris punctata), and long-leaved milkvetch (Astragalus ceramicus). Only a few species of those tested appeared to show little or no promise for use in revegetation of mine spoil materials. Limitations of the trial were that the forbs were grown without competition from a grass vegetation component, and the immediate environmental conditions under which the forbs were grown were not as severe as those potentially existing on spoil banks on previously mined lands in western North Dakota.



CLASS DETERMINATION OF IMMUNOGLOBULINS IN SERUM,  
COLOSTRUM AND NASAL SECRETIONS FROM PROGRESSIVE  
PNEUMONIA VIRUS-INFECTED SHEEP

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Progressive pneumonia is a "slow" virus disease of sheep caused by an agent antigenically and morphologically related to the visna-maedi viruses; the disease is chronic, prolonged and invariably fatal. Previous studies have demonstrated high levels of virus-specific antibody in infected sheep; however, antibodies acquired as a result of infection did not prevent death. This may be a result of provirus formation in infected cells (1). The provirus, integrated into host DNA, is not subject to attack by immunoglobulins.

Successful immunization against this virus will likely depend upon prevention of initial infection and provirus formation. Since the primary site of infection is respiratory tract, secretory immunoglobulins present in respiratory mucous may play an important role in this immunity.

This study was initiated to develop methodology for class identification and quantitation of virus-specific immunoglobulins in secretions and serum of sheep. Previous studies had not resolved whether sheep produced a secretory immunoglobulin related to IgA; Heimer et al. (2) found three antigenically distinct immunoglobulins in ovine colostrum and identified two of them as IgA subclasses. However, Sullivan et al. (3) detected IgM, IgG and a 10S immunoglobulin antigenically identical to IgG; no IgA was detected.

To determine the classes of immunoglobulins present in progressive pneumonia-infected sheep, serum and colostrum collected from these animals were fractionated by column chromatography and analyzed by immunoelectrophoresis, discontinuous electrophoresis and ultracentrifugation. Sephadex G200 resolved three serum and colostrum components which reacted with rabbit anti-sheep IgG (specific for L and H chains). A 19.5S molecule identified as IgM, was present in both serum and colostrum; 6.9S IgG was also detected in both samples. The third immunoglobulin, an 11S molecule, was antigenically identical to IgG; however, it was considerably heavier and showed less cathodic migration (Fig. 1).

Serum treated with rivanol for immunoglobulin enrichment and fractionated on DEAE cellulose was also resolved into three immunoglobulin components; this treatment produced 5S and 6.9S molecules which bound DEAE in addition to IgG which did not bind the resin in 0.01 M phosphate (pH 8.0); the 6.9S molecules eluted first after application of the NaCl gradient while 5S units eluted last.

Heavy chains were prepared from IgM and IgG by reduction in 0.1 M 2-mercaptoethanol, alkylation in 0.02 M iodoacetamide, and separation on Sephadex G100 prepared in 1.0 M propionic acid. Anti-sera specific for each species of heavy chain will be used to determine the class of the 11S molecule; it seems likely that this represents a polymeric IgG and that IgA, if present, is produced in only minor amounts.

1. Weiss, M.J. et al. (1975) J. Gen. Virol. 29, 335-339.
2. Heimer, R., D.W. Jones, and P.H. Maurer. (1969) Biochem. 8, 3937-3944.
3. Sullivan, A.L. et al. (1969) J. Immunol. 103, 334-344.

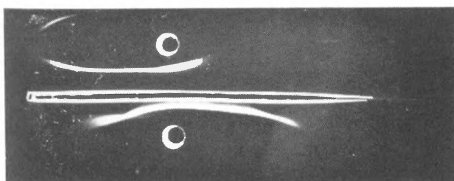


Figure 1. Immunoelectrophoresis of sheep immunoglobulins. Top-6.9S IgG; bottom 11S molecule. Trough-rabbit anti-sheep IgG. Anode to right.

BIOCHEMICAL CHARACTERIZATION OF HEMOLYMPH PROTEINS FROM TWO *HELIOTHIS* SPECIES AND A STERILE HYBRID

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*Heliothis virescens*, the tobacco budworm, is a pest of certain economically important crops including cotton. Larval infestations were previously controlled by chemical methods; however, environmental considerations and the appearance of strains highly resistant to chemical pesticides have necessitated the development of alternative control methods. One such method, applied successfully to other insect pests (1), is the release of sterile individuals which compete with wild type insects for mating. Previous studies have demonstrated that male progeny of *Heliothis virescens* ♂ x *H. subflexa* ♀ crosses are sterile, and when female progeny are backcrossed to *H. virescens* males, they continue to produce sterile males through at least 60 backcross generations (2, 3).

Before these backcross larvae can be safely introduced into native populations, methods are needed to specifically identify and monitor their populations. This study examined hemolymphs from backcross and parent insects to determine whether a unique protein existed that would serve as an immunological marker for backcross larvae.

Hemolymphs were first analyzed by discontinuous acrylamide electrophoresis; *H. subflexa* hemolymph yielded a protein pattern readily distinguished from *H. virescens* and the backcross (Fig. 1). Since *H. virescens* and backcross hemolymphs were very similar, a two-dimensional analysis was conducted to determine whether subtle differences would be detected with this increased resolution. Hemolymphs were first electrophoresed in acrylamide; slices were then cut from the gel and placed at the origin of an isoelectric focusing slab and proteins were focused into the second dimension. Results from this analysis also indicated that *H. virescens* and backcross larvae are very similar (Fig. 2); no unique protein was resolved in backcross hemolymph.

A further study employed SDS-acrylamide electrophoresis to determine the effect of developmental stages on hemolymph proteins. As each species progressed through the fifth instar, protein content and diversity increased. These data are consistent with previous reports (4) and indicate that analysis of *Heliothis* hemolymphs must be conducted on a well-defined developmental stage of the larvae. Insects late in the fifth instar, characterized by fatty swollen prolegs, were used in the one- and two-dimensional analyses performed in this study.

1. Bushland, R. C. (1974) *Science* 184, 1010-1011.
2. Pair, S. D., Laster, M. L., and Martin, D. F. (1977) *Ann. Entomol. Soc. Am.* 70, 665-668.
3. Martin, D. F. Personal communication.
4. Wyatt, G. R., and Pan, M. L. (1978) *Annu. Rev. Biochem.* 47, 779-817.

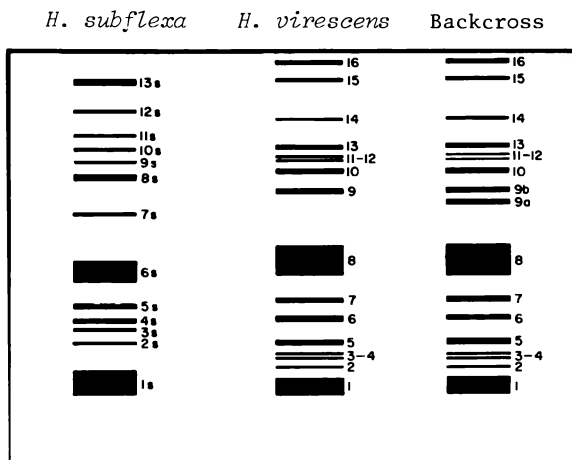


Fig. 1. Disc electrophoresis of larval hemolymphs. Anode (+) at bottom.

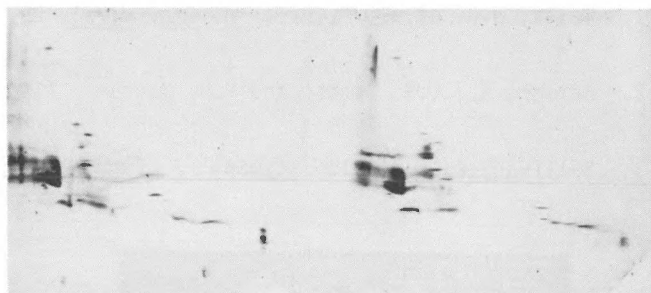


Fig. 2. Electrofocused second dimension of electrophoretically separated larval hemolymphs from *Heliothis virescens* (V) and a sterile backcross (B). First dimension anode (+). Second dimension pH 9.5-3.5 top to bottom.

A COMPARISON OF SERUM NEUTRALIZATION AND AGAR-GEL IMMUNODIFFUSION  
FOR THE DETECTION OF PSEUDORABIES ANTIBODY

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Pseudorabies has been recognized since the early 1900's as a disease in baby pigs and on occasion in other animals exposed to infected pigs. The etiologic agent involved is pseudorabies virus (PRV), a member of the Herpes group. Recently, increased incidence and severity has caused major economic losses to the swine industry.

With the rising incidence of PRV infection in swine, authorities throughout the midwest have required routine testing of transported animals. PRV infection may be transmitted by animals without clinical symptoms. These animals may harbor virus and transmit the disease; this phenomenon is known as latency. The presence of serum antibody is taken as proof of exposure and possible latency. Animals with positive PRV titers can not be transported across state lines and in many states are either quarantined or destroyed. As a result a rapid, accurate, economical serological test is desperately needed.

This study was initiated to compare the most commonly used method, microtitration serum neutralization (SN) and the agar-gel immunodiffusion (AGID) test using two different antigen preparations. One antigen was developed at the National Animal Disease Laboratory at Ames, Iowa (1), and one was developed at North Dakota State University, Veterinary Science Department (2).

Five hundred fifty-nine serums were analyzed by both methods. The results are shown in Table 1. Bacterial contamination or toxicity prevented the use of SN in the case of 34 serums. This illustrates an advantage of the AGID test; serums unsatisfactory for SN whether contaminated, grossly hemolyzed, or toxic can be tested. Additionally, serum volumes in the range of 25 to 75  $\mu$ l can not be tested by SN, but can be tested by AGID. It should be noted that SN did produce more positive results than did AGID.

Using NDSU antigen, 316 of the 559 serums were positive for PRV antibody by AGID, a 97.1% correlation with SN. Using NADL antigen, 300 of the 559 serums were positive for PRV antibody, a correlation of 92.4% with SN.

Serum neutralization is the recognized diagnostic test for the detection of PRV antibody. The AGID test offers many advantages; first, it permits testing of contaminated, hemolyzed or toxic serums. Secondly, AGID may be performed in the field, since it does not require specialized techniques, trained personnel and/or facilities, (e.g. sterile conditions, incubator space, hoods and tissue culture equipment) which are all necessary for SN. Thirdly, AGID yields preliminary results within 24 hours with final readings at 48 hours. This compares to a minimum of 48 hours before SN results can be read. The difference in the percent correlation (97.1% and 92.4%) for the NDSU antigen preparation and the NADL antigen preparation is not known.

The AGID test is a rapid, sensitive and economical test for the detection of PRV antibody.

TABLE 1: Comparison of SN and AGID Results

Test	Number Positive	Number Negative	Total
SN	320	205	525
AGID	316	243	559

1. Gutekunst, D.E., Pirtle, E.C., Mengeling, W.C. (1977) Am. J. Vet. Res. 39, 207-211.
2. Pfeiffer, N.E., Schipper, I.A. Am. Assoc. Vet. Lab. Diag. Mpls., 33-45, Oct., 1977.

THE SEASONAL PHOTOSYNTHETIC BLOOMS IN BREWER LAKE, N. D.,  
DERIVED FROM FIVE YEARS DATA

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The seasonal photosynthetic rates and correlated chemical and physical data were measured over five years, 1974 through 1978, in Brewer Lake, Erie, N. D. in order to determine the number and extent of the photosynthetic blooms occurring during the year.

In each of the five years data there was a period of minimum photosynthetic activity very near the end of May or early in June, indicating the end of the spring pulse. Since this period of minimal activity was a clear indication of the end of one pulse and the beginning of another, it was possible to work backwards in time from this minimum, summing-up the equivalent rates from each of the five years and a mean rate was calculated from these. The same process of summation of equivalent dates was applied to all the data following the late-May early-June minimum. From the mean of the five years data, a single composite curve was constructed for not only the photosynthetic rates but also for all parameters measured. While all the data measured contributed to the ensuing analysis, the chlorophyll-*a* data were especially useful in elucidating the number and extent of the photosynthetic pulses. When the chlorophyll-*a* composite curve for the five years is superimposed on the composite photosynthesis curve, the two are closely aligned and their data are correlated highly significantly,  $r = 0.676$  ( $N = 25$ ,  $P = 0.001$ ). Two photosynthetic pulses are demonstrated by these two curves alone and the remaining chemical and physical data reinforce this.

Pulse I occurs from mid-April to about 1 June. Pulse II occurs from near 1 June to about 1 October. The dates of occurrence varied over the five years by about a week. From this analysis it was possible to establish the approximate dates and magnitude of the pulses for each of the five years. The photosynthetic pulses for the five years are compared with those of the composite in the following table:

	Pulse ( $g\ C\ m^{-2}$ )			Ratios (%)		
	I	II	Total	I/II	I/Total	II/Total
1974	34	199	236	17	14	84
1975	33	230	263	14	13	87
1976	29	93	122	30	24	76
1977	49	154	203	30	24	76
1978	36	139	175	26	21	79
Composite	38	167	205	23	19	81

## POSSIBLE ROLE OF SYMPATHETIC NERVOUS SYSTEM IN REGULATION OF ASCORBIC ACID PRODUCTION IN THE RAT

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This experiment was conducted to determine if the sympathetic innervation of the liver plays a role in increased urinary ascorbic acid due to certain pharmacological agents. A selective chemical sympathectomy of the liver was performed on nine male Holzman rats (200-250 g) by intraportal injection of 6-hydroxydopamine at a dose of 50 mg/kg<sup>1</sup>. Nine other males were sham-operated. After a week recovery period the two groups were placed on differential diets. Six rats, three from each group, were fed normal ground chow, six were fed ground chow treated with 500 ppm o,p'-DDT and six were fed ground chow with 1000 ppm barbital. Urine was collected every 24 hours for seven days and analyzed for ascorbic acid. Liver glycogen was determined at the end of the seventh day. Urinary ascorbic acid increased in both the sympathectomized and sham-operated rats following treatment with o,p'-DDT.

Table 1

Urinary Ascorbic Acid  
 (Micrograms/24 hr)

Group	Day of Treatment					
	0	1	2	4	6	
Sham-Operated:						
Control	1,372 ± 951*	313 ± 307	1,276 ± 1,901	183 ± 318	442 ± 491	
o,p'-DDT	Trace	8,734 ± 6,155	9,192 ± 4,267	15,570 ± 6,250	17,252 ± 2,839	
Barbital	Trace	Trace	1,071 ± 1,855	4,787 ± 1,435	6,272 ± 2,549	
Sympathectomized:						
Control	Trace	Trace	68 ± 96	Trace	241 ± 133	
o,p'-DDT	415 ± 383	1,323 ± 2,291	4,914 ± 4,275	4,912 ± 6,393	14,557 ± 4,809	
Barbital	Trace	Trace	5,944 ± 4,307	4,263 ± 3,845	7,497 ± 2,860	

\*Mean ± Standard Deviation

The increase in urinary ascorbic acid in the sympathectomized rats was significantly lower than that of the sham-operated group. The barbital treatment also caused an increase, although the rise was somewhat lower than with o,p'-DDT, and no significant group difference was apparent. Sympathectomy caused a decrease in liver glycogen. Treatment with o,p'-DDT and barbital seemed to cause a further liver glycogen reduction. These data suggest that more than one mechanism is responsible for increased urinary ascorbic acid. The mechanism(s) by which o,p'-DDT causes this increase appears to involve, in part, sympathetic stimulation of the liver.

1. Cote, M.G., Blouin, A. and Brodeur, J. (1975) Proc. Can. Fed. Biol. Soc. 18, 102. Effect of chemical sympathectomy of rat liver with 6-OHDA.

THE INFLUENCE OF HORMONES,  $\alpha$ - AND  $\beta$ -ADRENERGIC EFFECTORS ON CARBOHYDRATE METABOLISM IN ISOLATED RABBIT HEPATOCYTES

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Huibregtse *et al* (1) reported in 1977 that isolated, perfused, rabbit livers convert 10 mM L-lactate to glucose and that glucagon, epinephrine and cyclic AMP stimulate lactate's conversion to glucose 1.6 to 1.8 fold. For a variety of reasons, we continued our studies in isolated hepatocytes.

Hepatocytes were isolated from livers from fasted rabbits by modifications of the methods of Berry and Friend (2) and Hutson *et al* (3). Trypan blue was routinely excluded by 90 to 95% of the cells.

Cells were incubated in oxygenated buffer containing 10 mM L-lactate in the absence and presence of various effectors. Reactions were initiated by addition of cells and terminated with  $\text{HClO}_4$ . Samples taken either before or after deproteinization were analyzed for their content of glucose, glycogen and cyclic AMP.

Data in Table I indicate that glucagon, epinephrine (EPI), dibutyl cyclic AMP (DcAMP), and the  $\beta$ -adrenergic agonist isoproterenol (ISO), all significantly increase both gluconeogenesis and glycogenolysis. The  $\beta$ -adrenergic antagonist propranolol (PROP) essentially eliminates both of isoproterenol's effects and epinephrine's glycogenolytic effect. Epinephrine's gluconeogenic effect is only partially negated. The  $\alpha$ -adrenergic antagonist phentolamine (PHENT) exerts essentially no influence on epinephrine-mediated gluconeogenesis or glycogenolysis.

Data demonstrate that both gluconeogenesis and glycogenolysis in rabbit hepatocytes can be regulated by glucagon, epinephrine and cyclic AMP. Both the gluconeogenic and the glycogenolytic effects of epinephrine appear to be primarily  $\beta$ -adrenergic in nature. Data obtained with rabbits contrast with observations of epinephrine's effects in rats where both gluconeogenesis and glycogenolysis are considered by various investigators to be either mixed or predominantly  $\alpha$ -adrenergic in nature.

TABLE I

Data on Gluconeogenesis and Glycogenolysis from Isolated Hepatocytes

ADDITION	GLUCONEOGENESIS <sup>1</sup>	STIMULATION <sup>2</sup>	GLYCOGENOLYSIS <sup>1</sup>	STIMULATION <sup>2</sup>
10mM Lactate (9) <sup>3</sup> +10 <sup>-8</sup> M Glucagon	3.46 $\pm$ 1.1 <sup>4</sup> 5.31 $\pm$ 1.7*	1.56 $\pm$ 0.18	1.18 $\pm$ 0.7 2.91 $\pm$ 1.7*	2.57 $\pm$ 0.39
10mM Lactate (7) +10 <sup>-4</sup> M DcAMP	3.20 $\pm$ 0.4 4.33 $\pm$ 0.6*	1.36 $\pm$ 0.15	0.80 $\pm$ 0.5 2.28 $\pm$ 1.0*	2.93 $\pm$ 0.99
10mM Lactate (12) +10 <sup>-6</sup> M Iso	3.38 $\pm$ 1.1 4.24 $\pm$ 1.3*	1.26 $\pm$ 0.20	0.80 $\pm$ 0.8 2.51 $\pm$ 2.2*	3.52 $\pm$ 1.00
10mM Lactate (7) +10 <sup>-5</sup> M Prop + Prop + Iso	3.57 $\pm$ 1.5 3.40 $\pm$ 1.6 3.52 $\pm$ 1.3	0.92 $\pm$ 0.14 0.99 $\pm$ 0.16	0.79 $\pm$ 0.9 0.66 $\pm$ 0.8 0.90 $\pm$ 1.0	0.82 $\pm$ 0.13 1.28 $\pm$ 0.40
10mM Lactate (9) +5x10 <sup>-6</sup> M Epi	2.98 $\pm$ 0.7 4.07 $\pm$ 0.7*	1.40 $\pm$ 0.16	0.25 $\pm$ 0.1 0.51 $\pm$ 0.3*	2.34 $\pm$ 0.61
10mM Lactate (7) +10 <sup>-5</sup> M Prop + Prop + Epi	2.74 $\pm$ 0.6 2.77 $\pm$ 0.4 3.21 $\pm$ 1.1	1.03 $\pm$ 0.08 1.19 $\pm$ 0.12	0.41 $\pm$ 0.3 0.31 $\pm$ 0.3 0.55 $\pm$ 0.6	0.74 $\pm$ 0.16 1.22 $\pm$ 0.45
10mM Lactate (6) +10 <sup>-5</sup> M Phent + Phent + Epi	2.81 $\pm$ 0.6 2.80 $\pm$ 0.5 3.82 $\pm$ 1.1	1.00 $\pm$ 0.06 1.37 $\pm$ 0.24	0.48 $\pm$ 0.3 0.33 $\pm$ 0.3 1.01 $\pm$ 0.7	0.63 $\pm$ 0.19 2.28 $\pm$ 0.70

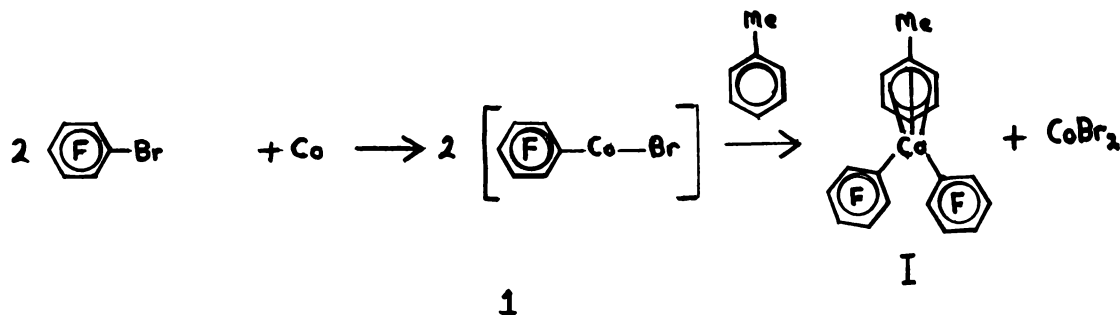
<sup>1</sup>Data expressed in terms of  $\mu\text{moles glucose}/30 \text{ min}/10^6 \text{ cells}$ <sup>2</sup>Times Control<sup>3</sup>Number of observations<sup>4</sup>Standard deviation from the mean\*significance of differences between sample means  $p < 0.05$  calculated by students T test

- Huibregtse, C.A., Rufo, G.A., Jr. and Ray, P.D. (1977) *Biochim. Biophys. Acta* 499, 99-110
- Berry, M.N. and Friend, D.S. (1969) *J. Cell. Biol.* 43, 506-520
- Hutson, N.J., Brumley, F.T., Assimacopoulos, F.D., Harper, S.C. and Exton, J.C. (1976) *J. Biol. Chem.* 251, 5200-5208

METAL ATOM SYNTHESIS AND CHEMISTRY OF A  $\pi^6$ -ARENE  
 COMPLEX OF Co(II)  $[(C_6F_5)_2-\pi^6-C_6H_5CH_3]$

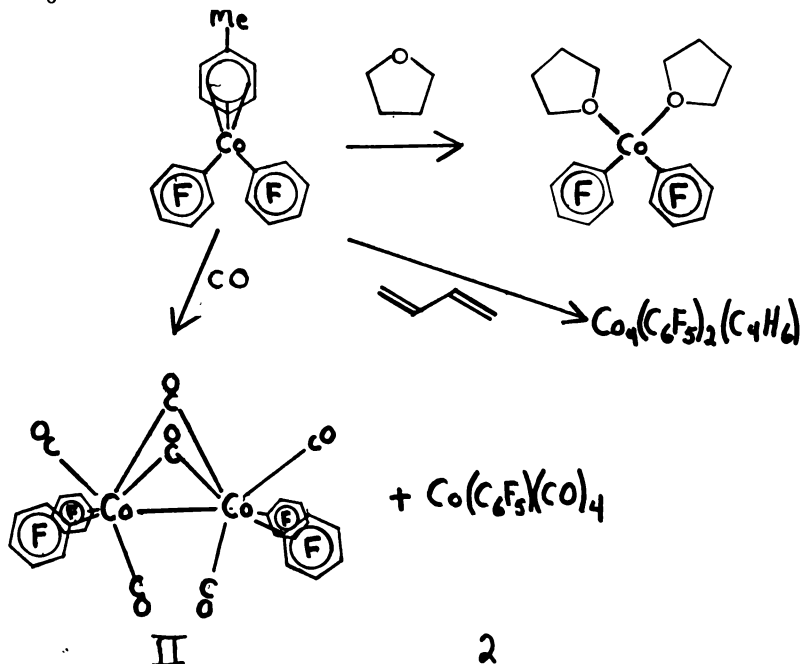
W. J. Martin\* and K. J. Klabunde  
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Using standard metal atom techniques (co-condensing metal vapors and organic compounds), we have been able to prepare  $\pi^6$ -toluenebis (pentafluorophenyl) cobalt (II) (1). Based on other reactions of metals with organic halides, we believe that the first step is the insertion of cobalt into the carbon-bromine bond to form an intermediate that readily reacts with toluene to give I and  $CoBr_2$ .



This was the first  $R_2M$  compound ever to be formed by metal atom techniques and the first  $\pi^6$ -arene complex of an  $R_2M$  compound to be prepared by any means. The pentafluorophenyl groups are  $\sigma$ -bonded to the metal, while the arene is  $\pi$ -bonded to the metal thus making it a five-coordinate, 17 electron system. The  $\pi$ -arene is very labile and can be replaced by other arenes.

The lability of the  $\pi$ -arene and the unpaired electron of the cobalt gives I a rich and unusual chemistry. We have found that CO bubbled through a toluene solution of I yields 50% conversion to  $Co_2(C_6F_5)_4(CO)_6$ , which probably has a structure similar to II, and 10%  $Co(C_6F_5)(CO)_4$ .

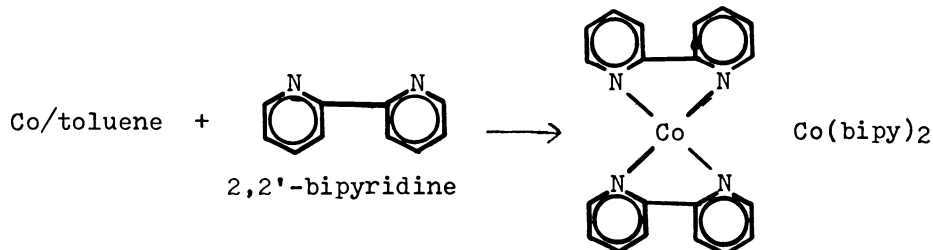


When I is dissolved in tetrahydrofuran (THF) the blue  $Co(C_6F_5)_2(THF)_2$  is formed. The reaction of I with 1,3-butadiene yields a compound whose structure hasn't been determined but it appears to be a cobalt cluster compound whose stoichiometry is  $Co_4(C_6F_5)_2(C_4H_6)$ . Likewise I has been found to react with cyclopentadiene and cyclooctadiene to form complexes containing metal-metal bonds which haven't yet been fully characterized.

METAL ATOM SYNTHESIS AND CHEMISTRY OF A  
BIPYRIDYL COMPLEX OF Co(O)

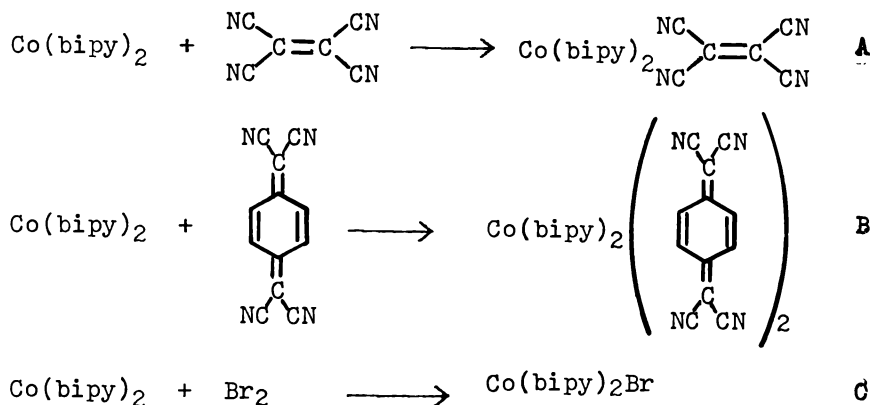
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Interest in the recent discovery of the unusual electrical conducting properties of the tetrathiafulvalene — tetracyanoquinodimethane (TTF-TCNQ) (1) and tetracyanoplatinate (2) type solids led us to investigate compounds which may exhibit similar behavior. Since these properties require stacking of the molecules in the crystal structure possible only with planar compounds, we are studying the reactivity of planar ligands such as 2,2'-bipyridine with zero-valent transition metals by the metal atom method. (3) The cocondensation of cobalt atoms and toluene at liquid nitrogen temperature produces a very reactive complex which is stable to above the meltdown temperature of the matrix. The solution of this reactive cobalt/toluene complex is stirred together with a toluene solution of bipyridine to produce a compound shown to be bis bipyridyl cobalt by elemental analysis.



Bisbipyridyl cobalt is an air sensitive black solid with a melting point of 145°C which forms very dark solutions in most organic solvents. Although it would be expected to be paramagnetic we have been unable to obtain either a ESR or NMR spectrum.

The chemistry of  $\text{Co}(\text{bipy})_2$  with certain electronegative ligands has been investigated including tetracyanoethylene (A), tetracyanoquinodimethane (B), and bromine (C). All products are deeply colored solutions in polar solvents and have been shown by elemental analysis to have formulas according to the following equations:



Further studies of  $\text{Co}(\text{bipy})_2$  and the reaction of other planar ligands with transition metals are being investigated.

1. Ferraris, John (1973) J.A.C.S. 95, 948
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3. Klabunde, K.J. (1975) Acc. Chem. Res. 8, 393



WEB ORIENTATIONS TO WIND AND LIGHT IN THE SPIDERS ARANEUS DIADEMATUS CLERCK AND ARANEUS GEMMOIDES CHAMBERLIN AND IVIE (ARANEAE: ARANEIDAE).

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The web is the most costly expenditure of energy to the orb-web spider (1), and the spiders sole source of energy input. Since the reproductive fitness of spiders is related to amount of prey consumed (2), selection should operate for web orientations which maximize energy input and minimize building and maintenance costs. Two factors, wind and light, have been suggested as effecting web orientation. To investigate the effects of these two factors, two orb-web spiders, A. diadematus and A. gemmoides were subjected to wind and light in the laboratory.

Populations of the two spiders were raised in a controlled room according to Witt (3). A wind tunnel with a 20x20x20 inch test cell was positioned in the room. Wind was provided by a window fan, and was approximately 2 miles per hour. Above the test cell was a panel of florescent lights and an incandescent bulb in a hood. These lights were on timers and provided a "sunrise" for the spiders to orient to. The lights could be swung 90° to provide an alternative light direction. Spiders were exposed daily in the cell to one of three catagories of wind and light: 1) no wind/lights normal (NW/LN), 2) no wind/lights 90° (NW/L90) and 3) wind/lights normal (W/LN). The same spider was used for each set of three exposures, and exposure order was random. Web face orientation angles were measured for each web, with the source of the factor being tested as 0°. Surface area of the webs was also measured in A. diadematus.

Table 1

Web Orientation Data for A. diadematus and A. gemmoides

Species	Category	N	$\bar{X}$ Orientation	S.D.	95% C.I.
<u>A. dia</u>	W/LN	16	350°	±58°	23° to 317°
<u>A. dia</u>	NW/LN	26	81°	±44°	56° to 106°
<u>A. dia</u>	NW/L90	14	29°	±46°	1° to 57°
<u>A. gemm</u>	W/LN	8	99°	±18°	84° to 114°
<u>A. gemm</u>	NW/LN	9	3°	±37°	33° to 333°
<u>A. gemm</u>	NW/L90	7	349°	±21°	14° to 325°

the two other catagories. A. gemmoides showed different orientations. Rayleighs Test indicated significant preferred orientations ( $p < .001$ ,  $p < .001$ ,  $p < .001$ ) to the three catagories. Watsons  $U^2$  Test indicated significant differences ( $p < .001$ ) between the wind and no wind catagories. Web areas were not compared.

A. diadematus shows no directed web orientation to wind or light, but does show reduced web area in wind situations. This may be a response to wind damage. A. gemmoides shows significant responses to wind and light. The web orientation to wind appears to be a response to reduced wind damage. The response to light may be thermoregulatory in nature, or a response to potential areas of high prey concentration or movement.

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Web orientation data for A. diadematus and A. gemmoides is given in Table 1. Rayleighs Test (4) indicated significant preferred orientations ( $p < .001$ ,  $p < .01$ ,  $p < .001$ ) in A. diadematus to the three catagories. Watsons  $U^2$  Test (5) indicated significant difference ( $p < .001$ ) in orientation between NW/LN and W/LN, but these two catagories represent the ends of a range of web orientations, and all three catagories are probably from one population. A one-way ANOVA (6) indicated significant differences ( $N=53$ ,  $p < .001$ ) in web area between the three catagories, with the webs from the W/LN group significantly smaller (T-test,  $p < .001$ ) than

THE INFLUENCE OF HORMONES,  $\alpha$ - AND  $\beta$ -ADRENERGIC EFFECTORS ON CARBOHYDRATE METABOLISM IN PERFUSED RABBIT LIVERS

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The influences of hormones,  $\alpha$ -, and  $\beta$ -adrenergic effectors on carbohydrate metabolism have been studied in isolated, perfused, rabbit livers. Livers isolated by previously described procedures (1) from 48 hr fasted, male, New Zealand rabbits were perfused with a recirculating perfusate consisting of washed rabbit erythrocytes suspended in a Krebs-Ringer-bicarbonate buffer containing heparin and "fatty acid poor" bovine serum albumin oxygenated with 95% O<sub>2</sub>:5% CO<sub>2</sub>.

Livers were "pre-perfused" for 30 min prior to initiation of experiments. After the "pre-perfusion" period, experiments were started and perfusate glucose levels were monitored in the absence of added substrate for an additional 30 min. At the end of this period, dihydroxyacetone was added to a final concentration of 40mM which was maintained by infusion. Effector(s) were added at 60 min and perfusate glucose levels were monitored throughout the rest of the experiments. Liver biopsies taken just prior to addition of substrate, just prior to addition of effector and 20 min after addition of effector, were analyzed for glycogen content. Additional biopsies taken just prior to and 3 min following addition of effector were analyzed for their content of cyclic AMP.

Listed in Table I are the rates of glucose release (expressed as  $\mu$ mole perfusate glucose/min/g liver) in the presence of 40mM dihydroxyacetone (DHA) alone and in combination with added effector(s). Also listed are the rates of glucose incorporation into glycogen (glycogenesis) as well as the rates of glucose liberation from glycogen (glycogenolysis). Table I also lists the influence of each effector on the tissue levels of cyclic AMP (cAMP), (expressed as times control). Data indicate that livers make glycogen in the presence of DHA alone. Conversely, glucagon (GLU), epinephrine (EPI), isoproterenol (ISO) (a  $\beta$ -adrenergic agonist), and phenylephrine (PHE) (an  $\alpha$ -adrenergic agonist), all cause glycogen breakdown to a statistically significant extent. However, the glycogenolytic effect of PHE is considerably and consistently less than the higher and essentially identical glycogenolytic effects of the other three effectors. The observations that EPI (known to exert both  $\alpha$  and  $\beta$ -adrenergic effects) and ISO (a known  $\beta$ -agonist whose glycogenolytic effects are essentially eliminated by the  $\beta$ -antagonist propranolol (PRO)) stimulate glycogenolysis far more than does PHE strongly suggest that the glycogenolytic effect of EPI is mediated primarily via its  $\beta$ -adrenergic effect. These observations are consistent with data obtained with isolated rabbit hepatocytes (2) and are in contrast with observations that EPI-induced glycogenolysis in rats is either mixed or predominantly  $\alpha$ -adrenergic in nature.

The large increase in tissue content of cAMP by treatment with GLU and ISO is considerably more than the consistent but marginal increase observed with EPI. Nonetheless, the apparent  $\beta$ -adrenergic mediated effect of EPI on glycogenolysis is presumably related to its ability to increase cAMP and thereby activate the glycogen phosphorylase system. Consistent with this presumption is the coincidental inhibition of the ISO-induced increases in both cAMP and glycogenolysis by PRO.

Table 1

Data From Livers Given 40mM Dihydroxyacetone(DHA)

Additions	Perfusate Glucose <sup>1</sup>	Glycogenesis <sup>2</sup>	Glycogenolysis <sup>2</sup>	Tissue cAMP Content Times Control
DHA (7) <sup>1</sup>	0.58:0.13 <sup>3</sup>	0.12:0.10		
+ 10 <sup>-9</sup> M GLU(7)	1.18:0.35		0.43:0.28*	9.05:5.00
DHA (6)	0.51:0.10	0.24:0.17		
+ 10 <sup>-6</sup> M EPI(6)	1.13:0.28		0.38:0.33*	1.46:0.64
DHA (6)	0.67:0.22	0.16:0.13		
+ 10 <sup>-4</sup> M ISO	1.23:0.24		0.45:0.41*	3.33:1.03
DHA (3)	0.60:0.14	0.28:0.13		
+ 10 <sup>-3</sup> M PRO	0.38:0.10	0.15:0.10		1.14:0.24
DHA (4)	0.62:0.02	0.13:0.03		
+ ISO + PRO(4)	0.84:0.16		0.09:0.08	0.95:0.21
DHA (5)	0.58:0.09	0.23:0.14		
+ 10 <sup>-5</sup> M PHE	0.99:0.33		0.18:0.27*	1.24:0.37

<sup>1</sup>number of observations<sup>2</sup>data expressed in  $\mu$ mol glucose/min./g liver<sup>3</sup>data expressed as standard deviation from the mean

\* indicates data with effector is significantly different at p &lt; 0.05 from data with substrate alone as calculated by the Students' T-test

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## ANAEROBIC DENITRIFICATION OF SYNTHETIC IRRIGATION RETURN FLOW

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Eutrophication of lakes and streams is often caused by high concentrations of nitrate-nitrogen ( $\text{NO}_3\text{-N}$ ). Excessive  $\text{NO}_3\text{-N}$  concentration is responsible for methemoglobinemia in infants and is detrimental to livestock. The National Interim Primary Drinking Water Regulations (1) specify a maximum  $\text{NO}_3\text{-N}$  level of 10 mg/l in drinking water. Agricultural irrigation return flows, such as that from the proposed Garrison Diversion Unit (2), normally contain high concentrations of  $\text{NO}_3\text{-N}$ . The removal of excessive  $\text{NO}_3\text{-N}$  concentration from irrigation return flow is necessary to safeguard the quality of the receiving waters.

Methods of removing  $\text{NO}_3\text{-N}$  from wastewater include both chemical methods using ion-exchange processes and biological methods using acclimated microorganisms. The purpose of this study was to investigate the feasibility of using anaerobic upflow continuous biological filters for the removal of  $\text{NO}_3\text{-N}$  from synthetic irrigation return flow. Since there is no irrigation return flow water available in North Dakota at the present time, synthetic irrigation return flow was used in this study. Clear polycarbonate columns of 3-in. (7.62cm) inside diameter and 6-ft. (1.83m) height filled with 5/8-in. (1.59cm) plastic rings as filter media were used. In the laboratory study the feed  $\text{NO}_3\text{-N}$  concentration varied from 5 to 100 mg/l, the methanol to  $\text{NO}_3\text{-N}$  ratio ranged from 0.5 to 3.0, and the hydraulic loading varied from 130 to 1,220 gpd/ft<sup>2</sup> (5.4 to 49.8 m<sup>3</sup>/day/m<sup>2</sup>). Results of the denitrification study are summarized in Table I. For feed waters containing 40 mg/l or less of  $\text{NO}_3\text{-N}$  concentration a 2-ft. (0.61m) column is sufficient to produce effluents of 10 mg/l or less of  $\text{NO}_3\text{-N}$ . For  $\text{NO}_3\text{-N}$  concentration of 60 to 100 mg/l a 6-ft. (1.83m) column height is needed. For  $\text{NO}_3\text{-N}$  concentration of 100 mg/l a maximum hydraulic loading of 815 gpd/ft<sup>2</sup> (33.1 m<sup>3</sup>/day/m<sup>2</sup>) can be used to produce effluent of 10 mg/l or less of  $\text{NO}_3\text{-N}$ . For feed water containing 40 mg/l or less of  $\text{NO}_3\text{-N}$ , a methanol to  $\text{NO}_3\text{-N}$  ratio of 0.5 is sufficient to have the required  $\text{NO}_3\text{-N}$  removal. For higher concentrations of 60 to 100 mg/l, a ratio of 1.6 is needed. Based on this study, the anaerobic filters appear to be a feasible method of removing excessive  $\text{NO}_3\text{-N}$  concentrations from irrigation return flow to meet the drinking water standards.

Table I

Data for Denitrification Columns at 49.8 m<sup>3</sup>/day/m<sup>2</sup>

Column height m	Effluent $\text{NO}_3\text{-N}$ (mg/l)			
	20 - 0.5*	40 - 0.5	60 - 1.6	100 - 1.6
0.30	9.8 ± 3.1	21 ± 4.6	20 ± 4.4	50 ± 7.1
0.61	6.8 ± 2.6	14 ± 3.7	17 ± 4.1	46 ± 6.7
0.91	4.6 ± 2.1	7.6 ± 2.7	14 ± 3.7	30 ± 5.5
1.22	2.9 ± 1.7	5.0 ± 2.2	8 ± 2.8	29 ± 5.0
1.52	2.5 ± 1.5	3.8 ± 1.9	7 ± 2.6	20 ± 4.5
1.83	2 ± 1.4	2.7 ± 1.6	6.4 ± 2.5	18 ± 4.0

\*Feed  $\text{NO}_3\text{N}$  concentration (mg/l) - methanol/ $\text{NO}_3\text{-N}$  ratio

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## WHY TEACH ENVIRONMENTAL ETHICS?

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A joint venture in teaching Environmental Ethics between the Biology and Philosophy Departments at the University of South Dakota was initiated as an experiment in interdisciplinary teaching in 1974 and has been continued in alternate years. A professor trained in Environmental Biology represented the Biology Department and one trained in Ethics represented the Philosophy Department.

There were no formal lectures. Instead, thorough discussions were held on controversial environmental issues having ethical implications. Source books used as a basis for discussions are listed in the bibliography. Other specific readings were also assigned. The purpose was to give a philosophical, economic, sociological, practical, scientific, and hopefully, ethical background to the environmental problems and crises of our time. Later, specific, difficult, controversial, environmental topics were chosen by student groups for study and discussion.

Students represented many disciplines, both scientific and non-scientific, and were a good academic mix. All were upper classmen or graduates. To increase participation they were often divided into small interdisciplinary groups.

Out of the course has come a clearer understanding of the complexity of environmental problems and the necessity in many cases of making compromises to achieve a workable solution. The costs of correction, both economic and sociological, are often considerable. The experiences in the course have been most worthwhile and have been both revealing and humbling to students and professors alike.

We would certainly recommend that other colleges and universities in the region initiate such a course. We believe such courses represent interdisciplinary education at its most useful best.

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## SWIMMING PERFORMANCE OF MISSOURI RIVER FISHES

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In the remaining unchannelized Missouri River below the mainstem dams a degradation erosion cycle occurs resulting in bank erosion. Congress in 1974 directed the U. S. Corps of Engineers to develop bank stabilization methods which will prevent erosion but not drastically alter the river environment. Structures and methods previously used to control bank erosion have usually proven detrimental to the biota, particularly the fish community. However, there is little conclusive evidence as to what factors cause community changes following bank stabilization programs. This study addressed the problem of determining what role swimming abilities may have in the successful habitation of lotic ecosystems which have been altered by man.

Field experiments were conducted on 281 fish representing 17 species using an open, flow-through system. A centrifugal pump pulled water from the river through 25.4 cm (ID) irrigation pipe, an acrylic observation chamber and back to the river. Fish were retained in the chamber by screens and current velocities in the system were monitored with a pitot tube-manometer combination. The entire apparatus was mounted on a pontoon boat. Two types of swimming speeds were determined, prolonged and sustained speeds. Critical speeds are prolonged speeds and represent the highest swimming speeds fish can maintain for 10 minutes. Sustained speeds represent the highest swimming speeds fish can sustain for 200 minutes and are usually expressed as a percentage of the critical speed.

The blue sucker, a species restricted to large rivers, was the best swimmer with a mean critical velocity (CV) of  $80.1 \pm 41.8$  cm/sec ( $\bar{x} \pm 2SD$ ). On a sustained basis this species swam at 83% of its critical speed. Other species which were considered good performers included the shovelnose sturgeon ( $\bar{x} = 75.2$ ), goldeye ( $\bar{x} = 79.5$ ), walleye ( $\bar{x} = 74.7$ ), and freshwater drum ( $\bar{x} = 75.2$ ). Shortnose gars ( $\bar{x} = 50.5$ ) were the poorest swimmers.

In general, critical speeds were significantly related to standard length ( $r=0.2979$ ,  $P < 0.01$ ) and to water temperature ( $r=0.1298$ ,  $P < .05$ ). No consistent relationship existed between critical speeds and the sex of the specimen or the method of capture used in securing the fish although in some species these variables contributed to the critical speed variance.

Although longer fish of each species performed better than smaller fish on an absolute basis, young-of-the-year and older immature fish were better relative performers. Critical speeds expressed as SL/sec ranged between 1.0 and 3.9 for adults, 1.0 and 13.2 for immatures and 1.4 and 17.2 for young-of-the-year.

There was relatively good agreement between the experimentally determined critical speeds for each species and their distribution in Missouri River habitats. The best swimmers were usually common or abundant in habitats with higher current velocities. Ubiquitous species like the carp ( $\bar{x} = 67.3$ ) and shorthead redhorse ( $\bar{x} = 67.6$ ) exhibited intermediate critical speeds (cm/sec).

All 17 species performed at prolonged and sustained speeds which would permit permanent habitation of some Missouri River habitat. However, none of the species had prolonged or sustained swimming speeds which would permit continuous habitation in the main channel or near revetments in the channelized Missouri River. Current velocities at these locations reached  $> 200$  cm/sec and exceeded the critical speeds of the best swimmers although all species could swim well enough to spend short time intervals in these habitats.

## OZONE INJURY SYMPTOMS ON PLANT BIOINDICATORS IN THE PRESENCE OR ABSENCE OF POWER PLANT EMISSIONS.

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Since 1970 air pollution monitoring at 22 locations in South Dakota has detected ozone (O<sub>3</sub>) injury on leaves of Nicotiana sp. bioindicators. In an effort to extend our knowledge of the distribution and severity of air pollution injury to plants in the state, plant indicator plots were established at Mobridge, South Dakota in 1977 and 1978. The cultivars used, sensitivity to pollutant and response to ambient air are shown in Table 1.

In 1977, none of the test plants responded with leaf symptoms predicted for O<sub>3</sub>, PAN, or fluoride. Gladiolus sp. did show symptoms typical of sulphur dioxide (SO<sub>2</sub>) (1). The tobacco cultivars were resistant to SO<sub>2</sub> (1).

In 1978, leaves of tobacco cultivars Bel C, Bel W<sub>3</sub> and Wisconsin 38 all showed typical "weather-fleck" symptoms of O<sub>3</sub> injury that were evident as black, brown, tan, or white necrotic spots on newly matured and older leaves. The spots were largest (1-3mm) on Bel W<sub>3</sub> and smallest (0.1-1.0mm) on Bel C and Wis. 38. The resistant Bel B leaves were not injured. The absence of O<sub>3</sub> injury in 1977 and presence in 1978 suggested that something in the power plant plume protected the indicator plants from O<sub>3</sub> injury in 1977.

Two other studies (2,3) have shown that O<sub>3</sub> was depleted in air near fossil-fuel power plants and "bulged" after two to four hours in the down-wind plume. The depletion reaction near the source was believed to be due to "conversion of nitric oxide to nitrogen dioxide" (2) and favoring the "ozone bulge" was "nitrogen oxides (NO<sub>x</sub>) emissions are apt to cause elevated O<sub>3</sub> concentrations in nonurban areas where there are natural concentrations of non-methane hydrocarbons and where low NO<sub>x</sub> concentrations normally prevail" (3).

TABLE 1

Response of selected air pollution bioindicator plants at Mobridge, SD for years 1977 and 1978.

Plant Bioindicator	Air Pollutant	Actual Response*	
		1977	1978
<u>N. tabacum</u> 'Bel B'	Ozone	None	None
<u>N. tabacum</u> 'Bel C'	Ozone	None	Positive
<u>N. tabacum</u> 'Bel W <sub>3</sub> '	Ozone	None	Positive
<u>N. tabacum</u> 'Wis 38'	Ozone	None	Positive
<u>N. tabacum</u> 'Turkish'	Ozone	None	Positive
<u>N. glutinosa</u>	PAN**	None	None
<u>Gladiolus sp.</u> 'Glacier'	Fluoride	None	None

\*Leaf symptom expression to predicted air pollutant.

\*\*Peroxyacetyl nitrate, one of the oxidant air pollutants.

Our results coupled with other research (2,3) suggest that, in the Dakotas, we might expect ozone injury to plant life 2-4 hours (3) downwind or 2<sup>4</sup> or more kilometers (2) beyond large fossil-fuel electric power generators. Plants should be protected from O<sub>3</sub> but might be injured by SO<sub>2</sub> near the pollution sources. Meteorological conditions and mechanical factors at each location also might influence the type and amount of plant injury.

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Mechanism of Specific and Nonspecific Intravenous Immunotherapy Against Herpesvirus hominis Type 2 Infection and Encephalitis in Rabbits

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It was reported (1, 2) that intravenous immunization of two strains of rabbits with viable Mycobacterium bovis (BCG) cells provided protection against subsequent infections with Herpesvirus hominis (HVH) Type 2. Assessment of in vivo immunity was made in vitro by measuring the protection of indicator vero cells against HVH type 2 infection in the presence of different immune systems.

The present study is an attempt to obtain some further insight into the mechanism of specific and nonspecific immunity and to enhance it in animal model.

Different groups of young New Zealand white rabbits were immunized intravenously with 0.5 ml (k-200) of BCG (canada strain), HVH Type 2 antiserum, HVH Type 2 vaccine and any combination of two of the above three. Immunization with BCG and HVH Type 2 vaccine were given two weeks before the challenge with  $2 \times 10^3$  TCID (Tissue culture infectious dose) virus. When HVH Type 2 antiserum used, all animals were challenged with virus within 24 hours after immunization with 0.5 ml of antiherpetic serum (average pool titer 1:64). When macrophage destruction was required, the animals were given size 5 Min-u-sil (silica) particles (1.6 gms/1800 grams of animal). All animals were examined for delayed hypersensitivity and sera were tested for antibody, interferon and macrophage inhibition factor (MIF). Attempts were made to reinfect animals recovered from HVH Type 2 infection with higher doses ( $4 \times 10^3$  TCID) of virus by corneal scarification.

Treatment with silica was fatal for all the BCG vaccinated and normal animals, but not in animals with high herpes specific antibody titer (1:32 to 1:64). Animals recovered from HVH Type 2 infection had shown resistance to subsequent infection. No keratitis, kerato-conjunctivitis or encephalitis was observed in this group of animal. This experiment shows that HVH Type 2 antibody was effective in neutralizing the virus in the lacrimal fluid and phagocytized the virus before it could establish infection.

Recovered BCG vaccinated rabbits were immunized with HVH Type 2 vaccine, produced highest titer (1:64) of antibody in contrast to BCG vaccinated control and normal control. No mortality was observed in recovered BCG vaccinated animals later receiving virus vaccine. BCG and virus vaccinated rabbits were observed to be more resistant to HVH Type 2 infection than those of BCG sensitized alone. But BCG sensitized animals with low antibody titer are more susceptible than BCG sensitized alone. The mortality rate was 75 percent in control virus vaccinated animals and 100 percent in normal control animals. In the presence of very low (1:24-1:4) to non-detectable antibody titer, BCG vaccinated animals were very susceptible to fatal encephalitis. Probably in the presence of a very low titer of specific antibody, a membrane surface property of BCG sensitized macrophages was changed and phagocytic mechanism was blocked as virus bound itself to specific cellular receptors. Results indicated that, when antiherpetic serum (rabbit) was used as an immunizing agent on BCG vaccinated and non-vaccinated rabbits, BCG vaccinated rabbits injected with specific HVH type 2 serum became more susceptible to fatal infection than serum vaccinated alone.

Observations of complement fixing antibody titer was made in both BCG vaccinated and unvaccinated animals following challenge with virus. The highest complement fixing antibody titer was 1:4 after 384 hours. No interferon titer was observed. Migration inhibition test was performed to see whether the immunity is related to MIF. The results indicate that most of the BCG sensitized animals with observed MIF did not show any delayed hypersensitivity reaction. Only one survived in BCG sensitized group and none survived in the control. This data suggests that migration inhibition factor was not responsible for protection of rabbits, if any. The second group of BCG sensitized animals did not show the presence of any migration inhibition factor but most of the animals showed delayed hypersensitivity reactions in contrast to the previous groups. In this experiment 75 percent of the BCG sensitized animals survived HVH type 2 infection in contrast to 28 percent of the control animals. The above data indicated that delayed hypersensitivity might be responsible for the protection of rabbits against fatal infections with type 2 virus and because of absence of MIF. MIF secretion starts 6-8 hours after contact with antigen and continued for four days.

The above results indicate that New Zealand rabbits were protected against HVH type 2 infection with subsequent fatal encephalitis due to increased activity of macrophage or effective neutralization of virus by active macrophages in the presence of specific antibody.

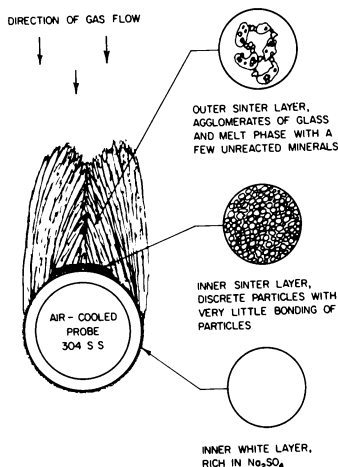
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MICROSCOPIC STUDIES OF ASH DEPOSITS AND  
COAL RELATED TO THE ASH FOULING PROBLEM

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A major problem in the combustion of low rank, western coals is ash fouling, a buildup of ash deposits on power plant boiler tubes and walls. The GFETC has studied the ash fouling potential of western coals for several years. The major index for fouling is the sodium content in the ash<sup>1</sup>, the higher the sodium content (>5%), the more severe the fouling. GFETC operates a pilot plant combustor which has probes designed to simulate power plant boiler tubes. Microscopic studies of the ash deposits from these probes have produced a typical structure of an ash deposit shown in the Figure<sup>2</sup>. Microscopic studies of the various particles from the inner and outer sinter layers from a Beulah N.D. lignite comprise the majority of this work. Characterization of the inorganic constituents in the coal is also an ongoing concern at the Center. Additional microscopic techniques utilized in this work are also discussed.

Equipment utilized to study the ash deposits and coal includes a heated stage microscope (HSM) capable of temperatures to 1700°C, a scanning and analyzing electron microscope (SEM), and a polarized light microscope (PLM) with photographic capabilities.



Typical Ash Deposit

Analysis of individual ash deposit particles is done using a combined HSM and SEM approach to determine the melting behavior of ash particles in relation to their chemical composition. This information is used to help delineate ash deposit formation and to predict possible materials to be added to the coal to reduce deposit formation. Reactions involving sodium are of particular interest due to their causative affects in fouling.

Thin section analysis of ash deposit particles using PLM and SEM were undertaken to characterize the crystalline components of the deposits. This work lends support to deposit formation theories, showing probable chemical components recrystallizing from the deposit melt phase. Crystalline coatings of  $\text{CaSO}_4$  have been identified surrounding ash deposit particles and the average chemical composition of the crystalline matrix material was determined.

SEM investigations of coal to determine the occurrence and distribution of inorganic constituents is a continuing project. Some of the SEM methods used to study the coal for inorganic constituents include:

1. Line scans of a polished coal surface for specific elements. These scans depict elements which are uniformly dispersed such as Na, Ca, S and Mg; and those elements which are locally concentrated, mainly Al, Si and Fe, implying particles of inorganic material such as clays, quartz and pyrites.
2. Elemental mappings of a polished coal surface showing particle distribution and frequency. Pyrite particles,  $\text{FeS}_2$ , have been identified using this method.
3. SEM analyses of sink particles from  $\text{CCl}_4$  float-sink separations, the sink fraction being the high density, mineral particles in the coal.

Inorganics observed in the coal are correlated to the composition of the ash deposits and intermediate reactions are formulated. Individual small coal particles after partial combustion were analyzed in the SEM and have assisted in this area. This work shows reactions which occur instantaneously upon combustion.

Combined microscopy efforts are yielding valuable information towards understanding the mechanisms involved in ash fouling and in predicting possible additives for fouling control.

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## QUANTITATIVE ANALYSIS OF COAL DERIVED LIQUIDS BY LOW VOLTAGE MASS SPECTROSCOPY

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At the Grand Forks Energy Technology Center, major emphasis is placed on the conversion of low ranked coals to pipeline products through the CO-Steam liquefaction process<sup>1</sup> and by slagging fixed-bed gasification. CO-Steam products and gasifier tars are very complex mixtures of aromatic hydrocarbons and oxygen, nitrogen and sulfur heterocycles. The complexities of these mixtures pose some very difficult analytical problems.

Several methods of analysis, including gas chromatography, liquid chromatography and mass spectrometry, have been tried. Mass spectrometry has proven to be the best technique for analysis in that it has the advantage of direct molecular weight determination. Operating at ionizing potentials of 70 eV, which is a standard for analysis of pure compounds, is not practical due to the complex nature of the samples and the great amount of fragmentation produced. By lowering the ionizing potential to 10 eV, fragmentation is eliminated and the only species remaining are peaks due to molecular ions and their isotopes.<sup>2</sup> Table 1 illustrates the determined accuracy of low voltage mass spectroscopy as compared to gas chromatography for a sample of coal tar oil. Table 2 is an example of the accuracy achieved on repeated analysis of a sample run at low voltage. The compounds selected represent three major component types, i.e. aromatic hydrocarbons, nitrogen heterocycles and oxygen heterocycles.

Quantitation of these complex mixtures is possible by using the intensity of the molecular ion and an appropriate sensitivity factor. Computerized data reduction is used to calculate a percentage of total concentration for each component in the sample using the following equation:

$$\% \text{ Component} = \frac{I_i K_i C}{\sum I_j K_j} \times 100$$

where  $I_i$  = intensity of the peak at each mass,  $K_i$  = relative sensitivity factor for each mass<sup>3</sup>, and  $C$  = correction for % non-volatile material and % saturates in each sample.

Sensitivity factors were determined relative to naphthalene. Commercially available compounds known to be in coal derived liquids along with a variety of synthesized methylated polynuclear aromatics were used to determine sensitivity factors.

Since low voltage mass spectroscopy eliminates only peaks due to fragmentation and not those due to natural isotope abundance, it is necessary to correct for these isotopic contributions to peaks due in fact to a different compound.

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TABLE 1

## Accuracy of LVMS

Compound	Crowley Coal Tar Oil	
	MS(66) <sup>a</sup>	GC(28) <sup>a</sup>
Naphthalene	5.6	8.5
Methylnaphthalene	15.5	20.3
Fluorene	6.2	7.0
Phenanthrene	16.1	18.8
Pyrene & Fluoranthene	11.0	12.7

a Number of components detected

TABLE 2

## Precision of LV-MS

Compound	Trial Number					$\bar{X}$	$\sigma$	$\sigma R$
	1	2	3	4	5			
Fluorene	3.75	3.94	3.64	3.71	3.81	3.77	0.11	3.0
Carbazole	0.58	0.57	0.61	0.62	0.55	0.59	0.03	4.9
Dibenzofuran	6.17	6.35	6.22	6.33	6.50	6.31	0.13	2.0

## AN OVERVIEW OF EFFLUENTS FROM THE GASIFICATION OF SOME WESTERN COALS

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The Process Measurements Branch, Industrial Environmental Research Laboratory - Research Triangle Park, has developed a phased analytical approach for Environmental Assessment Programs (1). The objective of the first phase, Level 1, is to provide within broad general limits the approximate concentrations of inorganic elements and classes of organic compounds by gas chromatography (GC), liquid chromatography (LC), infrared analysis, low resolution mass spectrometry and elemental analysis. Level 1 techniques have been applied to tar/water condensate samples from the spray washer of the Grand Forks Energy Technology Center slagging fixed bed gasifier (2). Samples from the gasification of three coals were obtained for study: 1. Indianhead Lignite, 2. Gascoyne Lignite, and 3. Rosebud Subbituminous.

Solvent extraction of the tar with methylene chloride yielded approximately 28 to 35% extracted organics, 0.34 to 1.2% unextractable residue, and 64 to 72% water. Methylene chloride/diethyl ether extraction of the liquor at pH 8 and pH 2 yielded 1200 to 2100 ppm extracted organics. The extracted organics from the tar and liquor were analyzed by GC simulated distillation and separated into eight approximate classes by gradient elution LC. The volatility of the tar correlates generally with the geological age of the coal gasified. Gascoyne tar had a lower boiling point range than Rosebud tar, respectively the younger and older coals. The mass distribution of the LC fractions of the tar extract is 10-13% paraffins, 32-40% aromatic species, 45-52% polar species, and 1-6% very polar species. LC separation of the liquor extract indicates a mass distribution of 0-2% paraffins, 0-8% aromatic species, 81-89% polar species and 7-19% very polar species. Infrared spectra were used to determine the functional groups present in each LC fraction.

The unextractable residues of the tar, coal fines, were analyzed for elemental content and size distribution. Sulfur and nitrogen content of the coal fines is greater than in the coal feed. This indicates loss of carbon, hydrogen, and oxygen early in the carbonization process.

The substances in tar and liquor are exceedingly complex and require fractionation before specific characterization can be accomplished. Level 1 analyses will supply samples of isolated chemical groups that may be considered important gasifier pollutants due to toxicity or high concentration for further investigation. Level 1 analyses also provide a means of studying the effect of various coals and parametric variations on the production of effluents. Data will be presented showing the results of Level 1 analyses, types of coals used, and range of conditions used in the gasification experiments.

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THIOCYANATE DEGRADATION BY AN ARTHROBACTER

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Thiocyanate is present in the waste waters produced during coal gasification and coke production. It can be used as an indicator of pollution from these sources. In Eastern Europe, on an experimental basis, such waste waters have been diluted with surface water and used to irrigate crop land (1). Soil was used as a means of biologically treating the waste. Because thiocyanate is somewhat resistant to biodegradation, we have undertaken a study of the degradation of thiocyanate by soil microorganisms.

A gram-positive, heterotrophic bacterium was isolated from soil which utilized thiocyanate as nitrogen-source. The isolate, identified as an Arthrobacter, was polymorphic and gave salmon-colored, opaque colonies. No flagella were present. The organism was able to grow on 31 different carbon compounds, including alcohols, amines, carbohydrates and acids.

The Arthrobacter isolate could tolerate thiocyanate up to 0.1M (Fig. 1). It was able to degrade thiocyanate in the presence of ammonium or nitrate ions. This is in contrast to a Pseudomonas reported to degrade thiocyanate only in the absence of nitrate or ammonia (2). The Arthrobacter isolate showed equal ability to use as nitrogen-source either nitrate, ammonia, or thiocyanate (Fig. 2). Thiocyanate-degrading activity was greatly diminished as the culture approached stationary phase.

This is the first Arthrobacter reported to degrade thiocyanate. Two other members of this genus, A. globiformis ATCC 8010 and A. tumescens ATCC 6947, were unable to degrade the ion. The other bacterial genera reported to degrade thiocyanate are Thiobacillus and Pseudomonas.

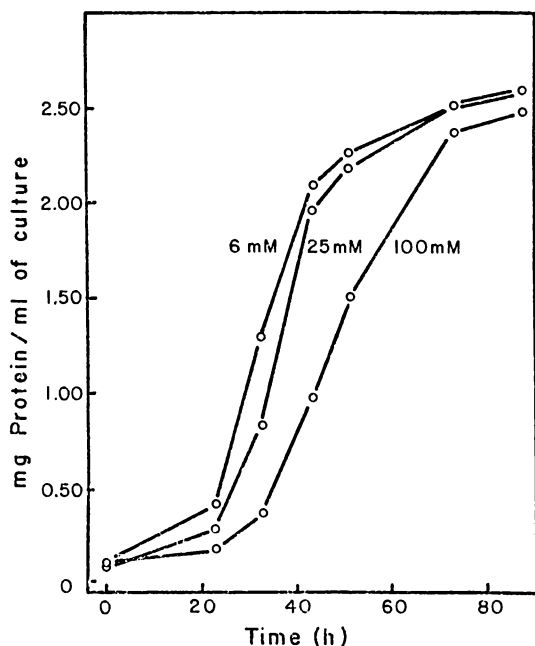


Fig. 1. Growth of the isolate on media containing 6, 25 or 100 mM sodium thiocyanate.

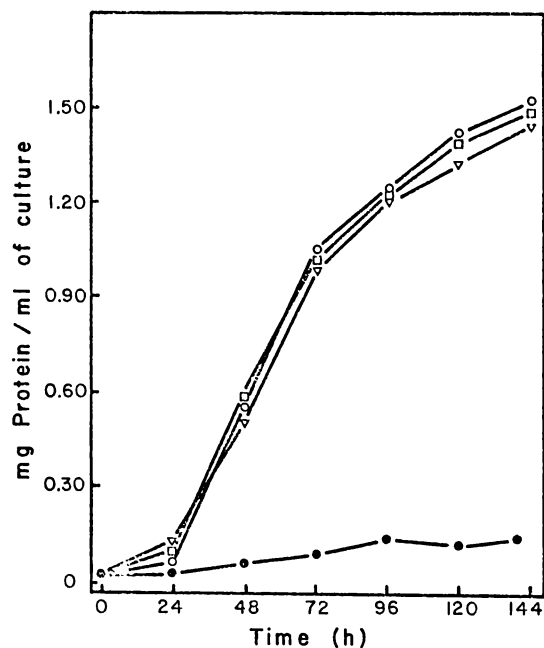


Fig. 2. Growth of the isolate on media containing as N-source (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>, (▼); NaNO<sub>3</sub>, (■); thiocyanate, (●) or no nitrogen added, (●).

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PRIMARY PRODUCTION OF PLANT COMMUNITIES  
WITHIN A WESTERN MINNESOTA TALL GRASS PRAIRIE

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The Red River Valley comprises the northern extent of the tall grass prairie in North America. Studies of the production ecology of prairie plant communities in the Red River Valley are few in number and, with one exception (3), are quite localized (1, 2, 4, 5). Literature reporting root production of prairies is even more sparse. The present investigation was initiated to study net primary production (aboveground and belowground) of selected plant communities in a native tall grass prairie in west-central Minnesota.

Bluestem Prairie (owned and managed by the Minnesota Chapter of The Nature Conservancy) is a 486 ha tract located along the Campbell beachline of glacial Lake Agassiz, approximately 23 km east of Moorhead, Minnesota in Clay County. The soils consist of very poorly to moderately well drained material formed in glacio-lacustrine deposits or lacustrine sediments on glacial lake plains. Topography is slightly rolling with slopes ranging from 0 to 6%. The mean annual temperature of the area is approximately 5.3°C, and the average annual precipitation is 49 cm with about 75% of the total occurring from April to September.

Six plant communities within the prairie were selected for an intensive two year study, 1978-1980. (This paper reports partial results for the 1978 growing season.) These areas include three upland and two lowland communities, all of which are relatively undisturbed, and one overgrazed area only recently set aside. Within each community a permanent study area 25 m on a side was delimited. From May to October, eight random 0.25 m<sup>2</sup> quadrats of aboveground plant material (separated into graminoids, forbs, standing dead, and litter) were harvested at biweekly intervals from each community. Root samples were collected at monthly intervals (June-September) using a bucket auger. Five cores 9.5 cm in diameter (consisting of two consecutive 30.5 cm increments) were extracted from previously clipped quadrats. Inorganic material was washed free of the root/rhizome mass, and the belowground and aboveground plant material was oven-dried to constant weight. Average dry weights for each component were considered to represent mean standing crop for that component for each sampling period. Basal plant cover was determined from 100 systematically placed point frames within each of the study areas.

The three upland communities, Bouteloua gracilis-Stipa spp., Andropogon scoparius-Sporobolus heterolepis and Andropogon scoparius-Sorghastrum nutans, all exhibited lower aboveground peak standing crop values than the lowland communities, Calamagrostis inexpansa-Andropogon gerardi and Carex spp.-Calamagrostis inexpansa (Table 1). However, the greatest aboveground peak was noted in the disturbed Melilotus alba-Agropyron repens community which had been free from grazing for two growing seasons and which was completely dominated by a luxuriant growth of Melilotus. That same trend was not noted in the belowground component wherein the standing crop for two of the upland communities (Bouteloua-Stipa and Andropogon-Sorghastrum) was greater than that for all except the Carex-Calamagrostis area, a moist swale community. Other than at that one site, the community peak standing crop values are quite comparable, in spite of distinct variations in plant basal cover and species composition. These data reinforce the view that a fuller understanding of grassland primary production must include analysis of belowground contributions.

Table 1. Basal cover and peak standing crop values for the 1978 growing season

Community Type	a (%)	b (g/m <sup>2</sup> )				
		b	c	d	e	f
<u>Bouteloua-Stipa</u>	23.3	203.9	258.2	462.1	4222.5	4612.4
<u>Andropogon-Sporobolus</u>	44.6	437.6	23.2	460.8	3383.1	3809.7
<u>Andropogon-Sorghastrum</u>	28.4	323.4	11.4	334.8	4233.8	4560.3
<u>Calamagrostis-Andropogon</u>	39.8	462.2	119.7	581.9	3905.1	4487.0
<u>Carex-Calamagrostis</u>	25.7	602.2	35.1	637.3	8703.2	9332.2
<u>Melilotus-Agropyron</u>	11.4	212.1	924.2	1136.3	3621.5	4565.1

a = community basal cover; b = graminoid, c = forb and d = total biomass at aboveground peak; e = belowground peak plant material (living and dead); f = community peak standing crop (aboveground and belowground peaks were usually not simultaneous)

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Soil and Plant Relationships of Selected Wetlands  
on the Missouri Coteau

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Correlation between the vegetation and soils of wetlands is desirable for purposes of soil classification and mapping (1). Water depth and permanence are known to influence zonation of wetland vegetation (2) and soil horizon development in temporary and seasonal wetlands (3).

A preliminary investigation of vegetation and soil relationships was conducted on semipermanent ponds on the Missouri Coteau 25 miles northwest of Jamestown, North Dakota during the summer of 1978. Four wetlands having similar dominant species in the wet meadow, shallow marsh, and deep marsh zones were selected. At each site a transect of soil profiles was established perpendicular to the vegetation zones in order to classify and characterize soil properties (Table 1—chemical properties only). Near each profile the vegetation was sampled with ten quadrats (0.25 m<sup>2</sup>) for species present, above ground biomass and density (Table 2). Water level (or depth to water table) was measured periodically during the summer beginning in early June.

Correlation and significance values were calculated for soil, vegetation and water level parameters. Significant differences between ponds indicating landscape changes were found for Na, Mg, Ca, Electrical Conductance (EC) and Sodium Adsorption Ratio (SAR) at the 1% probability level. The vegetation zonation correlated to CaCO<sub>3</sub> and pH at the 5% probability level and nearly so for organic carbon (5.1% level) indicating that these are soil changes. Water level changes appear to influence soil type, organic carbon, pH, Na and SAR (1% probability level). Additionally, density, biomass and species presence correlated to water level changes at the 5% probability level. The density of the vegetation was a better indicator of soil properties than the type of vegetation. Vegetation density correlated to zone, CaCO<sub>3</sub>, Na and SAR at the 1% probability level.

Table 1

	Soil Chemical Properties <sup>1</sup>			
	Site 1	Site 2	Site 3	Site 4
pH	8.0 ± 0.1 <sup>4</sup>	7.5 ± 0.4	8.0 ± 0.2	7.8 ± 0.5
Organic carbon (%)	3.2 ± 2.1	7.4 ± 2.6	2.7 ± 2.3	5.1 ± 3.3
CaCO <sub>3</sub> (%)	12.3 ± 2.6	7.9 ± 5.9	8.0 ± 1.6	16.7 ± 16.8
Ca (meq/l)	1.8 ± 0.4	2.1 ± 0.6	1.8 ± 0.8	0.9 ± 0.3
Mg (meq/l)	7.0 ± 3.2	2.2 ± 0.8	6.2 ± 2.8	1.8 ± 1.3
Na (meq/l)	3.9 ± 2.6	0.7 ± 0.2	2.3 ± 1.3	0.5 ± 0.4
E.C. <sup>2</sup> mmhos	8.4 ± 2.9	3.9 ± 0.9	7.4 ± 2.6	2.6 ± 1.4
SAR <sup>3</sup>	1.8 ± 0.8	0.5 ± 0.1	1.2 ± 0.5	0.5 ± 0.2

<sup>1</sup>Mean ± Standard Deviation. <sup>2</sup>Electrical Conductance. <sup>3</sup>Sodium Adsorption Ratio. <sup>4</sup>Each value represents three 6" depth increments from four profiles.

Table 2

Vegetation	Above Ground Biomass (g/m <sup>2</sup> ) <sup>1</sup>			
	Site 1	Site 2	Site 3	Site 4
Wet meadow	396 ± 53	484 ± 192	234 ± 55	444 ± 177
<i>Scolochloa festucacea</i>	176 ± 124	754 ± 321	512 ± 246	728 ± 235
<i>Typha latifolia</i>	---	686 ± 353	---	744 ± 269
<i>Typha glauca</i>	1404 ± 500	---	924 ± 301	---
<i>Scirpus acutus</i>	1092 ± 508	828 ± 530	508 ± 250	1032 ± 518

Vegetation	Density (shoots/m <sup>2</sup> ) <sup>1</sup>			
	Site 1	Site 2	Site 3	Site 4
Wet meadow	984 ± 219	730 ± 104	447 ± 170	995 ± 283
<i>Scolochloa festucacea</i>	202 ± 140	638 ± 178	479 ± 204	601 ± 170
<i>Typha latifolia</i>	---	25 ± 13	---	42 ± 9
<i>Typha glauca</i>	71 ± 24	---	58 ± 12	---
<i>Scirpus acutus</i>	353 ± 154	178 ± 77	174 ± 47	370 ± 158

<sup>1</sup>Mean ± Standard Deviation

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DETERMINATION OF LINEAR REGRESSION FOR NET SOLAR  
RADIATION AS A FUNCTION OF SOLAR RADIATION

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The use of aircraft and satellite for monitoring land-surface emittance in the thermal infrared region of the spectrum has become common in recent years. Scanners with detection capability in the thermal infrared region are being used extensively on a routine basis at aircraft altitudes. Existing and proposed satellites also include a capability for thermal infrared data collection. The synoptic view provided by these remote sensors is especially required when monitoring surface temperature patterns over large areas due to the dynamic nature of land-surface temperatures.

Since the emittance of the land surface is dependent upon a multitude of factors such as time of day, month of the year, elevation of land surface, aspect of slope, land use, soil moisture, and climatological variables the isolation of emittance variations caused by a single terrain feature is very difficult. Therefore, models describing emittance variations associated with various physical features must be developed to isolate their effects and to understand their interdependence.

One of the most important input parameters to these types of models is net solar radiation which consists of the total solar radiation incident on the earth minus that reflected and emitted from the earth's surface. Solar radiation is generally available from weather recording stations, however, net radiation is not usually recorded. Several investigators in various regions of the country have shown that for their area net radiation is a linear function of solar radiation. Thus if appropriate parameters are found net radiation may be calculated from solar radiation. This work is an attempt to show the linear relationship exists for this area and find the appropriate parameters for a barley field and bare soil.

Solar radiation and net radiation were collected during a five day period, August 5-9, 1978, at the Agricultural Engineering Research Farm at South Dakota State University near Brookings, SD. Net radiation was measured with a Swissteco net-radiometer and solar radiation with an Epply pyrhelimeter. Data was taken with a crop cover of barley for the first three days and the remaining two on bare soil. (All days were relatively cloud free.) Net radiation was plotted as a function of solar radiation and linear regression was applied to the data. Results of the regression equation are given in Table 1.

Table 1

<u>August</u>	<u>Equation (ly/min)</u>	<u>Corr. Coef.</u>	<u>Slope Std. Error</u>	<u>Intercept Std. Error</u>
5 (cover)	$R_n = 0.823 R_s - 0.090$	.998	$9.29 \times 10^{-3}$	$6.87 \times 10^{-3}$
6 (cover)	$R_n = 0.771 R_s - 0.077$	.981	$2.22 \times 10^{-2}$	$1.77 \times 10^{-2}$
7 (cover)	$R_n = 0.764 R_s - 0.063$	.998	$9.50 \times 10^{-3}$	$8.97 \times 10^{-3}$
8 (bare)	$R_n = 0.716 R_s - 0.103$	.995	$2.20 \times 10^{-2}$	$1.68 \times 10^{-2}$
9 (bare)	$R_n = 0.716 R_s - 0.063$	.998	$1.70 \times 10^{-2}$	$1.18 \times 10^{-2}$

From the analysis of the data and the results acquired net radiation is a strong linear function of solar radiation for the given conditions. Thus these results show promise for calculating values with either barley cover or bare soil with sufficient accuracy for soil model programs.

RELATIONSHIP BETWEEN ANTIPROTEOLYTIC AND ANTIHEMOLYTIC PROPERTIES AND ANTIINFLAMMATORY ACTIVITY OF NONSTEROIDAL ANTIINFLAMMATORY AGENTS

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Nonsteroidal antiinflammatory agents have demonstrated effectiveness in ameliorating the inflammatory response by reducing edema formation, inhibiting enzymes that may cause formation of permeability increasing factors and stabilizing cellular membranes which may prevent release of pro-inflammatory substances. Several quinazolones have been reported to possess antiinflammatory activity and that methylation of a phenyl at position 3 increased the activity (1). These observations prompted the synthesis (2) and evaluation of the antiinflammatory properties of a series of 2-methyl-3-(2'-methyl-substituted phenyl)-6-substituted-4-quinazolones.

The antiedema property was reflected by protection against carrageenin-induced rat hind-paw edema (3) in albino male rats. The substituted quinazolones (100 mg/kg, ip) and indomethacin (15 mg/kg, ip) provided up to 56% protection and 45% protection, respectively. The unsubstituted quinazolone demonstrated the greatest protection from carrageenin-induced rat paw edema. All substituted quinazolones (1 mM) possessed antiproteolytic activity as indicated by *in vitro* inhibition of trypsin-induced hydrolysis of bovine serum albumin (4). The protection against trypsin-induced proteolysis was 20.9% to 40.3% for the quinazolones and 89.9% and 83.5% for indomethacin and sulindac, respectively. The ability to prevent heat-induced hemolysis of canine erythrocytes (5) was used to evaluate the membrane stabilizing property of these agents. The membrane stabilizing property was concentration dependant and biphasic in nature. The dosage required to inhibit 50% of heat-induced hemolysis ( $I_{50}$ ) were determined. Substituted quinazolones demonstrated  $I_{50}$  values from 0.16 mM to 1.0 mM and  $I_{50}$  values for indomethacin and sulindac were 0.02 mM and 0.024 mM, respectively. The results obtained from these experiments have failed to provide any relationship between the antiproteolytic, antihemolytic and the antiinflammatory properties of these nonsteroidal antiinflammatory agents. (Supported in part by the Max Baer Heart Fund of the Dakota State Aerie Fraternal Order of Eagles and The Burroughs Wellcome Fund)

Table 1  
Antiedema, Antiproteolytic and Antihemolytic Properties of  
2-Methyl-3-(2'-Methyl-Substituted\*-phenyl)-6-Substituted-4-Quinazolones

Compound No.	R*	Antiedema Activity % Protection 100 mg/kg	Antiproteolytic Activity % Protection 1 mM	Antihemolytic Activity $I_{50}$ mM
1	H	56	21.2	0.22
2	3'-OH	11	32.3	0.34
3	4'-OH	13	28.1	0.21
4	6'-OH	16	40.3	0.31
5	6-OH	27	28.9	0.33
6	5'-NH <sub>2</sub>	32	20.9	0.34
7	4'-OCH <sub>3</sub>	Nil	23.5	0.16

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## EFFECT OF NEAR SURFACE MOISTURE ON THERMAL EMITTANCE

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The recently launched NOAA5, TIROS N and HCMM satellites all have the capability of monitoring land surface emittance in the thermal infrared region over large areas of the earth. Availability of such data may allow the monitoring of near surface soil moisture if water related effects can be isolated from non-water effects during data analysis. This research is a combined theoretical and experimental investigation with the general objective of understanding the effect of near-surface soil moisture on thermal emittance in order to develop such a data analysis technique.

A finite-difference heat flow model (1) has been modified to allow calculations of the difference in surface temperature to be expected for two plots with different soil moisture profiles but identical in every other respect. Surface temperature differences were then calculated as a function of time for various moisture profiles and the surface temperature differences plotted. Figure 1 shows a typical result of these calculations. This figure shows the dry plot to be warmer during the day and cooler during the night with a zero difference at approximately 0800 hours and 2100 hours. Temperature differences at about 1400 hours and about 0600 hours are very dependent on the difference in soil moisture.

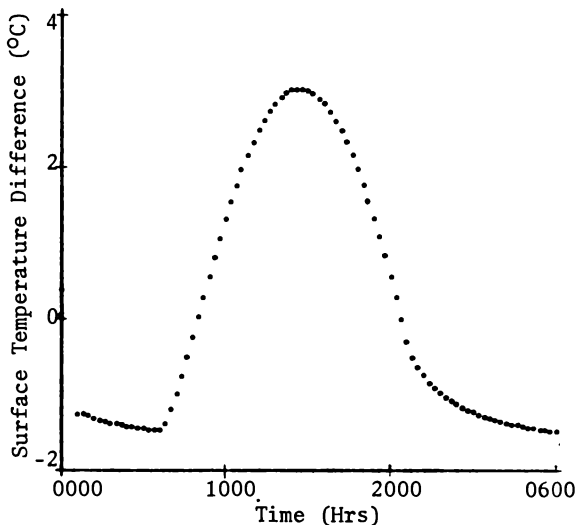


Figure 1: Surface temperature difference between two plots of different soil moisture.

Field experiments were conducted to evaluate the functional form of temperature differences predicted by the model. Data were collected on small barley plots at the Agricultural Engineering Research Farm located near Brookings, South Dakota. One plot was irrigated to increase its soil moisture while the other was left as a dryland plot. Soil temperatures were measured with thermocouples buried at depths of 1, 5, 10, 25, 50 and 100 cm. Thermal emittance was measured for both plots using a Barnes PRT-5, infrared radiometer mounted on a scanning apparatus. Net radiation, incoming solar radiation and other select data were also collected. These data were collected for six successive diurnal cycles. The barley plants were removed from the plots for the last three days of data collection.

Preliminary analysis of these experimental measurements gives considerable support to the qualitative validity of the calculations. If these calculations can be validated, they hold great promise for the development of a method to monitor soil moisture by satellite. Using points on the curve comparing apparent surface

temperature differences at two or more times during the diurnal cycle one could calculate soil moisture differences for a group of chosen sites. If soil moisture is then measured at one site, soil moisture may be calculated for the other sites. This procedure is well suited for utilization of satellites such as NOAA 5 and HCMM since they overpass more than once per diurnal cycle.

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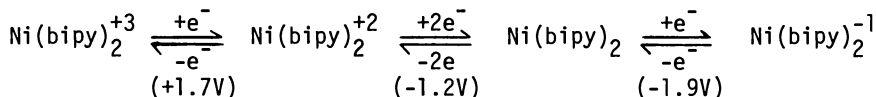
Partial support for this work was provided by OWRT Project No. A-063-S Dak, Agreement No. 14-34-0001-7088 and NASA Contract No. 2406.



## ELECTRON-TRANSFER CHEMISTRY OF ZERO-VALENT NICKEL AND COBALT BIS(BIPYRIDINE) COMPLEXES.

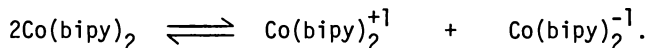
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The electrochemistry of bis(bispyridyl)nickel(0) [Ni(bipy)<sub>2</sub>] and bis(bispyridyl) cobalt(0) [Co(bipy)<sub>2</sub>] complexes, which were prepared by metal atom chemistry,<sup>1</sup> was investigated using cyclic voltammetric and controlled potential coulometric techniques. The electron-transfer chemistry of these complexes was found to be rich with several stable oxidation states observed for both complexes. Cyclic voltammetry studies for Ni(bipy)<sub>2</sub> in acetonitrile indicated four stable oxidation states with reversible couples at +1.7, -1.2 and -1.9 V vs. saturated calomel electrode (SCE). Controlled potential coulometry indicated that the couple at -1.2 as one-electron processes. A scheme for the electrochemistry of Ni(bipy)<sub>2</sub> is as follows:

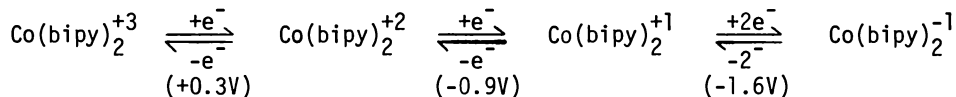


All of the above oxidation states were found to be stable in acetonitrile in the absence of oxygen and water. The solvent, acetonitrile, can act as a ligand with the higher oxidation states (i.e., +2 and +3) of both nickel and cobalt.

Zero-valent Co(bipy)<sub>2</sub> as prepared by hot metal atom chemistry, undergoes an apparent disproportionation reaction in acetonitrile to the (+1) and (-1) oxidation states:



Cyclic voltammetric studies of Co(bipy)<sub>2</sub> in acetonitrile indicated four stable oxidation states with reversible couples at +0.3, -0.9 and -1.6V vs. SCE. Controlled potential coulometry showed that the -1.6V couple was a 2e<sup>-</sup> process with all other processes being one-electron. A scheme for Co(bipy)<sub>2</sub> electrochemistry is:



These data are consistent with reports in the literature on Co(bipy)<sub>3</sub><sup>+2</sup> and Co(bipy)<sub>3</sub><sup>+3</sup> electrochemistry.<sup>2</sup> The synthesis of some of these low-valent complexes by electrochemical techniques will be discussed. Since these low-valent complexes will probably have a planar configuration and are excellent electron donors, mixed-valence salts of these materials have possibilities as electrically conducting solids.<sup>3</sup> Table I is a summary of the electrochemical data on these complexes and includes data on possible electron acceptors, such as tetracyanoquinodimethane (TCNQ) and tetracyanoethylene (TCNE).

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Table I. Peak Potentials in Acetonitrile at 25°C<sup>1</sup>

Redox Couple	E <sub>pc</sub>	E <sub>pa</sub>	ΔE <sub>p</sub>	Redox Couple	E <sub>pc</sub>	E <sub>pa</sub>	ΔE <sub>p</sub>
Co(bipy) <sub>3</sub> <sup>+3</sup> / Co(bipy) <sub>3</sub> <sup>+2</sup>	+0.31	+0.38	70mV	Ni(bipy) <sub>2</sub> <sup>0</sup> / Ni(bipy) <sub>2</sub> <sup>-</sup>	-1.98	-1.90	80mV
Co(bipy) <sub>3</sub> <sup>+2</sup> / Co(bipy) <sub>3</sub> <sup>+</sup>	-0.94	-0.88	60mV	TCNE / TCNE <sup>-</sup>	+0.19	+0.25	60mV
Co(bipy) <sub>2</sub> <sup>+2</sup> / Co(bipy) <sub>2</sub> <sup>-</sup>	-1.57	-1.53	40mV	TCNE <sup>-</sup> / TCNE <sup>-2</sup>	-0.80	-0.70	100mV
Ni(bipy) <sub>3</sub> <sup>+3</sup> / Ni(bipy) <sub>3</sub> <sup>+2</sup>	+1.66	+1.76	100mV	TCNQ / TCNQ <sup>-</sup>	+0.15	+0.21	60mV
Ni(bipy) <sub>2</sub> <sup>+2</sup> / Ni(bipy) <sub>2</sub> <sup>0</sup>	-1.27	-1.21	60mV	TCNQ <sup>-</sup> / TCNQ <sup>-2</sup>	-0.38	-0.32	60mV

1. All peak potentials are in volts vs. a saturated calomel electrode. The solutions are 0.2F tetraethylammonium perchlorate in acetonitrile. The scan rate is 100 mV/sec on a platinum electrode.

## BIOGENIC AMINE MEDIATION OF THE ANTICONVULSANT ACTIVITY OF CHLORDIAZEPOXIDE

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Chlordiazepoxide, a 1,4-benzodiazepine derivative, is noted for its ability to promote tranquilization, relaxation of muscle spasms, relief of anxiety and suppression of convulsive episodes (1,2). Although selective activity of chlordiazepoxide on the central and peripheral nervous system is clearly indicated, the specific involvement of various neurotransmitter substances in the anticonvulsant activity of chlordiazepoxide has not yet been definitely established. Earlier investigations have provided evidence which has indicated a link between benzodiazepine activity and gamma-aminobutyric acid (GABA) mediated neurotransmission (3,4,5). In addition, studies have also indicated a role for glycine (5,6), serotonin (5,7,8) and catecholamines (9,10) in facilitation or inhibition of the central nervous system mechanism of action of the benzodiazepines. The current investigations provide evidence for the possible involvement of the central nervous system biogenic amines as neurotransmitters in the mediation of the anticonvulsant activity of chlordiazepoxide.

Chlordiazepoxide was administered intraperitoneally to male, albino, CF<sub>1</sub>, mice (Sprague Dawley) in doses of 2.5 mg/kg and 5.0 mg/kg two hours prior to the subcutaneous administration of a convulsive dose (ED<sub>100</sub>) of pentylenetetrazol (80 mg/kg). The mice were then observed for a period of 60 minutes for the occurrence of seizures. An episode of clonic spasm persisting for at least five seconds was considered a threshold convulsion. Transient intermittent jerks or episodes of tremulousness were not counted as a seizure. Animals devoid of a threshold seizure during the 60-minute observation period were considered to be protected.

Chlordiazepoxide provided 35% and 68% protection from seizures at the two doses, respectively. The ED<sub>50</sub> value was graphically determined (11) and was found to be 3.40 mg/kg. Pretreatment with intraperitoneal administration of  $\alpha$ -methyl-p-tyrosine (250 mg/kg, 2 hr), phenoxybenzamine (7.5 mg/kg, 1 hr) or propranolol (50 mg/kg, 1 hr) prior to the administration of chlordiazepoxide resulted in the potentiation of the anticonvulsant activity. This was evidenced by a decrease in the ED<sub>50</sub> value of chlordiazepoxide to 1.70 mg/kg, 1.00 mg/kg, and 2.35 mg/kg, respectively. Similarly, intraperitoneal administration of L-Dopa (200 mg/kg, 1 hr) prior to chlordiazepoxide administration also potentiated the anticonvulsant activity as evidenced by a decrease in the ED<sub>50</sub> value, for chlordiazepoxide, to 1.70 mg/kg. These results have provided evidence that the central nervous system biogenic amines may possibly mediate the expression of the anticonvulsant activity of chlordiazepoxide.

(Supported in part by a research grant from the Hoffmann-La Roche Foundation and the Max Baer Heart Fund of the Dakota State Aerie Fraternal Order of Eagles)

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## STUDIES ON ANTIPROTEOLYTIC AND ANTIHEMOLYTIC PROPERTIES OF SOME SUBSTITUTED-2-METHYL-3-(3,4-DIMETHOXY/DIHYDROXYPHENYLETHYL)-4-QUINAZOLONES

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The synthesis of various substituted quinazolone compounds and the ensuing investigation of their possible clinical value (1,2) led to these investigations concerned with the antiinflammatory properties of six substituted-2-methyl-3-(3,4-dimethoxyphenylethyl)-4-quinazolones and six substituted-2-methyl-3-(3,4-dihydroxyphenylethyl)-4-quinazolones (3). The evaluation of the various quinazolones was reflected by their ability to stabilize cellular membranes and/or inhibit proteolytic enzyme activity, since inflammation and central nervous system affects are shown to be altered by agents which provide such stabilization and inhibition.

In the present study, the quinazolones (1 mM) demonstrated very little antiproteolytic activity (Table 1) as indicated by their failure to provide *in vitro* inhibition of trypsin-induced hydrolysis of bovine serum albumin (4). The standard, indomethacin (1 mM), provided 94% protection against proteolysis whereas the substituted quinazolones provided only 0.9% to 18.3% protection (Table 1). Of these compounds, the substituted-(3,4-dimethoxyphenylethyl)-derivatives demonstrated more protection against trypsin-induced proteolysis than did their corresponding substituted-(3,4-dihydroxyphenylethyl)-derivatives.

All of the substituted quinazolones tested (.05 mM), demonstrated *in vitro* membrane stabilizing activity (Table 1) since they provided protection against heat-induced hemolysis of canine red blood cells (5). This antihemolytic activity was concentration-dependant and biphasic in nature. In general, the substituted-(3,4-dimethoxyphenylethyl)-derivatives provided more protection as indicated by a greater reduction of heat-induced hemolysis than the substituted-(3,4-dihydroxyphenylethyl)-derivatives. It has not yet been established, however, that a relationship exists between antiproteolytic and antihemolytic activity of these substituted quinazolones. (Supported in part by the Max Baer Heart Fund of the North Dakota State Aerie Fraternal Order of Eagles and The American Parkinson Disease Association)

Table 1  
 Antiproteolytic and Antihemolytic Properties of Substituted Quinazolones

R*	% Protection§			% Protection†		
	Antiproteolytic Activity	Antihemolytic Activity		Antiproteolytic Activity	Antihemolytic Activity	
	1 mM	.05 mM	.1 mM	1 mM	.05 mM	.1 mM
H	3.5 ± 1.4	44.3	74.1	1.4 ± 0.8	-	-
6-Cl	14.1 ± 1.5	-	-	9.8 ± 3.1	43.7	62.4
7-Cl	11.5 ± 4.2	36.4	61.1	18.3 ± 1.8	75.3	80.4
8-CH <sub>3</sub>	0.9 ± 0.7	34.9	61.4	1.7 ± 0.9	23.3	23.9
6-CH <sub>3</sub>	5.9 ± 2.4	61.1	72.9	15.7 ± 0.8	76.4	73.6
6-I	0.9 ± 0.8	64.5	82.7	14.3 ± 0.8	72.7	63.3

§Substituted\*-2-Methyl-3-(3,4-Dihydroxyphenylethyl)-4-Quinazolones

†Substituted\*-2-Methyl-3-(3,4-Dimethoxyphenylethyl)-4-Quinazolones

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SODIUM AND CALCIUM IN OVERBURDEN, LIGNITE AND UNDERCLAY AT THE BEULAH AND  
BAUKOL-NOONAN MINES, MERCER AND OLIVER COUNTIES, NORTH DAKOTA

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Sodium generally increases with depth while calcium decreases in the overburden, lignite and underclay of the Sentinel Butte Formation (Paleocene) at the Beulah and Baukol-Noonan mines suggesting major chemical effects caused by hydrogeochemical processes. This investigation is part of a Grand Forks Energy Technology Center project to study the character, distribution and origin of the inorganic constituents in lignite. We have examined the chemistry and mineralogy of a stratigraphic section of overburden, lignite and underclay at each mine. Sodium and calcium distributions have been emphasized because of their importance in ash fouling during the combustion of lignite. Chemistry has been determined by rapid analysis of powders using a newly developed electron microprobe technique and mineralogy by x-ray diffraction and electron microprobe analysis.

At the South Beulah mine, 4 samples of overburden above the Beulah-Zap Bed, consist largely of clayey silt grading downward to organic-rich clay. Illite, quartz and montmorillonite are the major minerals and kaolinite, chlorite, plagioclase and mica are present in lesser amounts. Calcite is present in the uppermost sample while dolomite occurs in the other three samples; both decrease in abundance downwards. The Beulah underclay contains major illite, kaolinite and quartz and lesser montmorillonite and calcite. At the Baukol-Noonan mine, 12 samples of overburden above the Hagel Bed and one sample of underclay, are mineralogically similar to the Beulah samples. The lower part of the overburden is silty sand and the upper part, clays and silty clays. Overall, montmorillonite is somewhat less abundant than at Beulah, but is highly variable. Major element chemistry was determined for the overburden and underclay samples from both mines and for a section of eight lignite ash samples from the Beulah-Zap Bed and six samples from the Hagel Bed. Table 1 illustrates the following aspects of sodium, and calcium and Ca/Na variation: (1) generally, sodium increases downward while calcium and CaO/Na<sub>2</sub>O decrease; (2) in overburden, discontinuities with sharp calcium decrease are marked by the disappearance of calcite and/or dolomite; (3) in lignite, sodium and calcium are concentrated in the center of the seam; (4) sodium is higher in the Beulah-Zap Bed and calcium in the Hagel Bed while CaO/Na<sub>2</sub>O averages 1.97 and 2.94 respectively; (5) overall, there is less Na<sub>2</sub>O + CaO in the Beulah-Zap Bed; and (6) sodium and especially calcium and CaO/Na<sub>2</sub>O are low in underclay.

The mineralogy and chemistry of these samples and the observation that the lignites are aquifers suggests that hydrogeochemical processes have strongly affected sodium and calcium distribution by: (1) removal of calcium by dissolution of carbonate minerals in the lower part of the overburden; (2) concentration of sodium and calcium in the central parts of the lignite seams; and (3) increasing the CaO/Na<sub>2</sub>O in the upper parts of the lignite seams and increasing the calcium in the upper part of the Hagel Bed.

Table 1 Sodium and Calcium Variation at South Beulah and Baukol-Noonan Mines

Baukol-Noonan Mine				South Beulah Mine			
Height above lignite in m	CaO normalized	Na <sub>2</sub> O normalized	CaO/Na <sub>2</sub> O	Height above lignite in m	CaO normalized	Na <sub>2</sub> O normalized	CaO/Na <sub>2</sub> O
<u>Overburden</u>				<u>Overburden</u>			
8.3	3.6	0.8	4.50	12.0	7.2	1.8	4.00
7.7	4.6	0.8	5.75	6.0	1.7	1.4	1.21
6.6	5.5	0.8	6.88	4.9	1.8	1.6	1.12
5.5	2.3	1.2	1.92	3.8	0.8	1.5	0.53
4.4	3.0	1.1	2.73	<u>Lignite Ash</u>			
3.4	2.0	1.2	1.67	3.7	15.8	6.0	2.63
<u>Lignite Ash</u>				3.1	20.2	11.2	1.80
2.8	38.7	7.8	4.96	2.4	22.6	9.0	2.51
2.1	29.4	11.8	2.49	1.8	27.5	19.2	1.43
1.5	32.2	11.8	2.73	1.5	25.9	11.1	2.33
0.9	32.1	11.4	2.82	1.2	28.0	17.3	1.62
0.3	26.5	11.8	2.24	0.6	27.0	17.3	1.56
0.1	24.2	10.1	2.50	0.1	14.4	7.8	1.85
<u>Underclay</u>				<u>Underclay</u>			
-0.1	0.5	1.0	0.50	-0.3	0.2	0.7	0.28

SECONDARY PYRITE, BARITE AND GYPSUM ON FRACTURE SURFACES  
IN LIGNITE AT THE BAUKOL-NOONAN MINE, NORTH DAKOTA

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Secondary sulfide and sulfate minerals occur in a fracture zone associated with a normal fault, with about 2 m throw, displacing the Hagel lignite bed of the Sentinel Butte Formation (Paleocene) at the Baukol-Noonan mine near Center, Oliver County, North Dakota. Samples were collected as part of a Grand Forks Energy Technology Center project to study the character, distribution, and origin of the inorganic constituents in lignite (1).

Pyrite occurs as spheroidal nodules up to a few centimeters in diameter and irregular lenses apparently consisting of coalesced nodules. The pyrite makes up about 1% of the volume of a gouge zone about 5 m thick on the hanging wall of the fault. Pyrite was identified as the major sulfide present in the nodules by x-ray diffraction and its composition verified by electron microprobe analysis. Within the nodules, pyrite is concentrated in irregular layers of partially pyritized lignite. Pyrite, identified by crystal form and x-ray diffraction, also occurs within the fracture zone as small crystals formed on coal fracture or cleavage surfaces. Scanning electron microscopy of the pyrite (Figure 1) shows that it occurs as individual octahedra averaging about 40 microns across. The crystals are combined octahedral {111} and dodecahedral {110} forms with the octahedral faces best developed and the dodecahedral faces modifying edges and corners (Figures 1,2). Single and multiple interpenetration twins are common and much of the pyrite occurs as aggregates of separate or twinned crystals.

Barite and gypsum, identified by x-ray diffraction and electron microprobe analysis, and possibly other sulfate minerals occur in the fracture zone. Barite occurs as larger splintery crystals up to about 50 microns long (Figure 1) and as small acicular crystals or fragments, typically 4-8 microns long, on pyrite crystals (Figure 2). Gypsum also occurs as small acicular crystals which are partly embedded in, or project from pyrite crystal faces (Figures 1 and 2).

Sulfide and sulfate distribution in the fracture zone suggests that the fault zone controlled groundwater circulation and that at least two stages of mineralization occurred. In the first stage, under reducing conditions, groundwater carried iron and sulfur into the fault zone and pyrite was precipitated forming nodules and octahedral crystals. In the second stage, under oxidizing conditions, groundwater carried barium and calcium, and barite and gypsum were precipitated in the fracture zone. Gypsum and possibly barite grew on pyrite crystals, a probable source of sulfur.

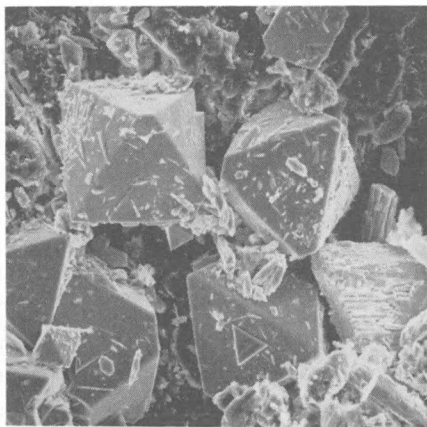


Figure 1. Scanning electron microscope photograph of aggregate of secondary pyrite octahedra and splintery barite crystals (lower right) on lignite fracture surface. Pyrite crystals show development of interpenetration twins and coatings of needle like crystals. Sample BN 2-21. 550X

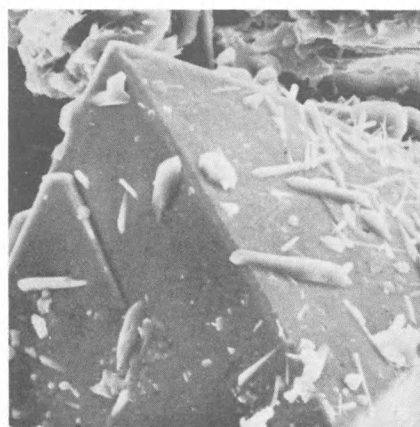


Figure 2. Detail of upper left crystal in Figure 1 showing acicular gypsum crystals on right face and barite on the left (large needle on raised surface). 2000X.

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VARIATION OF FE CONTENT AS A CAUSE OF COLOR CHANGES IN THE  
BULLION CREEK-SENTINEL BUTTE FORMATIONS, NORTH DAKOTA

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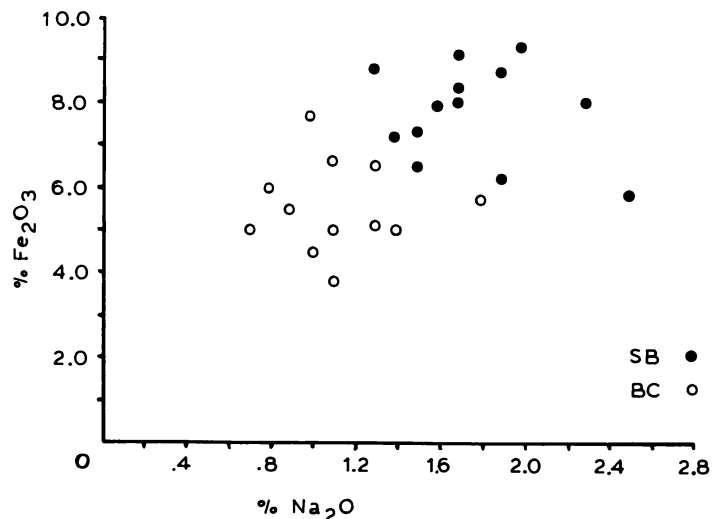
Chemical analysis of twenty-five samples from argillaceous units near the contact zone of the Bullion Creek and Sentinel Butte Formations (Paleocene) indicate a correlation between iron content and striking surface color difference. Although the exact mechanism is unknown, the higher iron and sodium content of the darker colored Sentinel Butte Formation may be the result of poorer drainage in the post depositional environment.

The Bullion Creek and Sentinel Butte Formations are composed of sandstones, siltstones, claystones, and numerous lignite beds. A distinct surface color change from light grays and yellows in the Bullion Creek to dark grays and browns in the overlying Sentinel Butte serves as a boundary marker. The color difference is sharp, mappable in the Little Missouri badlands, and generally independent of rock unit lithology. The colors appear to be a surface weathering phenomenon and is difficult to extend to the subsurface. Samples were collected on the eastern bluffs of the Little Missouri badlands in Billings County, North Dakota. Three sampling sites were selected 32 km north of Medora, near Medora, and 25 km south of Medora. Clay mineralogy and bulk mineralogy of the samples were determined by x-ray diffraction (1). The chemistry was determined by rapid energy dispersive microprobe analysis of bulk powders.

The major oxides present in the samples agree closely with published average analyses of shales primarily of eugeosynclinal origin. Since the Bullion Creek and Sentinel Butte Formations are calcareous and montmorillonitic, the samples are higher than average in CaO and MgO. Although not particularly higher than average in iron and sodium, there are consistent differences in these oxides between the two formations. The Sentinel Butte Formation has a higher total iron and sodium content than the Bullion Creek. The Sentinel Butte ranges 6.2-9.3% and averages 7.8% total iron expressed as Fe<sub>2</sub>O<sub>3</sub>. The Bullion Creek ranges 3.8-7.7% and averages 5.5%. Sodium in the Sentinel Butte ranges 1.3-2.5% and averages 1.8% Na<sub>2</sub>O. The Bullion Creek ranges 0.7-1.8% and averages 1.1%. A plot of iron versus sodium shows a two field distribution (Figure 1). Almost all of the Sentinel Butte samples contain greater than 6.5% iron and greater than 1.4% sodium. Almost all of the Bullion Creek samples are less than these amounts.

The exact mechanism of the iron content and color differences between the two formations is unknown. Iron is an effective coloring agent and is associated with many minerals in these sediments. Factors affecting the intensity and hue of coloration by iron include particle size, oxidation state, degree of dissemination, and total amount present. It appears there is a correlation between total iron content and the darker colors of the Sentinel Butte Formation. This could be the result of an environment with poorer drainage than the Bullion Creek Formation. The poorer drainage would produce reducing conditions which could result in the retention of iron and sodium and also maintain small particle size of the iron minerals in the Sentinel Butte Formation. This preconditioning of the sediments at the time of deposition is reflected in the darker colors observed in outcrop.

Figure 1. Percent total iron, expressed as Fe<sub>2</sub>O<sub>3</sub>, plotted against percent Na<sub>2</sub>O for all analyzed samples. SB, Sentinel Butte Formation; BC, Bullion Creek Formation.



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EFFECTS OF AGRICULTURAL LAND USAGE PATTERNS  
ON SHALLOW PRAIRIE LAKE ECOSYSTEMSJ. A. Olsen\*, D. Z. Hopewell, and W. R. Dorband  
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Runoff from agricultural cropland and pastureland is an important non-point source (NPS) pollutant to streams, lakes, and wetlands in the Northern Plains. This NPS pollution degrades water quality, destroys fisheries, reduces water based recreation, and increases the eutrophication rate of many watersheds. The major effects of agricultural runoff pollution are caused by sediment carried overland following storm events or through irrigation return flow, and by the input of dissolved nutrients associated with the runoff. This report describes a preliminary ecological study of several small lakes, in well defined drainage basins where the entire watershed of each lake was influenced by a single type of land usage pattern.

Four shallow lakes (max. depths < 3m) in different types of watersheds, 25-30 miles west of Sioux Falls, S.D., were chosen as study sites: 1) Muchow Lake, a 1.3 hectare impoundment within a 9.7 hectare basin used as a brome pastureland for cattle; 2) Dubbe Lake, a 4.0 hectare lake in a 13.5 hectare basin being used exclusively for dryland agriculture; 3) Retriever Lake, a 1.0 hectare wetland in a 17.5 hectare basin drained by planted alfalfa, which is used as a training location by hunters and their dogs; and 4) A-Frame Lake, a 25.0 hectare lake in a 68.5 hectare drainage basin which is a S. D. Wildlife Production Area, and is relatively undisturbed by agricultural activity. All four sites were sampled biweekly, from May-October, 1978. Zooplankton, phytoplankton, and physical-chemical parameters were collected at several locations from each site during each sampling.

The four lakes studied could be classified as mildly eutrophic by their nutrient levels and dissolved ion concentrations. Three of the four sites had conductivities ranging from 800-1000  $\mu\text{mhos/cm}$ . Relatively undisturbed A-Frame Lake had significantly lower conductivity levels (400-600  $\mu\text{mhos/cm}$ ) through the sampling period. Total alkalinities were lower (75-120 mg/l) and pH values were higher (7.0-9.5) for the two lakes drained by grazed pastureland and dry land agriculture, than for the other two sites (pH values, 6.5-7.5; tot. alk., 160-230 mg/l). Nitrate levels were somewhat unexpectedly low (< 0.01-0.1 mg/l) for all four sites, while significantly higher ( $p < 0.05$ ) ortho-phosphate levels (0.663-4.788 mg/l) were observed at the relatively undisturbed site, A-Frame Lake, than at the other three sites (0- $\text{PO}_4$  range, 0.007-2.754 mg/l). Total suspended solids loadings to the four sites were variable (0.9 -64.3 mg/l) and seemed to increase drastically following high intensity storm events.

Seasonal changes in water quality through the sampling period were gradual and expected. Most dissolved ions decreased in concentration with biological utilization and sedimentation through the late spring and summer period. However, the macronutrients, nitrogen and phosphorous, did not change appreciably in concentration through the sampling period at any of the four sites. Thermal stratification occurred in only one of the lakes (Muchow Lake) and it was only mild (25 $^{\circ}$  surface - 20 $^{\circ}$ C bottom). Oxygen concentrations throughout the water columns remained high at all sites through the sampling period.

The zooplankton communities at all four sites were similar in composition and density throughout the summer. Copepods were numerically dominant early in the sampling period, while cladocerans became dominant through the late summer.

Benthic communities in the four lakes reflected bottom substrate characteristics. Silt-clay substrates in Muchow and Retriever Lakes supported a chironomid larvae dominated community. Ostracods of several species were also abundant at these two sites. In Dubbe Lake, the bottom was almost completely covered with epipellic algal mats, and the macroinvertebrate benthos was dominated by herbivorous snails (*Physa* and *Gyraulus*). In A-Frame, the bottom substrate was a fine ooze of decomposing organic matter which supported a very sparse benthic community of several chironomid species. Oligochaetes were conspicuously absent from all four sites.

Comparisons of relationships between physical-chemical and biological parameters at the four sites did not reveal a clear picture of any apparent effects of agricultural NPS pollution. The unique natural geologic character of each drainage basin contributed significantly to the water quality and biological composition of these lakes. As a hypothesis which requires more study to validate, it appears that naturally eutrophic prairie lakes have a natural buffering capacity which allows them to maintain stable following mild environmental perturbations.

## THE MACROSCOPIC BENTHOS OF LAKE COCHRANE, SOUTH DAKOTA

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A quantitative survey of the macroscopic bottom fauna of Lake Cochrane was made between April 1976 and December 1976. Lake Cochrane is a small (148ha) moderately eutrophic lake (1) in eastern South Dakota. It has a maximum depth of 7.9m and a mean depth of 3.9m (2).

Three sampling stations were selected based on differences in depth and bottom type. The bottom at station 1 (depth 3m) was composed of mud with large amounts of detritus and mollusc shells. The bottom at station 2 (depth 2m) was composed of gravel, sand and mud with large amounts of detritus and mollusc shells. The bottom at station 3 (depth 7m) was composed of mud with little detritus or mollusc shells.

Four Eckman dredge (232cm<sup>2</sup>) hauls were taken at each station. Each sample was washed through a 30 mesh per inch brass screen and preserved in 10% formalin. Organisms were hand sorted using a binocular microscope. Dry weights were determined after oven drying at 95-100° C for 24 hours.

Analysis of variance showed that the stations differed from each other significantly in terms of the numbers of various organisms present and amount of standing crop. Station 1 had the highest mean seasonal numbers (13,631/m<sup>2</sup>) and mean seasonal standing crop (2.1227gm/m<sup>2</sup>) for total macrobenthos. Station 2 had the lowest mean seasonal numbers (9618/m<sup>2</sup>) and mean seasonal standing crop (.8858gm/m<sup>2</sup>). Station 3 had seasonal means nearly as high as station 1 with numbers of 13,452/m<sup>2</sup> and a standing crop of 1.6743gm/m<sup>2</sup>.

Three species dominated the benthos of Lake Cochrane. Together they made up 92.3% (11,287/m<sup>2</sup>) of the mean seasonal numbers and 91.5% (1.4284gm/m<sup>2</sup>) of the mean seasonal standing crop from all stations. They were in order of importance numerically Chaoborus punctipennis (56.5%), Cryptochironomus "harnischia" group (24.5%) and Chironomus attenuatus (11.3%) and gravimetrically C. punctipennis (42.3%), C. attenuatus (39.4%) and Cryptochironomus "harnischia" group (9.8%).

A study of the life cycles of the major species indicated that Chaoborus punctipennis had one generation per year, spent the winter in the fourth instar and was most abundant at the deepest station. Chironomus attenuatus appeared to have three generations with the fall emergence involving only part of the population. The majority of the larvae spent the winter in the fourth instar. Greatest numbers were found at the station of intermediate depth. Cryptochironomus "harnischia" group appears to have three generations per year. The larvae spend at least part of the winter in the third instar in an inactive state. They were most abundant at the station of intermediate depth but were also abundant at the shallower station.

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DIFFERENTIAL DEGRADATION RATES OF TECHNICAL AND  
FORMULATED CARBOFURAN IN SOIL

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Tests were performed under controlled laboratory conditions to determine degradation behaviour of 10% granular carbofuran (FURADAN 10G <sup>®</sup> FMC) and 99.8% pure technical carbofuran in soil samples labelled as S-1, S-2, S-3 and S-4 collected from 4 different experimental sites near Waseca, Minnesota. Soil sample S-1 was taken from an experimental plot where carbofuran had failed to provide effective control of corn rootworm larvae. On the other hand, sample S-2 was taken from an experimental plot where insecticide performance was rated as satisfactory in field trials. Soil samples S-3 and S-4 were from control field plots with no history of insecticide application. These soils contained 46.4, 30.8 and 22.8% of sand, silt and clay respectively. Soil pH varied between 6.4 and 6.6. Soil moisture content was 19.8 to 21.5%. Subsamples of soils weighing 50 and 10 g respectively were transferred to clean conical flasks (30 subsamples of 50 g and 15 subsamples of 10 g soil) for each of the 4 soil samples collected. All of the 50 g subsamples from S-1, S-2, S-3 or S-4 were fortified with 35 ppm of granular formulation while the 10 g soil subsamples were fortified with 8 ppm of technical carbofuran in acetone. All samples were then incubated at  $22 \pm 2^\circ\text{C}$ . Residues of carbofuran were analyzed at time intervals of 0, 7, 14, 28, and 35 days from the three treated soil subsamples, the untreated soil samples was also analyzed to determine the presence of carbofuran. Samples were extracted by the method of Cook et al (1969) and extracts were assayed with a Hewlett-Packard 5710A gas chromatograph equipped with Nitrogen-Phosphorus flame ionization detector (Alkali Flame) and a glass column (183 cm long x 3 mm O.D.) prepacked with 5% DC 200 on 100/120 mesh chromosorb W-HP (Hewlett-Packard Avondale, PA). The GLC conditions were the following: Temp. in  $^\circ\text{C}$ -- detector 300, isothermal column oven 165, and injection port 200, and the gas flow rates carrier gas Helium 30 ml/min. at 55 PSIG, detector gas Helium:Hydrogen (91.5:8.5%) mixture 36 ml/min. at 30 PSIG and air 50 ml/min. at 40 PSIG. Under these conditions 20 ng of carbofuran/ $\mu\text{l}$  in acetone produced a recorder response of 30-40%.

Our results show significant differences ( $P < .01$ ) in the degradation rates of carbofuran 10% granular formulation and in the technical material. Rapid dissipation of the technical carbofuran was evident in soil samples from 4 different experimental sites. More than 50% of the insecticide was lost within seven days after incubation in soil samples S-1, S-2 and S-3 while 14 days were required for 50% disappearance of technical carbofuran from sample S-4. Only 15, 19, 18 and 21% of applied technical carbofuran was present on the 21st day after application. On the contrary when carbofuran was applied as a granular formulation it dissipated from the soil samples at a very slow rate. For example, more than 75, 63, 70 and 65% of the applied dosage of the carbofuran formulation could be detected after 35 days incubation in S-1, S-2, S-3 and S-4 soil samples respectively. Thus, our data suggests that formulated carbofuran is much more persistent in soil than the technical material. Two way analysis of variance was used to compare degradation rates of the carbofuran in 4 soils and the effect of incubation time. Results of analyses showed no significant differences in the degradation of carbofuran in soil samples from different sites. However, highly significant ( $P < .01$ ) F-values indicated that residue levels were significantly different from each other at different time intervals. Our residue results indicate that carbofuran was equally persistent at experimental site 1 and site 2. Therefore, its failure to provide effective rootworm control may have been due to reasons other than its rapid degradation. Furthermore, if the performance inconsistencies of the carbofuran at S-1 were due to the presence of certain bacterial populations, the soil residue levels should have been influenced significantly under controlled laboratory conditions.

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EFFECT OF THE ETSI COAL SLURRY PIPELINE ON WATER RESOURCES  
IN WYOMING, SOUTH DAKOTA, AND NEBRASKA

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In 1974 the State of Wyoming gave permission to Energy Transportation Systems Incorporated (ETSI) to develop a well field in Niobrara County, Wyoming. The anticipated withdrawal of 15,000 acre-ft/yr (equivalent to 20.7 cfs or 9,300 gpm) would be obtained from about 40 wells in the Madison Limestone. The water would be sent in a 38 inch pipeline to the Gillette area, mixed with crushed coal, and the coal slurry pumped about 1,400 miles to Arkansas.

Three test holes were drilled by ETSI to the Madison in 1974, and were test pumped at 57 to 180 gpm. Using values of transmissivity (about 6,400 gpd/ft) and storage (about 0.000,065) derived from the pump test, predictive models using conventional theory show a lowering of the piezometric surface which would ultimately develop after the 45 year life of the project. A study by Rahn (1975) shows that a cone of depression would spread over 50 miles from the ETSI site. This prediction was further documented by University of Wyoming geologists (Huntoon and Womack, 1975) who show, for example, a drawdown of 500 feet at the town of Edgemont, South Dakota. (This would cause the artesian wells in Edgemont to cease flowing.) A computer study by the U. S. Geological Survey confirms these predictions and includes data showing the effects on springs (Konikow, 1976). Cascade Spring, probably South Dakota's largest spring, could be reduced 4 cfs in its discharge. It is not known to what degree the ETSI project would affect other springs in the area, some of which discharge into the Platte and Niobrara Rivers which drain into Nebraska. The loss in pressure may ultimately have effects on overlying aquifers, because studies by the U. S. Geological Survey show that the Madison may be a source of recharge to the Lakota Formation (Gott, et al, 1974) or the Dakota Sandstone (Swenson, 1968).

The quality of water in the Madison in the project area appears to be reasonable good. The total dissolved solids from water samples at the test site averaged 550 ppm. This is nearly the same as the Missouri River in the Dakotas for which massive diversion schemes such as the Garrison and Oahe Irrigation Projects and the West River Aqueduct have been proposed.

Adequate provisions should be made to protect the water in areas affected by this project. Water from the Madison Limestone has considerable potential for local use even though the depth prohibits its use in some areas due to high drilling cost at present.

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## STRENGTH AND GENERALITY OF PREFERENCE FOR IMMEDIATE STRESSOR

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Imagine for a moment that you are seated in my lab at Concordia College, with a shock electrode attached to your wrist. You have but one choice. You may delay the shock for 30 seconds (sec), or you may get the shock over immediately...But let's change the situation. Now the immediate shock lasts 3 sec while the delayed shock lasts only 1 sec. Now what are your choices? Immediate? Delayed? This is the procedure I have been using in recent studies. It has several advantages. First, it eliminates the problem of ceiling effects, which may have obscured independent variables in previous research. (In Bertilson & Dengerink (1975), the 1-sec versus 1-sec condition resulted in immediate choices 83% of the time.) Second, a larger immediate punishment may more closely approximate the phenomenology of people in the real world -- going to the dentist today to have that tooth filled is more real (thus larger) than going to the dentist next week (thus smaller).

An anxiety mediation hypothesis has been suggested by several authors -- that people choose immediate shock to avoid anxiety that would otherwise occur while waiting for the delayed shock. This hypothesis has been confirmed by Bertilson, Meyer, and Meyers (1978). Subjects who were relaxed, by listening to a relaxation tape for six minutes in a dimly lit room, were able to put off selecting the immediate 3-sec shock (by choosing the 30-sec delay and receiving only a 1-sec shock) for more trials than untreated control subjects, who almost always chose the immediate 3-sec shock on the first trial.

When faced with an inevitable stressor, most people prefer to get it over with immediately. Only two classes of people do not seem to prefer the immediate stressor -- children and psychopaths. This stress choice research is of particular interest for several reasons. First, it may tell us something about the development of anxiety in humans and how the anxiety mediates choice behavior. Second, it may tell us something about psychopathy. The series of experiments reported in this paper investigated the strength and generality of this preference. As is common in this research, subjects were asked to choose between an aversive event immediately or an aversive event delayed thirty sec. The strength of the preference for the immediate stressor was measured by asking subjects in different treatment groups to choose between larger stressors immediately versus smaller stressors delayed thirty sec. The generality of the preference for immediate stressors was evaluated in separate experiments by systematically varying duration of shock to the wrist, intensity of shock to the wrist, and different concentrations of the unpleasant tasting drink, quinine sulfate.

In Bertilson & Dengerink (1975), subjects in one group chose between a 1-sec shock immediately versus a 1-sec shock delayed 30 sec. Other groups of subjects chose between 2-sec versus 1-sec shocks and 3-sec versus 1-sec shocks. In the first study of the present research, subjects chose between shocks that were 100% of the voltage that subjects judged to be unpleasant versus 90%. Subjects in other groups chose between 100% versus 80% and 100% versus 70%. In the second study of the present research, subjects chose between 1-tablespoon drinks of quinine sulfate solution. The choices in one group were solutions containing .062 grains of quinine sulfate versus .062 grains delayed 30-sec. The other group chose between .062 grains now versus .031 grains delayed. As can be seen from the table, the preference for an immediate aversive event is both strong and general. Almost half the time, subjects chose an immediate event that was more aversive than the delayed event. This was true whether the choice was between durations of shock, intensities of shock, or concentrations of quinine sulfate.

TABLE

Percentage of Immediate Choices

Duration conditions choices	1-1 83%	2-1 61%	3-1 46%
Voltage conditions choices	100% - 90% 84%	100% - 80% 84%	100% - 70% 63%
Quinine sulfate choices	.062 - .062 95%		.062 - .031 48%

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## REPRODUCTIVE SUCCESS OF GIANT CANADA GEESE IN WESTERN SOUTH DAKOTA

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The South Dakota Department of Game, Fish and Parks, in cooperation with the U.S. Fish and Wildlife Service began a program in 1967 to restore the giant Canada goose (Branta canadensis maxima) in South Dakota (Pittman-Robertson Prog. No W-75-R-17, South Dakota Dept. Game, Fish and Parks, Pierre).

From 1967 through 1974, 1,528 free flying geese were released in the western half of the state. Six hundred forty-six were released on the study area which included the counties of Jackson, Haakon and portions of Pennington and Jones County. The goal of the restoration project was to provide a breeding population of giant Canada geese for people to hunt and to observe in a scientific and aesthetic sense. The objective of my study was to investigate the production of this flock.

Nests were found by searching near release sites and previous nest locations. Replies to newspaper solicitations and reports from conservation officers and landowners contributed additional information about nests and pairs of Canada geese. Nests were examined to determine clutch size and fate of eggs. Date of nest initiation was estimated by back dating, using an incubation interval of 28 days (1) and a 1.5 day interval for each egg laid (2).

One hundred fifty-nine nests were located during the nesting seasons of 1974 and 1975. First nests were started on 1 April 1974, and 8 April 1975. The last known nests were initiated 14 May 1974 and 21 May 1975.

Nests were located near intermittent streams, man made dugouts and stockponds (water impounded by small dams). A few natural wetland basins existed on the study area but were either dry or not used for nest locations by geese.

One hundred fifty of 159 nests (94 percent) were associated with stockponds. Two nests were associated with dugouts and seven nests were located on stream banks. One nesting pair of geese per water body was the general rule. Some exceptions were found when islands were present and on two ponds, both exceeding 3 ha, two nesting pairs were present. The large size and irregular shape of these stockponds evidently contributed to multiple nesting.

One hundred thirty-four nests (84 percent) were located on natural nest sites. All known islands on the study area were used for nesting. Other natural sites of nests were peninsulas and the shoreline of stockponds. Natural nest sites were located close to visible water. Mean horizontal distance of nests from water was 9.1 m. In all cases, a permanent water body could be seen from the nest site.

Artificial nesting structures were also present on some study area stockponds. Nest boxes attached to floating platforms and on posts in stockponds were the two types present. Forty percent of all usable structures contained nests during the 2 years. This amounted to 16 percent of the total nests found.

The mean clutch size of incubating nests was  $5.27 \pm 0.11$  eggs. Fifty-seven percent of all nests had at least one egg hatch. Nests on artificial structures and islands had similar hatching success rates of 64 and 63 percent respectively. Peninsula nest sites were less successful (54 percent) and shoreline nests had the lowest success rate (38 percent).

Four hundred sixty-one goslings were produced from known nests during the two years. Two hundred forty goslings were observed periodically from time of hatching to flight stage. Mortality of these goslings was 16.2 percent. Gosling mortality of this population may be greater because I was unable to locate numerous other broods that had hatched from known nest sites.

Production of Canada geese in western South Dakota may be enhanced by including islands in the design of future stockponds. Island construction may also be considered on existing stockponds during years of low water. Construction of artificial nesting structures may also be considered to encourage reproductive success.

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SULFUR CAPTURED BY SORBENT INJECTION  
IN AN ATMOSPHERIC FLUIDIZED-BED COMBUSTOR

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Atmospheric Fluidized-Bed Combustion (AFBC) of coal offers several important advantages over conventional combustion. The most significant advantage is the ability to control the emission of sulfur dioxide (SO<sub>2</sub>) by reacting it with alkaline materials in the fluidized region to form alkali sulfates, which are either retained in the combustion zone or emitted as solid particulate.

The Grand Forks Energy Technology Center (GFETC) as part of its continuing research on low rank coals and lignites, is operating a 6-inch I.D. AFBC test facility. The test program is designed to evaluate the ability of various low rank coal ashes to absorb the SO<sub>2</sub> produced. The alkali in these coals is primarily calcium, magnesium, and sodium.

Northern Great Plains lignites are particularly well suited for AFBC, because many have alkali-to-sulfur stoichiometric ratios greater than 1:1. These coals could theoretically exhibit sulfur retentions of 100 percent, although most actually retain between 40 and 60 percent of the coal sulfur when burned in the GFETC AFBC.<sup>1</sup> Retention of this amount of SO<sub>2</sub> by the inherent coal ash during AFBC allows nearly all of the coals tested at GFETC to meet the current Federal New Source Performance Standards (1.2 LB SO<sub>2</sub> emitted per million Btu input) without added sulfur sorbents. However, it appears likely that with the revised NSPS for SO<sub>2</sub>, the addition of some SO<sub>2</sub> sorbent material will be required.

To determine the effects of sorbent addition on sulfur retention in the AFBC, a series of tests was conducted on three sorbent materials: 1) Limestone obtained from the Davenport, Iowa area with 50 percent available CaO; 2) Trona, an impure form of hydrous sodium carbonate Na<sub>2</sub>CO<sub>3</sub>·NaHCO<sub>3</sub>·2H<sub>2</sub>O; 3) Nahcolite, a mineral whose primary constituent is NaHCO<sub>3</sub>. Nahcolite is associated with oil shale deposits and is not currently available. Two test coals were selected: 1) Decker, MT subbituminous; 2) Beulah, ND lignite.

The GFETC 6-inch I.D. AFBC has a combustor shell that is a 10-foot high stainless steel pipe with internal cooling coils to control bed temperature. Two positive displacement blowers provide combustion air and control combustor static pressure. The bed material used for these tests was either 20-mesh (US) quartz sand or 30-mesh (US) Al<sub>2</sub>O<sub>3</sub> grain, as noted in the results. Coal and sorbent were metered by volumetric screw feeders and transported into the fluidized combustion zone by an air ejector. Two cyclones and a fiber filter provide particulate control for the exhaust gas. Flue gases were continuously monitored for oxygen, carbon monoxide, carbon dioxide, nitric oxide, and sulfur dioxide.

Test results are shown in the table that follows. All tests were conducted at an average bed temperature of 1500°F and a fluidizing velocity of 7 feet per second. The test results demonstrate that all three sorbents are effective in reducing SO<sub>2</sub> emissions. The sodium compounds Trona and Nahcolite were more effective than the limestone.

Run #	Coal	Sorbent	Ratio of Added Alk to S	Excess Air, pct	Sulfur Retained, pct	Sulfur Dioxide Emitted LBS SO <sub>2</sub> /10 <sup>6</sup> Btu	Bed Material
GS-116A-78	Decker	--	--	19.6%	59.7%	0.27	SiO <sub>2</sub>
GS-126-79	Decker	--	--	19.0	57.1	0.29	SiO <sub>2</sub>
GS-121-79	Decker	Trona	0.5	19.2	82.8	0.12	SiO <sub>2</sub>
G-125-79	Decker	Trona	2.0	21.0	94.2	0.04	Al <sub>2</sub> O <sub>3</sub>
GS-119-79	Decker	Nahcolite	0.5	18.7	86.5	0.09	SiO <sub>2</sub>
GS-120-79	Decker	Nahcolite	0.9	20.0	89.1	0.07	SiO <sub>2</sub>
GS-118-79	Decker	Limestone	1.3	19.0	87.0	0.09	SiO <sub>2</sub>
GS-127-79	Decker	Limestone	1.7	18.6	87.7	0.08	SiO <sub>2</sub>
GS-104-78	Beulah	--	--	27.2	60.5	1.08	SiO <sub>2</sub>
GS-108-78	Beulah	Trona	1.0	28.4	99.0	0.005	SiO <sub>2</sub>
GS-109-78	Beulah	Nahcolite	1.0	29.3	98.2	0.05	SiO <sub>2</sub>
GS-111-78	Beulah	Limestone	1.0	31.6	79.8	0.55	SiO <sub>2</sub>

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## COLLEGIATE COMMUNICATIONS

The 1979 Annual Meeting of the Dakota Academies of Science, a special joint meeting of the North Dakota Academy of Science and the South Dakota Academy of Science, contained a Collegiate section where a total of thirty-three presentations were made by undergraduate and graduate students. The students were reporting the results of their own research activities, usually under the guidance of a faculty advisor. The students were required to prepare a Communication similar to those prepared for the professional presentations. These Communications all are published in this special section of the Proceedings.

Readers are referred to the Author Index of this volume in order to locate any particular Communication.

DETECTION OF LABILE METAL BINDING LIGANDS BY MODIFIED GEL FILTRATION CHROMATOGRAPHY  
ITS LIMITATIONS AND ADVANTAGES

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Trace metals and their ligand counterparts are very important in human nutrition (1). Detecting trace amounts of these metal-ligand complexes in biological fluids has been a perplexing problem for biochemists and nutritionists for a number of years now. In the past, classical gel filtration chromatography has been used to study these labile metal-ligand complexes and false results were obtained (2).

Many of the problems can be attributed to the fact that the metal ions are both absorbed and retarded on the column substrate. In this paper this absorption phenomenon is studied using widely used gels.

Then, there is also a base line study of a new technique called modified gel filtration chromatography. In this technique the metal-column substrate absorption-retardation interaction is held in check by keeping a constant metal concentration called  $[M]^0$  in the solvent system. The chemistry of the system will be explained.

In using modified gel filtration chromatography many problems involved with the detection of metal-ligand complexes are solved. However it does generate new problems that must be controlled. These problems and their control will be explained.

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THE USE OF TAURINE FOR CONTROLLING THE  
CONTRACTILITY AND IRRITABILITY OF HEART TISSUE\*\*

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Taurine is a free amino acid present in all animals. Its functions are believed to be (1) to aid in the formation of bile salts in certain mammals which then aids in lipid digestion, (2) to aid in the maintenance of intracellular osmotic pressure, and (3) its physiological role may be to maintain excitatory activity in muscles and nerves of mammals.

It is hypothesized that increasing work of the heart stimulates an increase of taurine formation which then tries to bring the body back into a homostatic condition by acting as a regulator of contractility of heart muscles. In these experiments frogs were kept in different thermal environments (4°C, 18°C, 25°C) creating various stressful situations. Taurine levels were measured on the extracted heart using the ninhydrin reaction.

In the northern grass frog (a hibernator) there is a seasonal variation in the taurine content of the heart. From a previous study the highest level of taurine occurs in July. The results of this study show that taurine levels decrease in the winter months, the lowest occurring in January. However, not only the seasons but also environmental temperatures affect taurine levels. High temperatures raise the level and low temperatures decrease the level. It was also found that the frog heart can synthesize taurine from cysteine and pantothenic acid at temperatures of 16°C but not at higher temperatures. Thus formation of taurine in the frog heart depends on several factors.

\*\*This research work was done under the supervision of Dr. W.O. Read and Don Niebel, University of South Dakota, Vermillion, during the January 1979 Interim.

## MOLAR ABSORPTIVITY OF Co(II) EDDA (ODII) COMPLEXES\*\*

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Co(III) EDDA (dye) compounds are very difficult to crystallize and thus have been difficult to characterize. In order to determine the molar absorptivity of compounds of this type, Co(III) EDDA (ODII), a more well-characterized dye, is used as a model system. From chemical analysis it has been determined that there is one Co<sup>3+</sup> for every dye molecule. The molar absorptivity for this dye is also known.

By reducing the Co(III) EDDA (ODII), a free Co(II), EDDA and OD(II) are produced. Using column chromatography these components are separated and a standard percentage of OD(II) recovered is quantified. The molar absorptivity of an unknown dye is calculated from the spectrally determined absorption and the molar concentration of the cobalt as determined by Co<sup>3+</sup> analysis and quantified by the percentage recovery of OD(II) from Co(III) EDDA (ODII).

\*\*This research was done under the direction of Dr. Ivan Legg during the summer of 1978 in an NSF-URP study at Washington State University, Pullman.

COMPARISON OF LIVER MITOCHONDRIA  
IN CARDIOMYOPATHIC AND NORMAL HAMSTERS

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Cardiomyopathic hamsters (BIO 14.6) normally die of heart failure between four to nine months of age. Although the reason for their predisposition toward heart failure is not known, heart mitochondria from these animals have increased levels of calcium (1), as well as decreases in rates of respiration, respiratory control ratios (RCR), and efficiency of ATP synthesis (ADP/O ratio) as compared to normal hamsters (2).

In this study, liver mitochondria from myopathic hamsters were studied to see if these differences are restricted to heart tissue, or reflect a generalized disturbance in mitochondria from other tissues. The liver mitochondria showed no significant differences with respect to RCR and ADP/O ratios as compared to random bred controls. However, the cytochrome oxidase content ( $a+a_3$ ) was twice that of control hamsters. The relationship of this increase to cardiac dysfunction is not apparent.

<u>Hamsters</u>	<u>Cytochrome Content</u>			<u>Cytochrome Ratios</u>
	(nanomoles/mg protein)			
	<u>b</u>	<u>c+c<sub>1</sub></u>	<u>a+a<sub>3</sub></u>	
180 day myopathic	0.11	0.15	0.084	1 : 1.34 : 0.74
300 day myopathic	0.18	0.25	0.10	1 : 1.39 : 0.56
300 day random bred	0.18	0.26	0.05	1 : 1.44 : 0.28

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<sup>†</sup>This experiment was performed under the direction of Dr. John A. Thomas, Biochemistry Section, The University of South Dakota School of Medicine, Vermillion, S.D. 57069.

Particle Size Determination From Settling Rates

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As part of an independent study course in fluid mechanics to improve my preparation in the joint engineering program between Augustana College and Washington University in St. Louis, I did some elementary experimentation on determining particle size based upon measurements of settling rates. An explanation is given of the formula which identifies terminal velocity of a settling particle by equating the Stoke's resisting force of a particle by a supporting fluid to the gravitational force. Experiments are first done with small plastic spheres of uniform size where the size can be verified through measurement with a micrometer. The method is extended to irregularly shaped particles having a distribution of sizes. In this case the measurement results in the determination of the amount by volume in different increments of sizes.



NEASCUS PYRIFORMIS, THE BLACK-SPOT TREMATODE OF  
FISHES IN BRULE CREEK, SOUTH DAKOTA

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A total of 2,482 fishes of 10 species from Brule Creek in Southeastern South Dakota were examined for black-spot. Seven of the 10 species were found infected with the trematode larvae. The number of fish of each species examined and the prevalence of infection was as follows: fathead minnow (Pimephales promelas), 726 of 1,311 (55%); creek chub (Simotilus atromaculatus), 213 of 272 (78%); plains minnow (Hybognathus placita), 83 of 167 (50%); white sucker (Catostomus commersoni), 14 of 46 (30%); stone roller (Campostoma anomalum), 15 of 21 (71%); common shiner (Notropis cornatus), 3 of 15 (20%); and red shiner (Notropis luterensis), 6 of 8 (75%). No infections were found in 244 sand shiner (Notropis stramineus), 392 bigmouth shiner (Notropis dorsalis), or 6 Johnny darter (Etheostoma nigrum).

There was no correlation between the prevalence and the intensity of infection among the different host species. For example, there was 71% infection in stone rollers with a range of 1 to 2 cysts/infected fish, whereas in fathead minnows there was only 50% infection but with a range of 1 to 30 cysts.

Based on morphological studies, the strigeoid metacercaria causing the black-spot was identified as Neascus pyriformis. This parasite was originally described from fishes in Lake Itasca, Minnesota (1). This is the first report of the parasite from fishes in South Dakota, although it has been reported from fishes in North Dakota (2).

1. Chandler, A.C. (1951) Am. Midl. Nat. 45: 711-721.
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FISHES OF THE SIOUX FALLS RIVER PARK AREA

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This taxonomic study was initiated in December of 1977 and was concluded in January of 1979. The purpose of the study was to establish baseline data on the fish inhabiting the Sioux Falls River Park area. The paper provides an annotated list of 25 species of fish representing four orders and ten families inhabiting the Sioux Falls River Park area. The majority of this area being the Big Sioux River or directly contiguous with the Big Sioux River, such as the connected oxbow lakes and the mouths of tributaries, within the city limits of Sioux Falls. The fish were collected by various means depending on the specific habitat and season of the year. A new species to the Big Sioux River (1), the golden shiner, Notemigonus crysoleucas, was collected in abundance in the oxbows and was definitely my most interesting find. This study also describes the individual habitats in which the fish were found as well as the relative abundance of each species.

1. Nickum, John G. and James A. Sinning (1971) Proc. S.D. Acad. Sci. 50, 143-154.

## Swimming Ability of the Northern Pocket Gopher

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Since little is known concerning the swimming ability of the northern pocket gopher, (Thomomys talpoides), this continuing study was initiated. Northern Pocket gophers were captured in live traps and swum in a wooden trough (2.4m by .3m). There was no significant difference noted in swimming ability due to variables of age, sex, weight, and water temperature. The average swimming time was 239 seconds and the mean distance swum was 26.2m. The longest swim time recorded was 395 seconds and the farthest distance swum was 47.4m. Average swimming speed was 11.5 cm/second.

Best and Hart (1) found that average swimming time for three species of plains-adapted pocket gophers (Geomys and Pappogeomys) was 362 seconds; their swimming speed was an average of 17 cm/second. A similar sized trough was used, showing a significant increase of an average of 123 seconds more than Thomomys individuals; swimming speed difference was markedly slower in the northern gopher.

Northern pocket gopher burrows are generally encountered in friable soils in inclined rather than low elevations. Flooding of low lying areas is an obvious threat and the drainage advantages of higher elevations are apparent. The not uncommon physical approximation of these gopher burrows to bodies of water, though usually in higher ground, suggests that swimming ability may be a factor though perhaps minor, in survival and self preservation during high water, if not in dispersal.

In typical Thomomys habitat, from gentle hills to abrupt mountain ranges, there is probably less opportunity and/or necessity to swim than is present for the plains-adapted pocket gopher. The latter occupies the relatively flat Great Plains, where flooding possibilities are ever present.

## Summary of pocket gopher swimming performance

	Swimming Duration (Sec)	Swimming Speed (cm/sec)	Distance Swum (m.)
Males	4(259; 193.63) <sup>a</sup>	1(13.1; _____)	4(29; 15.39)
Females	7(240; 82.46)	6(11.2; 3.0)	7(31; 12.64)

<sup>a</sup>Sample size (mean; standard deviation).

1. Best, T. L. and Hart, E. B. (1976). Texas J. Sci., 27(3): 361-366.

HABITAT PREFERENCE OF THE WHITETAILED DEER (ODOCOLIEUS VIRGINIANUS)  
 IN BROWN AND SPINK COUNTY SOUTH DAKOTA

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The purpose of this study was to examine any possible correlations between the amount, distance to, or number of land use types to the number of whitetailed deer (Odocoileus virginianus) sighted in a given area. The deer were spotlighted by two researchers using a specially equipped vehicle with two 500,000 candlepower lights. Nine individual routes were driven a minimum of four times each June in 1975, 1976, and 1977. A total of 1096 deer were observed. Linear regression analysis was conducted on a Cyber 74-28 CDC computer. No correlations between the number of deer per sighting and our habitat variables were noted. Areas with small grain showed a higher percent occurrence of deer than did other land-use types.

## DENSITY AND REFRACTIVE INDEX STUDIES OF NITROGEN FERTILIZER SOLUTIONS (1)

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Nitrogen fertilizer solutions composed of urea and ammonium nitrate are prepared and sold commercially under many different trade names. The common label is "UAN" solution. Quality control analysis of the manufactured product is carried out by drawing off a sample from the continuous flow pipeline. This sample is analyzed by titration with standard acid and represents a labor intensive procedure. A method to determine solution composition using triangular graphs and the two physical properties, refractive index and density, has been suggested by Sloan and Veales.(2) After the examination of refractive index and density of over sixty different concentrations of "UAN" solution, our results indicate that this technique is only accurate over a very narrow range of concentrations.

Table 1  
Typical Set of Refractive Index and Densities  
of Urea Ammonium Nitrate Solutions

% Urea	% Ammonium Nitrate	Refractive Index	Density Studies
29	40	1.43828	1.2825
29	41	1.43949	1.2800
30	38	1.43668	1.2862
30	39	1.43848	1.2807
30	40	1.43959	1.2781
30	41	1.43758	1.2694

The triangular graphing method of the three component system uses percent water, percent urea and percent ammonium nitrate, each as a side of the equilateral triangle. Refractive index and density lines are obtained by both observed data and by geometrical methods. Intersection of the refractive index and density lines of one solution gives the percent composition of that solution. This method is only accurate over a very narrow range, and in certain ranges can give results which are in error by as much as ten percent. The reason for this error can be seen from plots of refractive index and density versus

percent water. The former is linear but the latter shows no semblance of a straight line. An alternate method using a plot of refractive index versus percent nitrogen could be useful to determine the composition of solutions over the concentration range examined which was 29 to 38 percent urea and 38 to 42 percent ammonium nitrate.

1. Advisors: Worman, J.J. and Pearce, M. South Dakota State University and Homan, J., Terra Chemicals International, Sioux City, Iowa.
2. Sloan, D.M. and Veales, R.W. (1977) Journal of the AOAC, 60, 876.

A SYNTHESIS OF 2,7-BIPHENYLENE DIAMINE  
AND ITS USE AS A CROSSLINKING AGENT IN A POLYMER

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2,7-biphenylene diamine was synthesized by a Hofmann Rearrangement of 2,7-biphenylene dicarboxamide. The amine was incorporated in a 5% composition into a polyamide. The polymer was thermally cured using the amine as a crosslinking agent.

## PASSIVE SOLAR ENERGY USE: THE TROMBE WALL

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Use of the sun's energy in space heating is achieved by employing active and passive systems. Active systems contain water or air solar collectors, plumbing rock or water heat storage, heat exchangers, and fans, etc. Passive systems are more simple in utilizing the building design, components, and natural surroundings to maximize use of the sun's energy. Passive systems are less costly and require less hardware than active solar heating systems. Passive systems can be easily incorporated into new construction. Existing architecture can be retrofitted by changing or adding on components; and by effective use of vegetation. One of the earlier and still very innovative passive systems is the Trombe Wall.

The Trombe Wall was designed by Felix Trombe, Director of the Centre National de la Recherche Scientifique in Odeillo, France in the 1950's. The Trombe House, built in 1967, and designed by Trombe and the architect, Jaques Michel; utilized a double-glazed concrete wall approximately two feet thick to absorb sunlight. In addition, the wall stored thermal energy and released it by radiation and convection. Evenly spaced vents at the top and bottom of the wall distributed heat by natural convection from the heated exterior face of the wall.<sup>1</sup>

This paper will present an overview of the Trombe Thermal Wall and its possible use in retrofitting existing dwellings.

By way of summary, passive systems are less costly and require less hardware than active solar heating systems. The Trombe Wall is a very viable and innovative passive system.

1. Mazria, E. (1979) The Passive Solar Energy Book, pp. 44-45. Rodale Press, Emmaus, P.A.

## EFFECTS OF DIETARY FIBER ON CHICKS

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Two separate experiments were performed to determine growth and biochemical effects of dietary fiber on chicks. In one experiment, Columbian-New Hampshire Red cross chicks were fed diets that contained 12, 15, 18, 21, or 24 percent protein with 2.5 or 12.5 percent fiber. Results pointed to the possibility that fiber stimulates increased food and caloric intake. Food efficiency was increased by high levels of fiber but decreased at high protein levels. The data suggest that cellulose had a beneficial effect on protein utilization in low or deficient protein diets but was detrimental to protein utilization at high levels of dietary protein. Growth rate (gain) was not solely dependent on dietary protein levels but also upon fiber.

In a second experiment, Columbian-New Hampshire Red chicks were used to evaluate the effects of pectin, barley extract, and microcrystalline cellulose on serum cholesterol. A hypercholesteremic diet was formed with the inclusion of whole egg powder. Although differences were not significantly different, barley extract (15%) appeared to be the most effective in lowering diet induced hypercholesteremia. Pectin (3%) was somewhat less effective and cellulose (15%) was least effective.

## QUANTITATIVE ANALYSIS OF ZOOPLANKTON IN LAKE YANKTON

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Zooplankton make up the very important animal portion of the planktonic community in lakes. They are the chief consumers of the phytoplanktonic producers and are the chief food source for young fish. Zooplankton communities are influenced by physiochemical factors of water, especially temperature, oxygen content, light, and currents. These factors can cause stratification among the zooplankton community (1). Density of zooplankton, especially the rotifer component, changes over time, peaking in late fall, then falling off and hitting a low in late summer (2).

This study examines variations in the quantitative populations of zooplankton in opposite ends of the lake, with a focus on the vertical distribution of rotifers and rotifer density over time.

Weekly samples were taken from Sept. 9 to Nov. 10, 1978 at sites on opposite ends of the small shallow lake using a Van Dorn water sampler at surface, 1 meter, and 2 meters. The water temperature was then recorded. All samples were reduced to a 1 ml volume using a 30-40um silk net to collect the zooplankton. The net was rinsed well with 10 ml of tap water, the rinse collected in a vial, and the sample preserved with Lugol's iodine. Samples were counted using a Ward's circular zooplankton counting chamber under 36X magnification of a stereoscopic microscope.

Cyclopoid copepods, calanoid copepods, copepod nauplii, cladocera, and rotifers were counted and their numbers recorded. The populations of zooplankton seemed to be randomly distributed from one end of the lake to the other. The population of rotifers appeared to be randomly distributed from the surface down to two meters. There was a definite increase in the density of rotifers in late fall as compared to early fall. There was also a definite increase in copepod nauplii and a decrease in adult copepods as the season progressed.

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2. Benson, N.G. and Cowell, B.C. (1968) The Environment and Plankton Density in Missouri River Reservoirs.

THE POTENTIAL OF TRANSITION METAL  $\pi$ -ALLYL COMPLEXES  
IN ORGANIC SYNTHESIS\*\*Allan Tramp  
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The utility of transition metal  $\pi$ -allyl complexes as reagents in the synthesis of interesting organic compounds was investigated. In particular, the preparation of new  $\pi$ -geranyl complexes and their potential as synthons for stereospecific terpene synthesis was explored.

\*\*This research was done under the direction of Dr. W.E. Carroll at the University of Arkansas, Fayetteville, on an NSF-URP program in the summer of 1978.

ENZYME STUDIES OF INTRA- AND EXOCELLULAR  
 PROTEASES OF NEUROSPORA CRASSA\*\*

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Neurospora crassa strain 74A has been found to synthesize and release a neutral and an alkaline protease into its growth media under specific growth conditions. The regulation of the synthesis and release of these proteases has been studied in some detail. Induction and repression of the proteases synthesis was investigated under various chemical influences. Also investigated was the effect that these chemicals had on the proteolytic activity of these proteases.

Experiments were run using the chemicals in varying concentrations and also adding them at various stages of the synthesis cycle of the protease. Three main chemicals were investigated: thermolysin, ethylenediaminetetracetate (EDTA), and cycloheximide. The conclusions drawn were based on cell growth in mg of weight increase and protein assays using electrophoretic and spectrophotofluorometric data.

\*\*This study was done during the January 1979 Interim under the direction of Dr. L. Dudley Eirich at the Battelle Northwest Research Laboratory in Richland, Washington.  
 "By acceptance of this article, the publisher and/or recipient acknowledges the U.S. Government's right to retain a nonexclusive, royalty-free license in and to any copyright covering this paper. This research was supported by the Northwest College and University Association for Science (University of Washington) under Contract EY-76-C-06-2225 with the U.S. Department of Energy."

SOLUTION PROPERTIES OF AQUEOUS TETRAALKYLAMMONIUM SULFATE SOLUTIONS AT 25°C:  
 ON INVESTIGATION OF CONDUCTANCES OF TETRAETHYLAMMONIUM SULFATE

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The effect of salts upon the structure of water and the nature of electrolytic solutions were studied by measuring conductances, partial molar volumes, and viscosities of aqueous tetraalkylammonium sulfate solutions at 25.0°C. An attempt has been made to relate the results of these measurements to the structure effects of certain ions, principally the sulfate anion, on water and of the extent of ionic association or micelle type formation in aqueous tetraalkylammonium sulfate solutions at 25.0°C. Equivalent conductance, partial molar volume, and viscosity were selected for study since they can be measured quite accurately, and because these properties have been used in the past to elicit information concerning the questions we wished to investigate. Previous work at Augustana College has included measurements of viscosities and partial molar volumes for ammonium sulfate, tetramethylammonium sulfate, and tetraethylammonium sulfate solutions from 0.1 molar concentration to near saturation at 25.0°C. In addition conductances have been measured for ammonium sulfate and tetramethylammonium sulfate from 0.00002 molar concentration to near saturation at 25.0°C. In order to complete these investigations, work is presently underway to extend the conductance measurements to include tetraethylammonium sulfate. An alternative preparative method for tetraethylammonium sulfate has been developed. An older, adiabatic, heat capacity solution calorimeter acquired from Iowa State University was repaired and modified. This instrument will be used to study heats of solution, heats of dilution, and heats of reaction of tetraalkylammonium salts.

Faculty Advisor: Wayne M. Gildseth

SURFACE GEOLOGY FOR LAND USE PLANNING  
IN THE MINOT, NORTH DAKOTA AREA

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The Minot area includes about 330 square kilometers along both sides of the Souris River in north-central North Dakota. The area can be divided into the flat Souris River floodplain, the steeply sloping sides of the Souris and Des Lacs river valleys and larger tributaries, and the gently undulating uplands dissected by small streams. Surficial geologic units include Tertiary sandstone and shale, glacial till, glacial ice contact fluvial deposits, and both Pleistocene and Holocene fluvial and eolian deposits.

Expansion of the Minot metropolitan area has resulted in conflict between urban development and planned resource management. A study was therefore undertaken to evaluate the surface and subsurface geology as it relates to mineral resources, water supplies, waste disposal sites, and flooding. The result was a detailed geologic map and a series of interpretive suitability maps intended to aid in land use planning.

Moderate amounts of high-quality sand and gravel adjacent to and within the Souris River valley constitute the major mineral resource in the area. These are largely glacial meltwater deposits. Proper land use planning should restrict development on such deposits until maximum use is made of this resource. The area southeast of Minot between US Hwy. 2 and the Souris River valley, particularly, should be zoned to prohibit residential development.

The second major resource is water. Water supply has been a major problem for Minot in the past (1). Water is presently available from the Souris River and from groundwater aquifers. These two sources provide Minot with an adequate supply for the present, but major aquifers have already been developed and the discovery of significant new groundwater supplies in the Minot area is unlikely. Therefore it is important that all possible steps be taken to protect existing water supplies from potential pollution and misuse.

Waste disposal is also an important facet of land use planning. Sanitary landfill and sewage lagoon facilities should be located in upland areas away from both the Souris River and the major aquifers to minimize possible pollution of water supplies. The present practice of locating sanitary landfill sites within coulees tributary to the Souris River is not advisable due to the potential for surface and groundwater pollution and the high erosion potential.

Finally, flooding has been and continues to be a problem in the Minot area (2). Major floods in recent past have caused significant damage to homes and businesses. The potential for future flooding and the uncertain future of the Burlington Dam project make continued development of the floodplain in Minot inadvisable.

The inclusion and consideration of detailed geologic information in the land use planning decision-making process is important if effective use is to be made of all available natural resources. Specifically, the City of Minot, N.D. must include the results of such studies in its expansion planning if costly and environmentally unsafe alternatives are to be avoided.

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2. Anderson, D.B. and Schwob, H.H. (1970) USGS Open File Report 70-7.

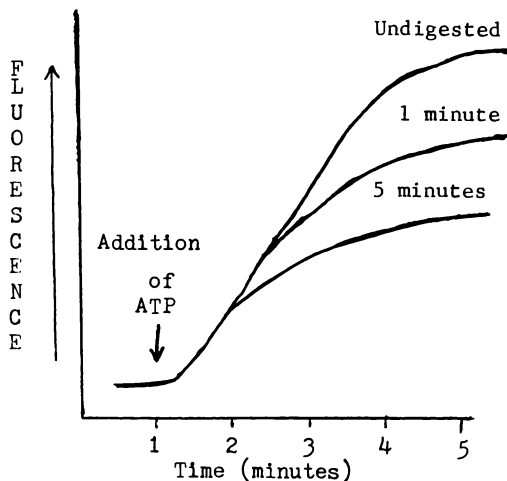
CALCIUM SPECIFIC ATP-ASE PUMP  
OF THE SARCOPLASMIC RETICULUM

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There is considerable evidence linking membrane proteins with active and passive transport of ions across membranes. The accumulation of calcium within the sarcoplasmic reticulum membrane system of skeletal muscle is facilitated by a  $\text{Ca}^{2+}$  specific ATP-ase pumping enzyme that spans the membrane. In the resting state of the muscle the enzyme is actively pumping in calcium and thus keeping the concentration of  $\text{Ca}^{2+}$  in the sarcoplasm very low. Upon stimulation the membrane releases  $\text{Ca}^{2+}$  and contraction takes place.

The high degree of specialization of the sarcoplasmic reticulum is reflected by its unusual simplicity; the membrane contains relatively few proteins. The major protein, accounting for as much as 60% of the total protein content, is the 106,000 dalton  $\text{Ca}^{2+}$  specific,  $\text{Mg}^{+}$  dependent, ATP-ase.<sup>1</sup> Thus the sarcoplasmic reticulum provides an excellent tool for the investigator in the study of membrane-protein interactions.

The major thrust of our investigation involved the isolation of sarcoplasmic reticulum vesicles from rat skeletal muscle and monitoring the  $\text{Ca}^{2+}$  transport *in vitro* utilizing a fluorescence technique described by Caswell.<sup>2</sup> The technique relies upon the fact that the antibiotic chlorotetracycline emits intense fluorescence when bound to  $\text{Ca}^{2+}$  in a hydrophobic membrane environment. Upon



addition of ATP, a properly prepared vesicle solution would show a marked increase in fluorescence due to the active pumping of  $\text{Ca}^{2+}$  into the vesicle by the ATP-ase enzyme. The active influx of  $\text{Ca}^{2+}$  in the intact vesicles was compared to the influx in similarly prepared vesicles that had undergone a series of proteolytic digestions. Our molecular weight studies, using polyacrylamide gel electrophoresis, showed that the 106,000 dalton protein had been fragmented into 55,000 and 45,000 dalton peptides. After five minutes of digestion the peptides were further broken down into smaller units. However, the fluorescence studies show that even after five minutes of digestion there is still considerable "pumping" activity. This indicates that even though the enzyme is not totally intact, it is still capable of limited calcium transport.

\*\*All laboratory work was done at Washington State University under a NSF Undergraduate Research Grant.

<sup>1</sup>Martonosi, A.N. and Halpin, R.A., (1971), Arch. Biochem. Biophys. 144, 66.

<sup>2</sup>Shamoo, A.E. and Goldstein, D.A., (1977), Arch. Biochem. Biophys. 169, 35.



EFFECT OF ELEVATED SELENIUM DIETS ON RAT LIVER PEROXISOMES<sup>1</sup>

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Liver cells contain distinct microbodies called peroxisomes. Some of the enzymes found in liver peroxisomes are catalase, urate oxidase, D-amino acid oxidase, glycolate oxidase, isocitrate dehydrogenase and enzymes for oxidation of lipids. Peroxisomes are enclosed by a single limiting membrane. The exact function of the peroxisomes is not known. The purpose of this study was to determine the stability of the peroxisomes during degeneration of the liver. Elevated levels of selenium are known to be toxic and to cause liver degeneration in young rats and so selenium was used in this study. Since catalase comprises about 40% of the protein in peroxisomes and is easy to assay, it was used as a marker for the isolation of the peroxisomes.

Female Sprague Dawley rats weighing about 50 gms. were fed rabbit food pellets containing oxytetracycline for 12 days and then they were divided into three groups of 14 rats each. Group 1, the control group, was fed a corn base diet, and groups 2 and 3 were fed the same corn diet but with 7 ppm and 10 ppm Na<sub>2</sub>SeO<sub>3</sub> added respectively.<sup>2</sup> The rats fed selenium did not grow as rapidly as the control rats. At about 9 weeks of age the selenium content in the diet was increased to 10 ppm and 15 ppm for groups 2 and 3 respectively. The isolation of the peroxisomes was carried out by differential centrifugation. The rat livers were weighed and then placed in 8.5% (wt/vol) sucrose in 0.05M phosphate buffer pH 7.5 and diced. Homogenization of individual or pooled livers was done with a Potter-Elvehjem homogenizer with a loose fitting pestle. The homogenized liver was filtered through cheesecloth and then centrifuged at 270g for 5 min. The supernatant was then centrifuged at 23,500g for 4 min. to isolate the peroxisomes. Resuspension of pellets and dilution of supernatants to about 30 ml was done with cold 0.05 M phosphate buffer pH 7.0. Fractions were assayed by following the decrease in H<sub>2</sub>O<sub>2</sub> concentration at 240 mμ. About 5 μl of each fraction was placed in a 3 ml assay solution consisting of room temperature 0.05 M phosphate buffer pH 7, 0.1% Triton X-100 and 0.059 M H<sub>2</sub>O<sub>2</sub>. The results are summarized in Table I.

In Experiment 1 individual rat livers were analyzed while in Experiment 2 pooled livers were analyzed. Each value in Experiment 2 represents three pooled rat livers. The results suggest that there is probably no effect of elevated selenium levels on peroxisomes. However, the data is highly variable and no firm conclusion can be drawn yet from this preliminary data. Only in Experiment 2 could gross discoloration of the livers be observed in the rats fed 10 ppm selenium. In all other cases, livers looked normal although once in awhile a very slight streak of discoloration appeared in livers of rats on selenium diets.

Table I  
 Catalase Activity in Rat Liver

Experiment Number	Control		7 ppm Se		10 ppm Se	
	Total Activity (μmoles min <sup>-1</sup> gm <sup>-1</sup> )	Peroxisomal Activity (%)	Total Activity (μmoles min <sup>-1</sup> gm <sup>-1</sup> )	Peroxisomal Activity (%)	Total Activity (μmoles min <sup>-1</sup> gm <sup>-1</sup> )	Peroxisomal Activity (%)
1	48946	24.3	106,753	14.2	45768	21.4
	91813	11.7	49705	16.7	60712	17.3
	46669	26.6	54535	24.3	45305	17.2
2	28885	23.1	22411	27.8	18758	29.1
	31479	32.6	26613	31.2	29548	27.2

1. This research was supported in part by Faculty Research Funds from Northern State College.
2. The authors thank Dr. O. Olson and Dr. I. Palmer, South Dakota State University, for their help.

## ENERGY IN SUNFLOWER STALKS AND RATES OF CELLULASE ACTIVITY ON SUNFLOWER STALKS

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The quantity of fossil fuel in the world is limited. It is then important that an unlimited renewable source of energy be found. One renewable source of energy is biomass. Biomass can be used either by compressing the material and burning it or by its conversion to compounds that can be fermented to provide gaseous or liquid fuel. The purpose of this research was to determine the energy content of sunflower stalks and determine how much glucose can be produced from sunflower stalks.

Three kinds of sunflower stalks were used, these being a confectionary type grown in 1978 and oil types grown in the 1977 and 1978 crop years. The stalks were ground in a Warring blender. The ground stalk was then passed through either a 149 micron or a 595 micron screen. The material that passed through the screen was then compressed to form a pellet using a Parr pellet press. The energy content of the pellet was determined by the combustion of the pellet in an oxygen enriched atmosphere (about 25 atmospheres) using a Parr isothermal jacket bomb calorimeter. The results are given in Table I. The energy content of the stalks was found to be lower than those previously reported (1). The stalks used for this study were from field grown plants and were collected about one month before harvest in 1978 and in 1977 collected about 2 months after harvest. The energy content did vary some between crop years but not significantly. Likewise there was no significant differences found between oil type and confectionary type sunflowers.

Table I  
 Energy Content of Sunflower Stalk

Crop Year and Type of Stalk	Particle Size (Microns)	Mean Energy Content (calories/gram)
1978 Confectionary	149	3830
1978 oil <sup>1</sup>	595	3667
1977 oil <sup>2</sup>	595	3421

1) Interstate Brand 8944  
 2) Interstate Brand 903

The production of glucose from sunflower stalk was accomplished by incubating 5 mg cellulase (*Aspergillus Niger*, 1.1 units of activity/mg) with 40 mg of ground sunflower stalk at 50°C for 30 minutes. After 30 minutes, 3 ml of 50 mM 3,5 dinitrosalicylic acid was added to the assay tube and then it was placed in boiling water for five minutes. The sample was then filtered and read at 550 m $\mu$  in a Beckman DB using a zero time blank as reference. The amount of glucose produced was determined by comparing the absorbance to a standard curve. The standard curve was "spiked" with sunflower stalk material and enzyme because of the absorbance caused by these substances. The results showed that more glucose was produced from finely ground sunflower stalk than from larger sunflower particles.

In many cases the production of glucose is limited because the cellulose of the plant is ensheathed by a lignin which cannot be hydrolyzed by the cellulase. Therefore some way of destroying or at least "punching holes" in the lignin will lead to increased glucose production. One method of delignification is that described by N. Toyama and V. Ogawa (2). The delignification procedure consisted of mixing approximately 3.0 gms of ground sunflower stalk and 40 ml of 1% (wt/vol) NaOH and autoclaving at 52°C for 15 minutes. After autoclaving, the residue was filtered and washed to remove the NaOH and then the residue was dried in an oven. About 50% of the ground sunflower stalk (595 microns or smaller) was lost by this delignification procedure. Thus a significant portion of the original energy present in the sunflower stalk was lost. It appeared delignification did not increase glucose production when compared to ground sunflower stalk that had passed through a 595 micron screen.

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PENTOBARBITAL INHIBITION OF  $\text{Ca}^{++}$  UPTAKE INTO RAT BRAIN SYNAPTOSOMES

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It is generally held that the most likely site of action of the barbiturates is at synapses located in the central nervous system. Investigators agree that in mammalian brain the barbiturates inhibit excitatory synaptic transmission by a predominantly presynaptic mechanism. Evidence indicates this presynaptic effect results in a decreased neurotransmitter release giving rise to the observed sedative effect (1).

Since calcium ion is required for excitation-secretion coupling of these neurotransmitters, it was of interest to investigate whether acute and/or chronic administration of barbiturates alters calcium levels and/or calcium fluxes at the nerve ending.

Brain nerve endings (synaptosomes) were prepared from rat brain stem and cortex using a modification of the buoyant density centrifugation method reported by Cotman and Mathews (2). Calcium levels in synaptosomes from brain stem and cortex were determined by the atomic absorption spectrophotometry by comparison to standards.

Calcium uptake into synaptosomes from brain stem and cortex was studied using the calcium-45 radioisotope method of Lust and Robinson (3). Aliquots of the synaptosomal suspension were incubated at 30°C with equal volumes of  $\text{Ca}^{++}$  uptake media and terminated at various times from 30 sec to 5 min with a sodium-rich stopping solution. Radioactivity was determined by liquid scintillation counting. Calcium uptake is reported as  $\mu\text{mole calcium/g synaptosomal protein/min}$ . Calcium uptake was measured in control synaptosomes, synaptosomes pre-incubated with  $\mu\text{molar}$  concentrations of pentobarbital and synaptosomes from barbiturate tolerant rats. Protein determinations were carried out according to the method of Lowry et al. (4).

In brain stem, the acute treatment of 30 mg/kg i.p. pentobarbital causes no changes in the calcium content of synaptosomes. The same treatment has little effect on the calcium content of those synaptosomes isolated from cortex. On the other hand, chronic administration of pentobarbital to produce tolerance results in a two-fold increase in the calcium content of cortical synaptosomes. A small but significant increase was observed in brain stem.

Pre-incubation of synaptosomes, isolated from brain stem or cortex, with  $10^{-3}$  M,  $10^{-6}$  M or  $10^{-9}$  M phenobarbital showed that in brain stem the uptake of calcium was decreased. Pentobarbital concentrations of  $10^{-3}$  and  $10^{-6}$  M reduced calcium uptake by 33% and 21% respectively in brain stem, while  $10^{-9}$  M has no effect on calcium uptake. *In vitro* incubation of synaptosomes from cortex with pentobarbital had no effect on calcium uptake. Synaptosomes isolated from brain stem of barbiturate tolerant rats show a two-fold decrease in calcium uptake. The cortex shows no significant alterations in calcium uptake in barbiturate tolerant rats when compared to control animals.

The induction of the liver microsomal drug metabolizing system contributes to the tolerance seen with repeated administration of barbiturates (5). This phenomena, referred to as drug dispositional tolerance, may not account completely for the observed decrease in the effect of these drugs in tolerant animals. Recent demonstrations of a pharmacodynamic tolerance involving some form of central nervous system adaptation, not associated with drug metabolism, may play a role in tolerance to the barbiturates. The preliminary experiments described here may offer some data on a possible mechanism for this central adaptation. Calcium levels and uptake into synaptosomes isolated from brain homogenates, are significantly altered in pentobarbital tolerant animals. The brain stem appears to be especially sensitive. These data indicate that brain stem from tolerant animals has an increased storage capacity for calcium in the nerve terminal accompanied by a decreased ability for calcium uptake. It may be likely that pharmacodynamic tolerance to barbiturates is mediated through an alteration in calcium dynamics. Experiments are in progress to further investigate this hypothesis.

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## MILK PROGESTERONE PROFILES IN THE DAIRY COW

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The relationship between the concentration of progesterone in milk, method of extraction, and time of collection have been studied during the bovine estrous cycle. Beginning, composite, and stripping milk samples were taken at days 0, 5, 10, 14, 17, and 21 of the cycle. Samples were assayed, in duplicate, for progesterone content by radioimmunologic procedures. Two different extraction methods were compared for their ability to recover progesterone from the milk sample. Progesterone levels varied considerably between beginning milk and those samples collected from composite and stripping portions of the milking (table 1). The concentration of progesterone was higher for stripping milk during the early luteal phase of the cycle but the concentration of progesterone falls below that of the composite milk sample during the late luteal phase. Of the two extraction methods, it was observed that the short extraction method (extraction A) recovered a greater amount of progesterone than the centrifuge method (extraction B). Progesterone recovery rates during the cycle utilizing extraction method A were greater for all three collection times (beginning, composite, and stripping) when compared to extraction B. However, both extraction A and extraction B indicated a consistent rise in progesterone levels occurred during the first half of the cycle. Progesterone concentration reached maximal levels for both extraction methods at day 14. From day 17 to 21 an abrupt decline in progesterone concentration was detected by both extraction methods, thus indicating the end of the luteal phase of the bovine estrous cycle. These results would indicate either composite or stripping milk samples could be used for progesterone analysis to predict stage of the estrous cycle for the bovine female.

Table 1

Progesterone Concentration of Milk as Influenced by  
 Day of Estrous and Time of Collection

Day of Estrous Cycle <sup>a</sup>	Time of Collection <sup>b</sup>	Method A		Method B	
		$\bar{x}$	$\pm$ SEM	$\bar{x}$	$\pm$ SEM
0	B	0.29	$\pm$ .07	0.16	$\pm$ .11
	C	0.28	$\pm$ .08	0.32	$\pm$ .12
	S	0.25	$\pm$ .07	0.15	$\pm$ .07
5	B	0.95	$\pm$ .33	0.25	$\pm$ .09
	C	1.38	$\pm$ .59	0.38	$\pm$ .18
	S	1.83	$\pm$ .55	0.44	$\pm$ .19
10	B	1.19	$\pm$ .58	0.41	$\pm$ .08
	C	2.33	$\pm$ .84	0.85	$\pm$ .23
	S	2.39	$\pm$ .77	0.92	$\pm$ .24
14	B	2.52	$\pm$ .78	0.56	$\pm$ .11
	C	3.58	$\pm$ .91	1.11	$\pm$ .35
	S	3.18	$\pm$ .71	1.01	$\pm$ .32
17	B	2.18	$\pm$ 1.13	0.43	$\pm$ .08
	C	3.39	$\pm$ 1.25	0.84	$\pm$ .36
	S	3.10	$\pm$ 1.10	0.76	$\pm$ .31
21	B	0.22	$\pm$ .14	0.15	$\pm$ .12
	C	0.41	$\pm$ .18	0.22	$\pm$ .16
	S	0.85	$\pm$ .63	0.47	$\pm$ .39

<sup>a</sup>Each mean an average of five cows.

<sup>b</sup>Time B = beginning milk; C = composite; S = stripping.

## HORMONAL PATTERNS IN MGA IMPLANTED ANESTROUS EWES

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plasma progesterone and estradiol levels of 18 anestrous crossbred ewes were determined prior to insertion, time in situ and after removal of silastic implants to ascertain their effect on hormonal secretion rates. The implants were impregnated with 50 mg of Melengestrol Acetate (MGA) and subcutaneously administered for 12 or 18 days. Placebo implants were inserted in 6 crossbred ewes which served as a control group. Blood samples were collected by venipuncture immediately prior to implant insertion, 7 and 10 days following insertion, upon removal and every four hours thereafter for 5 consecutive days. Progesterone and estradiol content of these plasma samples were measured by radioimmunoassay. Ewes were exposed to rams twice daily beginning 24 hours following implant removal for a period of 25 days as a further measure of mating activity. The mean plasma progesterone concentration for all ewes prior to implant insertion was  $0.85 \text{ ng/ml} \pm 0.15$ . Ewes having non-treated implants in place had lower plasma progesterone levels ( $0.38 \text{ ng/ml} \pm 0.06$ ) when compared to ewes having treated implants in place for 12 days ( $0.72 \text{ ng/ml} \pm 0.09$ ) and for 18 days ( $0.54 \text{ ng/ml} \pm 0.06$ ). Mean plasma progesterone concentration for 5 days following implant removal in the non-treated group was  $0.57 \text{ ng/ml} \pm 0.09$ . Ewes carrying treated implants for 12 or 18 days had mean plasma progesterone concentrations of  $0.54 \text{ ng/ml} \pm 0.06$  and  $0.41 \text{ ng/ml} \pm 0.05$ , respectively, for 5 days following implant removal. Differences in plasma progesterone concentrations as related to time after implant removal are presented in table 1.

Table 1  
 Plasma Progesterone Levels Post-Implant Removal

Time after Removal (Days)	Plasma Progesterone Concentration (ng/ml $\pm$ SEM)		
	Non-treated Group	Implanted 12 Days	Implanted 18 Days
1	$0.54 \pm 0.22$	$0.59 \pm 0.14$	$0.54 \pm 0.13$
2	$0.59 \pm 0.25$	$0.57 \pm 0.17$	$0.44 \pm 0.12$
3	$0.50 \pm 0.17$	$0.56 \pm 0.17$	$0.34 \pm 0.13$
4	$0.53 \pm 0.20$	$0.59 \pm 0.17$	$0.43 \pm 0.17$
5	$0.68 \pm 0.24$	$0.38 \pm 0.12$	$0.33 \pm 0.11$

Plasma estradiol concentration for all ewes prior to implant insertion was  $8.37 \text{ pg/ml} \pm 2.43$ . Ewes which received dummy implants retained this concentration while the non-impregnated implants were in place ( $8.10 \text{ pg/ml} \pm 1.72$ ). Ewes with MGA implants for 12 or 18 days had plasma estradiol concentrations of  $5.40 \text{ pg/ml} \pm 2.09$  and  $5.60 \text{ pg/ml} \pm 3.67$ , respectively.

Following implant removal estradiol concentration for all ewes not exhibiting estrual activity when exposed to rams was  $2.50 \text{ pg/ml} \pm 0.90$ . One non-treated ewe exhibited estrual activity two days following implant removal and had a plasma estradiol concentration of  $13.47 \text{ pg/ml} \pm 1.93$  on that day. Seven ewes exhibited estrual activity 6 to 25 days following implant removal. Four of these were in the non-treated group indicating a possible stimulatory effect from the rams. These results would indicate MGA implants of 50 mg. were not totally effective in causing hormonal profile alterations necessary to stimulate estrual responses in noncycling females.

Study of Immunity by Mycobacterium bovis (BCG) Cell Wall Fractions in Mice

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In spite of its extensive use in cancer immunotherapy in tumor bearing animals, as well as in some type of leukemic patients, the way Mycobacterium bovis (BCG) cell walls act to increase the resistance of host is not well defined (1).

The objective of this study is to obtain preliminary information on the mechanism of Immunity by Mycobacterium bovis (BCG) cell wall fractions in mice. The parameters used to meet the study objectives were: a) variation in liver and spleen size b) variation in total white blood cell count and c) variation in differential white cell count due to immunization by certain fractions of BCG cell wall.

The study consisted of preparation of immunizing agents and the study of the immunity index in mice following immunization by specific immunizing agents. 1. Mycobacterium bovis (BCG) was grown in Dubos broth for a week at 37°C. The cells were centrifuged and harvested after washing three times with physiologic saline. The wet cells were disrupted with Ribi cell disruptor, centrifuged and washed. Cell walls were treated with neutral solvent. The extract contained free lipids while the residue contained cell wall skeleton (CWS) and later fractionated by saponification. LH20 gel column was used for separation of fractions and was purified by high performance liquid chromatograph, waters model 440 uv detector measuring absorbance at 254 nm, and a water model 660 solvent programmer. One of the fraction containing trihalose dimycolate (P<sub>3</sub>) was used in this study. 2. Seven groups of Sasco ICR mice, each containing seven mice were used. Each group of mice except the controls were injected subcutaneously with specific immunizing agent 1. Group I: 0.03 mg CWS in 0.1 ml saline/mouse. Group II: 0.03 mg CWS in 0.1 ml saline-oil mixture (1:1), Group III: 0.015 mg CWS and 0.015 mg P<sub>3</sub> in 0.1 ml saline-oil mixture (1:1), Group IV: 0.03 mg P<sub>3</sub> in 0.1 ml saline-oil mixture, Group V: 0.03 mg of P<sub>3</sub> in 0.1 ml saline. Group VI and VII were control animal groups receiving 0.1 ml oil and 0.1 ml saline-oil mixture respectively. Beginning eleventh day after immunization, two mice from each group were sacrificed at weekly intervals after a few drops of blood were taken from each tail. The blood was used for total and differential white cell count. Spleen and liver were removed from each mouse and weighed.

The effect of BCG cell wall fractions was observed by examining the blood sample from each group of animals for total and differential white cell counts. The results indicate that Group III mice showed maximum increase in the total number of white blood cells (WBC) by 10,640 and the next highest increase was by 5710 in Group II animals. Group II mice were immunized with CWS in oil, whereas Group III mice were immunized with CWS and P<sub>3</sub> in oil-saline mixture. Whenever P<sub>3</sub> in saline was used as an immunizing agent, the total white count was first increased and then decreased with time in contrast to P<sub>3</sub> in oil, where it increased gradually. Higher WBC count was observed in mice immunized with CWS in oil than CWS in saline. But when total WBC count from mice immunized with P<sub>3</sub> in oil was compared with WBC count from mice immunized with CWS in oil after fifth week of immunization, the total WBC count was found to be more than two fold. The WBC count was decreased in each group of mice after immunization and then gradually increased with time. The result from differential WBC count indicates that in Group III mice the number of monocyte was doubled within three weeks, where as lymphocyte count stayed constant with slight decrease during third week after immunization. Whenever oil was used with any BCG cell wall fractions as an immunizing agent lymphocyte count was first decreased and then increased to contrast to neutrophil count which was first increased and then decreased. Both lymphocyte and monocyte are related to delayed hypersensitivity. When the correlation between total WBC count and differential WBC count are made, the data definitely indicate that there is a relationship between increase total WBC and increase in lymphocyte and monocyte.

Experimental data shows that there were a great variation of spleen and liver size due to immunization with different cell wall fractions. The highest percent weight increase (300%) was observed in the spleen of mice immunized with P<sub>3</sub> in oil. Similarly highest percent weight increase (68.34%) was observed in liver of mice immunized with combination P<sub>3</sub> and CWS in oil-saline mixture. Immunization with CWS in oil did not change the liver size to a great extent contrary to spleen size. In this case spleen size was increased 131.25% within five weeks after immunization.

This preliminary study shows that increase of liver and spleen sizes is related to increase in total white blood cells, lymphocytes, and monocytes due to immunization with P<sub>3</sub> and CWS in saline-oil mixture. The index of immunity is considered to be lymphocyte and monocyte and very definitely related to cell mediated immunity. This supports the treatment of leukemic patients using combination of BCG CWS and P<sub>3</sub> in oil-saline mixture.

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Aging South Dakota Red Foxes by Analysis of Canine  
Cementum Annuli and Enamel Lines

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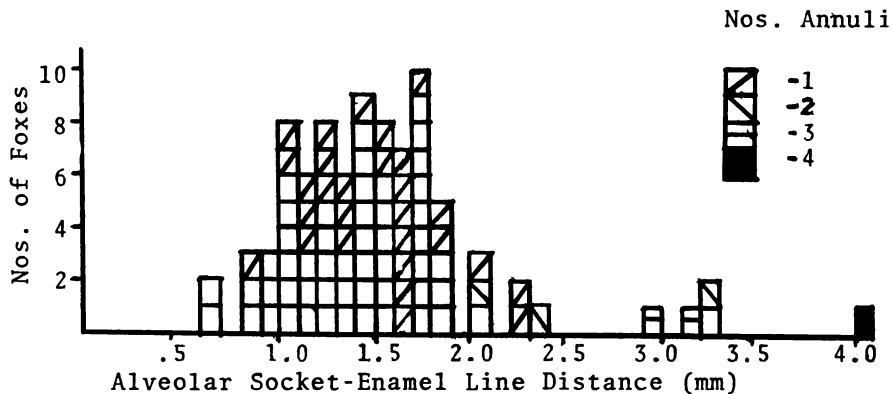
The objective of this continuing study was to assess dental aging characteristics of 86 representative northeastern South Dakota red foxes of unknown ages made available by a local fur buyer. Counts of dental cementum annulations and measurement of enamel lines were the primary criteria used. A combination of several preparation techniques of razor sectioned canines were utilized (1), (2). All upper right canines were examined for cementum annuli. Enamel line means were obtained by measuring upper canines. Results were compared to Allen's (1) study of North Dakota red foxes of known ages.

Inasmuch as Allen found 100 percent agreement between known-ages and cementum annuli ages in nearby North Dakota, and considering other correlative positive comparisons, it is reasonable to extrapolate that South Dakota red foxes of unknown ages can be accurately assigned ages also based on cementum annuli counts.

Allen found no adults with mean enamel line measurements of less than 2 mm. He found that 88 percent of known-age juveniles had enamel line measurements equal to or less than 2 mm. Results from the South Dakota red foxes indicated 50 to 52 (98%) foxes with no annulations had enamel line measurements less than 2 mm. Churcher (3), however, suggested the breakoff point as 1.0 mm between juveniles and adults. South Dakota foxes, using cementum annulations and skull ossification and other aging techniques, (4), as criteria, supported the North Dakota study. Churcher also suggested that age year classes could be assigned accurately from enamel line measurements. South Dakota foxes examined herein do not support Churcher's findings, as Allen also found.

As aging techniques become more accurate, larger samples can be analyzed to determine population composition of fur animals harvested. In turn, inferences can then be projected towards actual age classes present in local natural populations, and the ecological impact of artificial removal can be then projected.

Distribution of Enamel Line Measurements (mm)  
Correlative to Cementum Annuli



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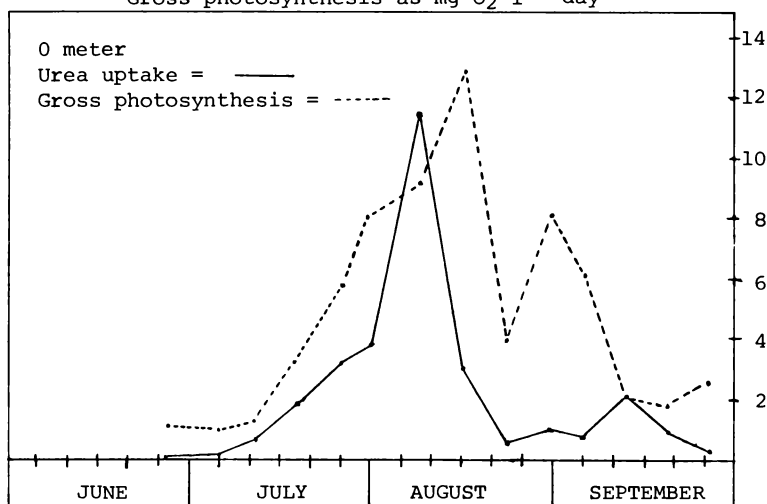
UREA UPTAKE AND RELEASE DURING MID-DAY HOURS ( 8AM - 3PM ) OF PHOTOSYNTHETIC  
ACTIVITY IN BREWER LAKE, ERIE, NORTH DAKOTA

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The uptake and release of urea as a nutrient by a predominately blue-green algal population in Brewer Lake, North Dakota, was measured during  $\approx 7$  hours ( $\bar{x} = 6.82 \pm .18$ ) of incubation in light and dark bottles, suspended at one meter intervals from the surface to 5 meters. Urea concentrations before and after incubation were determined using the urease method of McCarthy (1). During the period of maximum increase in photosynthetic activity (27 June to 8 Aug.), initial urea concentrations were zero. Immediately following the peak in the photosynthetic pulse, initial urea concentrations rose, and ranged from 1.0 to 5.3 microgram atoms urea nitrogen per liter ( $\mu\text{g at urea-N l}^{-1}$ ).

In order to obtain a measureable amount of urea present, a set of light and dark bottles were set-up and inoculated with  $10 \mu\text{g at urea-N l}^{-1}$ , and for comparison, a duplicate set was run simultaneously without urea added. In the surface (0 meter) bottles, after incubation was completed, during increasing photosynthetic activity (27 June - 8 Aug.), urea concentrations decreased in the light bottles containing the inoculated urea, and remained at zero in the light bottles with no urea added. In both sets, the dark bottles showed an expected increase in urea, probably from the decay of algae. Of 14 sampling dates spanning 91 days from 27 June to 26 September, there were 8 intervals where photosynthesis increased, with 7 (87.5%) of the intervals showing a concurrent increase in urea uptake. When photosynthesis declined on the remaining 5 intervals, urea uptake decreased simultaneously in 3 (60%) of the intervals. Thus, changes in urea uptake paralleled changes in photosynthetic activity in 10 (76.9%) of the 13 sampling intervals as seen in graph 1. During one of these measurement periods (15 Aug.), photosynthesis was at its peak, and therefore showed a much lower urea uptake than the previous periods of high photosynthesis. During the increasing pulse (27 June - 8 Aug.), urea uptake was significantly correlated,  $r = .972$ , ( $P < .01$ ,  $n=6$ ), with gross productivity (as milligrams oxygen per liter). During the decline of the pulse, 15 Aug. to 12 Sept., urea uptake versus gross productivity gave a correlation coefficient of  $r = .956$  ( $P < .01$ ,  $n=5$ ). Urea loss in light bottles of the inoculated set following the 7 hour incubation, also showed a significant correlation ( $r = -.761$ ,  $P < .01$ ,  $n=14$ ) with chlorophyll *a* with all sampling dates included. The present results suggest urea may be utilized by freshwater phytoplankton during high photosynthetic periods.

Graph 1  
Urea uptake as  $\mu\text{g at urea-N l}^{-1} \text{ hour}^{-1}$   
Gross photosynthesis as  $\text{mg O}_2 \text{ l}^{-1} \text{ day}^{-1}$



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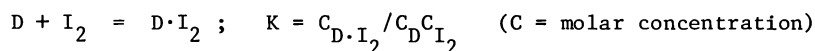


MOLECULAR COMPLEXES OF I<sub>2</sub> WITH ELECTRON DONORS

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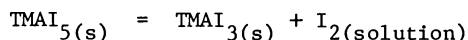
Elemental iodine forms complexes in solution with various electron donors (1) as shown below:



The complex is stabilized by electrons that move from the molecular orbitals of the donor into molecular orbitals closely associated with the I<sub>2</sub> (the electron acceptor). These complexes typically absorb in the visible and ultraviolet regions.

The donors studied were the aromatic compounds toluene, p-xylene, and mesitylene (1,3,5-trimethylbenzene). Solutions of iodine in heptane in the presence of these donors had the typical red-violet color and an isosbestic point near 519 nm.

The following equilibrium (2) was utilized to establish constant iodine activity heptane solutions:



where, TMAI<sub>5</sub> = tetramethylammonium penta-iodide  
TMAI<sub>3</sub> = tetramethylammonium tri-iodide

In the presence of such an equilibrium and a donor, any increased absorbance at the isosbestic point ( $\Delta A$ ) was attributed to the complexed iodine in the 1:1 complex.

Solid TMAI<sub>5</sub> was equilibrated in the inert solvent heptane, establishing a base absorbance ( $A^0$ ); similar solutions were prepared also containing donor having formal concentrations ( $F_D$ ) in the range of 0.2 to 1.0. The 1:1 association constants were evaluated from the slope of plots of  $\Delta A$  vs  $F_D$  using the relationship,  $\Delta A = A^0 K F_D$ . The association constants were measured at three temperatures; the reaction enthalpies were calculated using the van't Hoff relationship.

The results obtained are shown in the table below. The enthalpies are close to literature values; however, the equilibrium constants are somewhat different from those obtained by conventional spectral methods (1). The data is consistent with the prediction that methyl groups increase the basicity of the aromatic system.

Donor	K (M <sup>-1</sup> )				- $\Delta H$ (kcal/mole)
	14.5°C	25.0°C	30.0°C	36.0°C	
benzene(3)	-	0.36 ±.01	0.34 ±.01	0.32 ±.01	1.4 ±.2
toluene	0.64 ±.02	0.59 ±.03	-	0.52 ±.02	1.7 ±.2
p-xylene	0.90 ±.03	0.74 ±.02	-	0.66 ±.01	2.6 ±.3
mesitylene	1.27 ±.05	1.00 ±.05	-	0.88 ±.01	3.0 ±.4

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## ARTIFICIAL NESTING SITES AS A PRAIRIE FALCON MANAGEMENT TECHNIQUE

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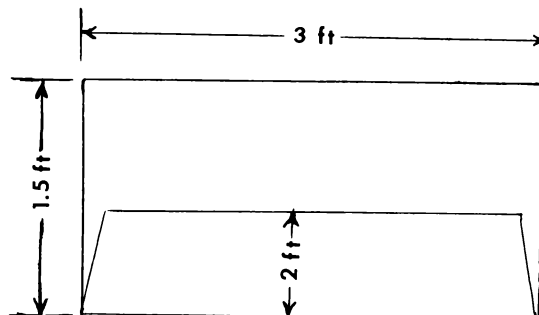
The prairie falcon is a crow sized raptor (bird of prey) of arid western North America. Historically, the breeding range of this species in North Dakota included much of the area west of the Missouri River; presently, breeding prairie falcons are limited to the North Dakota Badlands. In this region, prairie falcon eyries (nests) generally are located at least 10 meters above the ground in ledges or potholes of sheer faced cliffs. Strata in which prairie falcons nest are typically of sandstone or clay which are very common features of the topographically rugged Badlands. Eyrie sites in these cliffs are limited in number and highly susceptible to loss because of the continual effects of the wind and water erosion which has sculptured this area. In addition to nesting site limitations, increasing energy-related development may also pose a threat to nesting falcons. To alleviate these potential problems, we began to research the use of artificial eyries as a prairie falcon management technique in a 233 square kilometer Badlands study area in northern Dunn County.

Research was initiated during the 1977 breeding season during which we located three breeding pairs in our study area. From observations at those eyries and from past observations of nesting prairies in other parts of the Badlands we formulated criteria including height of cliff, exposure, and area below the cliff for selecting areas in which we could construct artificial eyries. After locating suitable cliffs we then dug twelve artificial eyries with average dimensions as given in the figure below.

Artificial eyries were completed before the 1978 breeding season. The use of the artificial sites by falcons was limited to one pair in 1978. This pair was successful up to the incubation stage of nesting but then a rodent got into the eyrie and destroyed the eggs. The nest was then abandoned and we were unable to relocate the pair. Although our success in attracting prairie falcons to artificial sites has been very limited so far, we still feel that artificial eyries may prove to be a worthy management tool. To test this we plan to modify some of our original eyrie diggings and construct new artificial eyries in the hope that prairie falcon utilization can be increased.

As an additional management goal we began, in 1978, to consider using artificial eyries, originally constructed for prairie falcons, as sites from which peregrine falcons could be reintroduced to North Dakota. One major problem that has plagued peregrine reintroductions elsewhere is great-horned owl depredation on the young releases. To initially test the possibility of other raptors depredating young peregrines released from our artificial eyries, feral pigeons were tethered into four of the artificial sites as surrogate peregrines. Golden eagles as well as great-horned owl inconsistently depredated on the tethered pigeons. Thus the potential for depredation on young peregrines at these sites does exist. We feel that the risk of raptor predation on reintroduction falcons needs further research and that means for thwarting the problem should be devised.

One novel solution might be to use the phenomenon of taste aversion. This technique has received most publicity with the coyote and sheep problem but it has also been shown to be successful in producing gustatory as well as visual aversions in the red-tailed hawk. We hope to investigate the potential of taste aversions in great-horned owls during 1979, in addition to monitoring the use of artificial eyries by prairie falcons.



## ETHER-HCl ASSOCIATION REACTIONS IN NONAQUEOUS SOLVENTS: THERMODYNAMICS AND SOLVENT EFFECTS

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The equilibrium constants and reaction enthalpies for the 1:1 association reaction of HCl with isopropyl ether in benzene and 1,2-dichloroethane were found using a solute isopiestic technique (1). The required experimental measurements were obtained by equilibrating a concentrated aqueous HCl solution (12 F) with a nonaqueous ether solution of known ether formality ( $f_E$ ) and then determining the increment ( $\Delta f_{HCl}$ ) by which the formality of the HCl in the organic phase exceeded the formal solubility of the HCl in the pure solvent ( $f_{HCl}^0$ ). The data were plotted using the relationship,

$$\Delta f_{HCl} = K_{11} f_{HCl}^0 (f_E - \Delta f_{HCl})$$

where  $K_{11}$ , the 1:1 ether-HCl association constant for the reaction, was found by its relation to the slope of the linear graph. Reaction enthalpies were obtained from the temperature dependence of the equilibrium constants using the van't Hoff relationship. Results are summarized in the following table:

Isopropyl Ether		$K_{11} (M^{-1})$				$-\Delta H^0 (kcal)$	$f_{HCl}^0$	Solvent Dielectric Constant
Solvent	5°C	15°C	25°C	35°C				
C1CH <sub>2</sub> CH <sub>2</sub> Cl	-	3.3 $\pm$ 0.1	2.4 $\pm$ 0.1	1.9 $\pm$ 0.1	5.1 $\pm$ 0.3	0.102	10.36	
CH <sub>2</sub> Cl <sub>2</sub> (2)	-	4.2 $\pm$ 0.2	3.4 $\pm$ 0.2	3.1 $\pm$ 0.1	4.9 $\pm$ 0.2	0.090	9.08	
CHCl <sub>3</sub> (2)	-	3.7 $\pm$ 0.4	2.6 $\pm$ 0.4	2.0 $\pm$ 0.1	5.5 $\pm$ 0.1	0.055	4.81	
C <sub>6</sub> H <sub>6</sub>	4.6 $\pm$ 0.3	3.7 $\pm$ 0.1	2.9 $\pm$ 0.1	-	3.8 $\pm$ 0.1	0.094	2.27	
CCl <sub>4</sub> (3)	-	5.7 $\pm$ 0.1	4.6 $\pm$ 0.1	3.1 $\pm$ 0.1	5.7 $\pm$ 0.4	0.037	2.23	
C <sub>6</sub> H <sub>12</sub> (2)	-	8.2 $\pm$ 0.1	5.6 $\pm$ 0.1	4.3 $\pm$ 0.1	5.6 $\pm$ 0.1	0.027	2.02	
Anisole (methoxybenzene)								
CCl <sub>4</sub> (3)	-	0.65 $\pm$ .05	0.57 $\pm$ .05	0.54 $\pm$ .01	1.5 $\pm$ 0.6			

The generalizations that follow from the data are that the association constant is inversely and the solubility directly related to the dielectric constant of the solvent. Also the association constant is inversely related to the solubility. There are, however, exceptions noted. For example, in CHCl<sub>3</sub>, the  $K_{11}$  predicted on the basis of dielectric constant and solubility would be between higher and lower values; however,  $K_{11}$  is relatively small because the "acidic" chloroform proton competes with the HCl for the ether basic site. In benzene,  $K_{11}$  is lower than might be predicted by the dielectric constant because the HCl forms a weak complex with the pi cloud of the benzene as shown by the relatively high HCl solubility in benzene. Thus, benzene competes with the ether for the HCl. The significantly lower enthalpy for anisole is attributed to a decrease in oxygen basicity due to the electron withdrawing effect of the benzene ring.

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WINTER ECOLOGY OF FIVE PRAIRIE AQUATIC  
ECOSYSTEMS

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Aquatic ecosystems in the Great Plains region are often stressed by very harsh environmental conditions during the winter. Depending on the ice depth, snow cover on the ice, and water levels prior to and during ice-cover, these aquatic systems can experience highly unstable conditions through the winter period, which vary through the winter and from year to year. Unfortunately, in many different types of aquatic ecosystems throughout the region, winter investigations of limnological conditions have been rare. This study was initiated by students in a winter aquatic ecology class at Augustana, to compare and contrast various physical, chemical, and biological parameters in five different types of aquatic ecosystems in southeastern South Dakota, during the winter period.

The five different aquatic ecosystems investigated during this project included: 1) Grass Lake, a shallow prairie lake; 2) Marindahl Lake, a relatively deep prairie impoundment; 3) Clay Creek, a spring-fed, ice-free outlet stream from Marindahl Lake; 4) Lewis and Clark Reservoir, an ice covered flowthrough Missouri River Impoundment; and 5) Gavins Point Dam tailwaters, an ice-free large river system just below Lewis and Clark Reservoir. All samples were obtained during one sampling trip to each system, in January, 1979. Samples were collected through depth profiles at several locations for each system.

There was little reverse temperature stratification in the three ice-covered systems, with temperatures remaining between 0 and 2.5 C from surface to bottom. Spring-fed Clay Creek maintained a warm 12 C temperature despite air temperatures which hovered around -32 C. The tailwaters of Gavins Point Dam maintained ice-free temperatures around 0 C due to the rapid currents generated by the turbines. Oxygen levels were adequate at all sites and depths except at Grass Lake where anaerobic conditions persisted throughout the water column at all sampling locations. All other physical-chemical parameters exhibited little variation with depth, and between sampling locations at the five ecosystems. Grass Lake had relatively high levels of  $\text{NO}_3\text{-N}$  ( $0.43 \pm 0.16$  mg/l), very high total alkalinity ( $189 \pm 15$  mg/l) relative to the other systems, and high total suspended solids loading ( $27.2 \pm 14$  mg/l) reflected by low light transparency (secchi depth 0.35 m). Marindahl Lake is located in a limestone drainage basin and was characteristically acidic (pH range 3.0-6.1), with unexpectedly low total alkalinity ( $52 \pm 12$  mg/l), moderate  $\text{NO}_3\text{-N}$  ( $0.24 \pm 0.18$  mg/l), and low suspended loading ( $8.99 \pm 9.69$  mg/l) reflected by secchi depths of 3.0-5.0 m. Clay Creek was characterized by low nutrients ( $\text{NO}_3\text{-N}$ , 0.005 mg/l), relatively high suspended loading (22.5 mg/l), and somewhat acidic conditions (pH, 6.0). The water quality of the two Missouri River systems was expectedly similar, despite ice-cover conditions. At these two large river systems, alkalinities were moderate (55-70 mg/l), pH was slightly alkaline (8.1-8.3),  $\text{NO}_3\text{-N}$  was relatively high (0.32-0.42 mg/l), and the total suspended load was expectedly very low (2.5-2.7 mg/l with secchi depths of 2.5-4.0 m).

Densities and diversities of zooplankton communities were very low in Grass Lake, Clay Creek, and Gavins Point Dam tailwaters. Diversity was also low at Marindahl Lake, but the density of zooplankton was somewhat higher. Lewis and Clark Reservoir also supported moderate densities of zooplankton, but had the lowest diversity of all five ecosystems. Cyclopoid copepods and cladocerans were the dominant taxa, found in about equal numbers, at four of the five ecosystems. In Lewis and Clark Reservoir, the copepods were conspicuously rare and only cladocerans were common.

Phytoplankton densities were moderate in the three lentic ecosystems, and very low in the two lotic systems. Diatoms and single-celled blue greens were numerically dominant in the three lentic ecosystems. Primary production, as estimated by *in situ* light-dark bottle incubations, and chlorophyll estimates, was very low to non-existent at all five locations. In the lentic systems, it would appear that primary production was light limited, due to heavy snow cover and thick ice (0.5-0.7 m).

Benthic habitat in four of the five ecosystems was an organic matter-silt, clay matrix, and it supported a typical oligochaete-chironomid community. Tailwaters of Gavins Point Dam had a scoured rock substrate which supported a diverse community dominated by caddis fly and chironomid larvae. Significant differences ( $p < 0.05$ ) in the benthos densities between sampling locations at the three lentic ecosystems indicated a clumped distribution of the common benthic inhabitants. Burrowing mayfly nymphs (*Hexagenia*) were common in Lewis and Clark Reservoir, but were absent in the other four ecosystems.

Multivariate step-wise linear regression provided some very interesting relationships between physical-chemical parameters and biological indices at the five systems. These relationships made apparent some possible causal mechanisms for community level differences among the five ecosystems.

## Sensitivity of a Test to Detect Bacteria in Bean Seed

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Bacterial blight caused by Xanthomonas phaseoli on dry edible beans is a serious problem in North Dakota. The disease causes yield loss and disqualifies seed beans from certification. Field inspections required for certification are not always definitive, and the standard lab test (2) for detecting blight is unreliable. A new lab test (1) involving enrichment culture and vacuum infiltration of seedlings is more rapid and reliable. Over 40 lots of seed have been tested and none have been found to be free of blight. Because clean seed was not available, the sensitivity of the new test could not be determined. This paper describes use of an antibiotic resistant bacterium to evaluate sensitivity of the test.

A sample of bean seed was surface sterilized and soaked in sterile tap water for 24 hours. Simultaneously a few (20-30) beans from the test lot were germinated in each of seven petri plates. The water used to soak the beans was divided into 7 flasks and pasteurized for 45 minutes at 65°C to kill plant pathogenic bacteria. The flasks were inoculated with 10 fold dilutions of a rifampin (3-[4-Methylpiperazinyliminomethyl] rifamycin SV) resistant mutant of Xanthomonas phaseoli (Xpr) isolated from diseased beans in a manner similar to that outlined by Weller and Saettler (3). One uninoculated flask served as a control. Serial dilutions from each of the flasks were plated on nutrient agar and colony counts were made after 36 hours to determine the number of viable bacteria. The lowest detectable number was  $6 \times 10^2$  bacteria/ml. Germinating seedlings were added to each flask and were vacuum infiltrated for 30-45 seconds. Infiltrated seedlings were then grown in enclosed plastic domes on vermiculite wetted with Maneb fungicide suspension. The domes were maintained under continuous fluorescent light. After 10 days, the lesioned leaves were macerated in sterile water and the liquid was plated on nutrient agar containing 50 ppm rifampin.

All bean seeds vacuum infiltrated with Xpr bacteria produced infected seedlings and rifampin resistant bacteria were isolated from the water-soaked lesions. A few seedlings from the control were also infected (natural inoculum) but no Xpr bacteria were detected.

We have therefore detected bacteria from the soak solution at approximately  $6 \times 10^2$  bacteria/ml and infer that the dome test for detecting seed borne bacteria is sensitive in this range.

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## A SAMPLING SURVEY OF BREEDING RAPTORS IN NORTH DAKOTA BADLANDS

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The North Dakota Badlands are a primitive area of rough topography that have recently become the focal point of increased exploration and drilling for oil reserves. To quantify the impact(s), if any, of future energy-related development on breeding raptors (birds of prey) in the Badlands it is necessary to have precise breeding population estimates that can be statistically compared with future estimates. Accordingly, we surveyed breeding raptors in a 233 square kilometer (km<sup>2</sup>) Badlands study area in northwest Dunn County in 1977 and 1978.

In 1977 we drew a simple random sample of nine 2.6 km<sup>2</sup> survey units (10% of the study area) and searched each unit for breeding raptors from 18 June through 12 July. Analysis of first year data indicated that survey efforts should have been initiated earlier in the breeding season and expanded to cover more area thus increasing sample size. We also felt that reducing the size of survey units and stratified sampling (1) would increase the precision of our raptor population estimates. Therefore, 1978 sampling units were 0.65 km<sup>2</sup> in size and each unit was placed into 1 of 2 strata on the basis of the amount of tree cover present. Tree cover on Stratum I units was <25% or >75% of the unit; tree cover on Stratum II units was >25% but <75% of the unit. A proportionally allocated stratified random sample of 117 units (33% of the study area) was drawn and surveyed from 1 May through 18 July. In both years, all sample units were surveyed for breeding raptors by extensive ground search for nests or territorial pairs. Survey results from each year were used to calculate breeding raptor population estimates at the 95% confidence level.

Table 1 summarizes data collected and population estimates  $\pm$  95% confidence intervals (C.I.) for each of the 11 species found nesting and for total breeding raptors in the study area. More nesting species were found in 1978 and the total population estimate for that year was higher than the 1977 estimate. The major factors causing the observed disagreement in yearly results probably were the differences in timing and extent of 1977 and 1978 surveys. The late start of 1977 surveys probably precluded finding nests of golden eagles and the 2 owl species because these species fledge prior to or during late June. Also, expanded survey efforts in 1978 increased the opportunities for finding nests of these species, harriers and red-tailed hawks. Riparian habitat was poorly represented in 1977 sample units and expanded coverage of that habitat in 1978 probably accounts for the large rise in kestrel numbers; most kestrel pairs were observed in riparian woods. The broad-winged hawk was the only species found in 1977 but not recorded in 1978; the study area is on the extreme edge of this hawk's range and the nest found in 1977 may be one of very few broad-winged nests that far west.

1978 sampling methods and survey efforts yielded gains in the precision of all population estimates (Table 1). Greatest precision was achieved by treating 1978 data as a simple random sample rather than as a stratified sample. This indicates that the observed gain in precision was primarily due to increased sample size and not a result of the stratification scheme. Therefore, for future general surveys in our study area, we recommend the use of simple random techniques with sample units of 0.65 km<sup>2</sup> and a total sample size  $\geq$ 117 units. The total estimate we calculated for 1978 should prove to be an adequate baseline for detecting changes in the breeding raptor population in our study area.

Table 1. Total Nests Found and Breeding Raptor Population Estimates, 1977 & 1978

Species	1977 Data; n=9			1978 Data; n=117		
	Nests Found	Pop. Est. $\pm$ 95% C.I.	Precision (%)	Nests Found	Pop. Est. $\pm$ 95% C.I.	Precision (%)
Kestrel	1	10 $\pm$ 22	219	15	45 $\pm$ 18	39
Merlin	1	10 $\pm$ 22	219	3	9 $\pm$ 8	92
Prairie falcon	1	10 $\pm$ 22	219	1	3 $\pm$ 5	160
Sharp-shinned hawk	3	30 $\pm$ 30	109	8	24 $\pm$ 13	55
Cooper's hawk	2	20 $\pm$ 29	145	4	12 $\pm$ 10	79
Red-tailed hawk	0	-- --	---	2	6 $\pm$ 7	113
Broad-winged hawk	1	10 $\pm$ 22	219	0	-- --	---
Golden eagle	0	-- --	---	1	3 $\pm$ 5	160
Harrier (marsh hawk)	0	-- --	---	2	6 $\pm$ 7	113
Great-horned owl	0	-- --	---	1	3 $\pm$ 5	160
Long-eared owl	0	-- --	---	4	12 $\pm$ 10	79
Total (simple random)	9	90 $\pm$ 57	63	41	124 $\pm$ 30	24
Total (stratified)		-- --	---	41	125 $\pm$ 31	25

## Membership of North Dakota Academy of Science

ADOMAITIS VYTAJITAS	41966301 16TH AVE NE	JAMESTOWN	ND58401
+ALBRECHT, LAURENCE J	1978802 STANFORD RD	GRAND FORKS	ND58201
ALFESSI JOSEPH	1962N GREAT PLAINS RES CENTE	MANDAN	ND58554
+ALLRICH, RODNEY D	1978ANIMAL SCIENCE NDSU	FARGO	ND58102
AVES, RICHARD	1978715 N 42ND ST	GRAND FORKS	ND58201
+ANDERSON EDWIN M	1962213 20TH AVE N	FARGO ND	58102
ANDERSON ORDEAN S	1972RURAL ROUTE 1	NEW PRAGUE MN	56071
ANTES, JAMES R	19793524 7TH AVE N	GRAND FORKS	ND58201
ANITA JR ALBERT E	1970MECH ENGR DEPT UND	GRAND FORKS	ND58202
ASCHBACHER PETER W	1958MET & RAD RES LAB NDSU	FARGO	ND58102
ASHWORTH ALLAN C	1974GEOLOGY DEPT	FARGO ND	58102
AUYONG THEODORE	1963MEDICAL SCHOOL UND	GRAND FORKS	ND58202
BALLINTINE, THOMAS	1978CHEMISTRY DEPT UND	GRAND FORKS	ND58202
BALTISBERGER RICHARD	1969CHEMISTRY DEPT UND	GRAND FORKS	ND58202
BANASIK ORVILLE J	1947CEPEAL TECH DEPT NDSU	FARGO	ND58102
BANOWITZ, GARY M	1978BACTERIOLOGY DEPT NDSU	FARGO	ND58102
BARBS, RICHARD	1977PROJECT RECLAYATION UND	GRAND FORKS	ND58202
BARKER WILLIAM T	1968BOTANY DEPT NDSU	FARGO	ND58102
BARNBY WILLIAM G	1957MECH ENGR DEPT UND	GRAND FORKS	ND58202
BARNHARD, MARY DIX	1975805 EAST ST	BOTTINEAU	ND58318
BARTON GEORGE	1972JAMESTOWN HIGH SCHOOL	JAMESTOWN	ND58401
BARTAK, DUANE	1977CHEMISTRY DEPT UND	GRAND FORKS	ND58202
BARTON, BILL	1978GF ENERGY RES CENTER	GRAND FORKS	ND58202
BECKERING WILLIS	1959U S BUREAU OF MINES UND	GRAND FORKS	ND58202
*BEHRINGER, MARJORIE	19698014-A PINEDALE COVE	AUSTIN	TX78578
BEIN, FREDERICK L	1978GEOGRAPHY DEPT UND	GRAND FORKS	ND58202
BEINSKEY CARL R	1958MINOT STATE COLLEGE	MINOT	ND58701
BEUKNAP, JOHN K	1979DEPT OF PHARM-UND	GRAND FORKS	ND58202
BEYSON, STEVEN A	1979ENFRGY TECH CENT BOX 8123	GRAND FORKS	ND58202
+BENZ, BRUCE	1978718 5TH AVE N	GRAND FORKS	ND58201
BENZ LEO C	19621407 N 23RD ST	BISMARCK ND	58501
BERKEY GORDON B	1970SCIENCE DIVISION MSC	MINOT	ND58701
BERRY, JAMES	1978U OF EVANSVILLE BOX 329	EVANSVILLE	IN47702
BERRYHILL DAVID L	1973BACTERIOLOGY DEPT NDSU	FARGO ND	58102
BERTILSON, HAL S	1978CONCORDIA COLLEGE	MOORHEAD	MN56560
BITZAN EDWARD F	1952U S BUREAU OF MINES UND	GRAND FORKS	ND58202
*BLISS, HAROLD N	1951MAYVILLE STATE COLLEGE	MAYVILLE	ND58257
BLUMELF JOHN P	1963N D GEOLOG SURVEY UND	GRAND FORKS	ND58202
*BOLFY, CHARLES	19671827 QUAIL ST #9	LAKEWOOD	CO80215
*BOLIN DONALD W	19461425 N UNIV DR	FARGO	ND58102
*BOLIN F M	19481505 6TH ST S	FARGO	ND58102
BOTTOMS, CHARLES L	1979940 BOX AVE	DICKINSON	ND58601
BOUDJOUK, PHILIP	1978CHEMISTRY DEPT NDSU	FARGO	ND58102
+BOURGOIS, JILL A	19781319 10TH AVE S	GRAND FORKS	ND58201
BRAMMER, J D	1978ZOOLOGY DEPT NDSU	FARGO	ND58102
+BRAND, MICHAEL	1976BOTANY DEPT NDSU	FARGO ND	58102
+BREKKE, DAVID	1979GEOL DEPT UND	GRAND FORKS	ND58202
BRIMEL MARY C	1969BACTERIOLOGY DEPT NDSU	FARGO	ND58102
BROPHY JOHN A	1960GEOLGY DEPT NDSU	FARGO	ND58102
BROSCHAT, MYRON D	1976215 S CASCADE	FERGUS FALLS	MN56537
BROWN RALPH C	1972GEOGRAPHY DEPT UND	GRAND FORKS	ND58202
BROWNLEVE STANLEY	1958PHYSIOLOGY DEPT UND	GRAND FORKS	ND58202
BRUSHMILLER, JOHN G	1978CHEMISTRY DEPT UND	GRAND FORKS	ND58202
BURTON, MICHAEL T	19771714 WHITESTONE	FARGO	ND58102
*CALLENBACH JOHN A	1954ENTOMOLOGY DEPT NDSU	FARGO ND	58102
CAMARA, MICHAEL	1977833 AILSIE APT 4A	KINGSVILLE	TX78363
CARMICHAEL, VIRGIL W	19791013 N ANDERSON ST	BISMARCK	ND58501
+CAMPBELL, RICHARD E	1978CHEMISTRY DEPT UND	GRAND FORKS	ND58202
CARLSON KENNETH	1960320 2ND AVE NW	MAYVILLE	ND58257
CARTER JACK F	1950AGRONOMY DEPT NDSU	FARGO	ND58102
CASSEL J FRANK	1954ZOOLOGY DEPT NDSU	FARGO	ND58102
CHERIAN, SEBASTIAN	1971BIOLOGY DEPT JAMESTOWN CO	JAMESTOWN	ND58401
CHRISTOFFERSON, LEE A	1952700 1ST AVE S	FARGO	ND58102
CLAFLIN, W JOSEPH	1974BOX 24 JAMESTOWN COLLEGE	JAMESTOWN	ND58401
CLAMREY, GARY K	1975BOTANY DEPT NDSU	FARGO ND	58102
CLAUSEN ERIC N	1968MINOT STATE COLLEGE	MINOT	ND58701
COLLINS CHARLES C	1962ELECT ENGR DEPT NDSU	FARGO	ND58102
COMITA GABRIEL W	1954ZOOLOGY DEPT NDSU	FARGO	ND58102
CONNELL MARVIN D	19722606 5TH AVE N	GRAND FORKS	ND58201
+CONWAY, CECILIA M	1977BIOLOGY DEPT UND	GRAND FORKS	ND58202
*COON ERNEST O	1923404 HAMLINE ST	GRAND FORKS	ND58201
CORNATZER WILLIAM E	1952BIOCHEMISTRY DEPT UND	GRAND FORKS	ND58202
COWARDIN LEWIS M	1967310 16TH AVE NE	JAMESTOWN	ND58401
CROSS, TIMOTHY	1977GEOLOGY DEPT UND	GRAND FORKS	ND58202
CVANCARA ALAN M	1963GEOLOGY DEPT UND	GRAND FORKS	ND58202
DANDY WILLIAM A	1975GEOGRAPHY DEPT UND	GRAND FORKS ND	58202
D'APOLLONIA BERT L	1968CEPEAL TECH DEPT NDSU	FARGO	ND58102
DAVIS DAVID G	1973MET & RAD RES LAB NDSU	FARGO ND	58102
+DAVIS, JOHN S	1978530 TULANA #11	GRAND FORKS	ND58201
*DEBOER, BENJAMIN	1952312 ALPHA	GRAND FORKS	ND58201
*DEBOER, KATHARINE	1963312 ALPHA	GRAND FORKS	ND58201
DINGA GUSTAV P	1961CONCORDIA COLLEGE	MOORHEAD	MN56560

DINUSSEN WILLIAM F	1950ANIMAL SCIENCE DEPT NDSU	FARGO	ND58102
DISEND DENNIS T	1963413 HILLCREST DR	MINOT ND	58701
DOERING EUGENE J	1066N GREAT PLAINS RES CENT	MANDAN	ND58554
DOGGER, JAMES R	1958RM 359 FED BLDG	HYATTSVILLE	MD20782
+DODD, STEVEN	1978BIOL DEPT UND	GRAND FORKS	ND58202
*DOOLBY, JOHN A	1950306 23RD AVE N	FARGO	ND58102
DOYLE, DARYL J	1978RUPAL ROUTE 2	VALLEY CITY	ND58072
+DRAVAGE, PHILIP	19792857 E OVERLOOK RD	CLEVELAND HEIGHTS	OH44118
+DRAVLAND, J ERIC	1977ANATOMY DEPT UND	GRAND FORKS	ND58202
DUFFEE JOHN A	1965MICROBIOLOGY DEPT UND	GRAND FORKS	ND58202
+DUH, SHOW-HONG	19798 BISON CT	FARGO	ND58102
DUSKY, JOAN	19771115 9TH AVE S	FARGO	ND58102
DUYSEN MURRAY F	1956ROTANY DEPT NDSU	FARGO ND	58102
+DZIADYK, BOHDAN	1978ROTANY DEPT NDSU	FARGO	ND58102
EBELTOST DAVID C	1964AGRONOMY DEPT NDSU	FARGO	ND58102
*EIDERSTROM, HELGE E	1953003 N 26TH ST	GRAND FORKS	ND58201
EDGEPLY CHARLES G	1955AIRY SCIENCE DEPT NDSU	FARGO	ND58102
EGINTON, CHARLES T	1979VETERAMS ADMIN CENTER	FARGO	ND58102
EL-ARINI, M OSAMA	19793201 PAR ST	FARGO	ND58102
ELLMAN, ROBERT	1957DNE ENGR RSCH/BOX 20 UND	GRAND FORKS	ND58202
ERICKSON, DUANE	1961ANIMAL SCIENCE NDSU	FARGO	ND58102
ERICKSON J MARK	1966ST LAWRENCE UNIV	CANTON	NY13617
EVANS GARY W	1975HUMAN NUTRITION LAB UND	GRAND FORKS ND	58202
EVANS HAROLD W	19612624 OLSON DR	GRAND FORKS	ND58201
FACEY VERA	1948BIOLOGY DEPT UND	GRAND FORKS	ND58202
FARNJM, BRUCE	1965DDE ENGR RSCH/BOX 20 UND	GRAND FORKS	ND58202
FARNJM, SYLVIA	1966CHEMISTRY DEPT UND	GRAND FORKS ND	58202
FEGLEY, MELVIN M	1961909 SANDERS	LARAMIE	WY82070
FELD, RICHARD W	19782208 1ST AVE N	GRAND FORKS	ND58201
FELICK, DUANE	19792010 DYKE AVE	GRAND FORKS	ND58201
FELL VERNON J	1964MET & RAD RESEA LAB NDSU	FARGO	ND58102
+FELLOWS, NILE	19782 SOUTH CT	MORRIS	MN56267
FILLIPI GORDON M	19721005 S 20TH ST	GND FORKS ND	58201
FISCHER ROBERT G	1964MICROBIOLOGY DEPT UND	GRAND FURKS	ND58202
FISH HAROLD E	197570X 338	WATFORD CITY ND	58854
FIVIZZANI, ALBERT J	1979BIOLOGY DEPT	GRAND FORKS	ND58202
FLEEKER JAMES R	1967BIOCHEMISTRY DEPT NDSU	FARGO	ND58102
*FLEETWOOD CHARLES W	1948CHEMISTRY DEPT NDSU	FARGO	ND58102
FLETCHER ALAN G	1970COLLEGE OF ENGR UND	GRAND FORKS	ND58202
+FLOSTIE, MICHAEL	1977PO BOX 28	ESKO	MN55733
FOSMIRE GARY J	1975HUMAN NUTRITION LAB UND	GRAND FORKS ND	58202
FOSSUM GUILFORD D	1957CIVIL ENGR DEPT UND	GRAND FORKS	ND58202
*FOWKES, WALTER W	1957422 W FARMER	INDEPENDENCE	MO64050
FOX, RICHARD A JR	1978ANTHRO/ARCHAE DEPT UND	GRAND FORKS	ND58202
FRAASE, RONALD G	1973PO BOX 223	BISMARCK	ND58501
*FRANK RICHARD E	19491020 ROYD DR	GRAND FORKS	ND58201
+FRECH, STEVEN J	1978179 D UNIV VILLAGE	FARGO	ND58102
FREEMAN MYRON L	1961DICKINSON STATE COLLEGE	DICKINSON	ND58601
FREEMAN PHILIP G	1958US BUR OF MINES UND	GRAND FORKS ND	58202
+FRETENBACH, DAVID J	1977ANATOMY DEPT UND	GRAND FORKS	ND58202
+FULTON, GARY W	19781123 17TH ST N	FARGO	ND58102
FUNKE B R	1966BACTERIOLOGY DEPT NDSU	FARGO	ND58102
+GABRIELSON, DAVID A	19791111 NORTHWESTERN DRIVE	GRAND FORKS	ND58201
GALITZ DONALD S	1974ROTANY DEPT NDSU	FARGO ND	58102
GANO, DAVID	1968MINOT STATE COLLEGE	MINOT	ND58701
GARDNER RUSSELL JP	1975700 1ST AVE SO	FARGO ND	58102
GARLASCO, CHRISTIANE	197817784 LUNNONHAUS DR #9	GOLDEN	CO80401
GARVEY ROY G	1967COLLEGE OF CHEM NDSU	FARGO	ND58102
GASSNER, GEORGE	1977MRP LAB UNIV STA NDSU	FARGO ND	58102
GILLES KENNETH A	1961VICE PRESIDENT AGR NDSU	FARGO	ND58102
GILMER, DAVID S	1978ROUTE #4 MEADOWLARK LN	JAMESTOWN	ND58401
GION, EUGENE R	19793017 MADISON AVE	FARGO	ND58102
GLASSER JAMES C	1973	REGENT ND	58650
GOETTLER, HANS J	1979MECH ENGR DEPT NDSU	FARGO	ND58105
GOETZ HAROLD	1968ROTANY DEPT NDSU	FARGO	ND58102
GRAN ARIENE H	1964710 PARK DR	FARGO ND	58102
GRENDA, JAMES C	1973PHYSICS DEPT ANGELO UNIV	SAN ANGELO	TX78901
GREENWOLD GERALD	1970ND GEOL SURVEY UND	GRAND FORKS ND	58202
GRINHOVD, GORDON H	1957US BUREAU OF MINES	GRAND FORKS	ND58202
GRUSE, PAUL A	1979SCHOOL OF PHARMACY NDSU	FARGO	ND58105
*GUSTAFSON BEN G	1939421 PRINCETON ST	GRAND FORKS ND	58201
GUTHRIE, PHYLLIS A	197810320 CARROLLWOOD LN #82	TAMPA	FL33618
+HALM, MICHAEL J	1978PHYSICS DEPT NDSU	FARGO	ND58102
HARPELL JAMES W	1970PHYSICS DEPT UND	GRAND FORKS	ND58202
+HARRIS, STEVE	19751521 SCOTT STREET	WILLIAMSPORT	PA17701
*HARWOOD THEODORE H	19549FD 2	ARLINGTON VT	02520
HASKINS ARTHUR G	1972MINOT ST COLLEGE	MINOT	ND58701
HASSETT, DAVID J	197920 FENTON AVE	GRAND FORKS	ND58201
HAJNZ EDGAR A	19511029 LINCOLN DR	GRAND FORKS	ND58201
HAZEN ARLO N G	1950COLLEGE OF AGR NDSU	FARGO	ND58102
HEINRICH, MICHAEL L	197880X 32	RHAME	ND58651
HELENBOLT, KENNETH S	19643563 LONGFELLOW RD	FARGO	ND58102
*HELGESON, E A	19362323 E WATER ST 37	TUCSON AZ	85719



+HOJCHERT, JOAN C	1976	ST THOMAS ND	58276
+HOFBERG, CRAIG	1978310 2ND AVENUE SOUTH	GRAND FORKS	ND58201
HILL LOREN W	1966COLLEGE OF CHEM NDSU	FARGO	ND58102
HINTJEWY, J WASYL S	1964COLLEGE OF CHEM NDSU	FARGO	ND58102
*HOFFNER, JEROME J	19492518 9TH AVE N	GRAND FORKS	ND58201
*HOFFMAN CHARLES A	1958MINOT STATE COLLEGE	MINOT ND	58701
+HOFFMAN, DENNIS	1979DEPT OF BIOCHEMISTRY-UND	GRAND FORKS	ND58202
+HOGANSON, JOHN W	1978GEOLOGY DEPT UND	GRAND FORKS	ND58202
HOLLAND FRANK D	1961GEOLOGY DEPT UND	GRAND FORKS	ND58202
HOLLWAY HARRY L	1973BIOLOGY DEPT UND	GRAND FORKS ND	58202
HOWELL, FRANCIS L	1970PHYSICS DEPT UND	GRAND FORKS	ND58202
HOWLETT LARRY D	1973913 SUBURBAN APTS	DEKALB	IL60115
HUNG, YUNG-TSE	1975CIV ENG DEPT UND	GRAND FORKS	ND58202
+HUNT, CURTISS	1978524 STATE ST	GRAND FORKS	ND58201
HUSAIN, SYED	1977PHYS & PHARM DEPT UND	GRAND FORKS	ND58202
+IVERSON, LOUIS	1978RDJ RECLAMATION UND	GRAND FORKS	ND58202
JACOB, ROBERT A	1974USDA HUMAN NUTR LAB UND	GRAND FORKS	ND58202
JACOBS FRANCIS A	1955BIOCHEMISTRY DEPT JND	GRAND FORKS	ND58202
JACOBSEN NEIL S	1967ZOOLOGY DEPT NDSU	FARGO	ND58102
JALAL SYED M	1965BIOLOGY DEPT UND	GRAND FORKS	ND58202
JENKINS, DENNIS R	1975493 MCMJLLIN DR	GRAND JUNCTION	CO81501
JENSEN, PAUL	1978909 6TH AVE NE	VALLEY CITY	ND58072
JOHANSEN ROBERT H	1955HORTICULTURE DEPT NDSU	FARGO	ND58102
JOHNSON, A WM	1961416 TERRACE DR	GRAND FORKS	ND58201
JOHNSON ARNOLD R	1966MINOT STATE COLLEGE	MINOT	ND58701
JOHNSON DOUGLAS H	1973BOX 1747	JAMESTOWN ND	58401
JOHNSON, GARY F	19721809 HARMON AVE	BISMARCK	ND58501
+JOHNSON, KIRK	1977	GLENBURN	ND58740
JOHNSON LESTER E	1969	BOTTINEAU	ND58318
JOHNSON, PHYLLIS E	1978HUMAN NUT LAB	GRAND FORKS	ND58202
JOHNSON ROBERT E	1969624 SINCLAIR	BOTTINEAU	ND58318
+JOHNSON, TARI	1978NDSU BACTERIOLOGY DEPT	FARGO	ND58102
JORDE, DENNIS G	1978BIOLOGY DEPT UND	GRAND FORKS	ND58202
JOSHI, MADHUSUDAN	1978ANATOMY DEPT UND	GRAND FORKS	ND58202
KANNOWSKI PAUL R	1960BIOLOGY DEPT UND	GRAND FORKS	ND58202
KARNER FRANK R	1963GEOLOGY DEPT UND	GRAND FORKS	ND58202
KEHEW, ALAN E	19791406 S 18TH	GRAND FORKS	ND58201
KEIFENHEIM, BRUCE	1978DOE ENGR RSCH/BOX 20 UND	GRAND FORKS	ND58202
KELLEHER JAMES J	1972MICROBIOLOGY DEPT UND	GRAND FORKS	ND58202
+KELLY, LYNN I	1978GEOLOGY DEPT UND	GRAND FORKS	ND58202
KEID, JUDY B	1978253 COLLEGE SW #3H	VALLEY CITY	ND58072
KIESLING RICHARD	1951PLANT PATH DEPT NDSU	FARGO	ND58102
KILLINGBECK, KEITH T	KS STATE U-BIOL DIV	MANHATTAN	KS66506
KISHORE, VIMAL	1978540 CARLTON CT #15	GRAND FORKS	ND58201
KLARINDE KENNETH	1971CHEMISTRY DEPT UND	GRAND FORKS	ND58202
KLEVAY LESLIE M	1973223 27TH AVE S	GRAND FORKS ND	58201
KLEVELAND DONALD F	1972HATTON HIGH SCHOOL	HATTON	ND58240
KNORLICH, JEROME	195880X 63	ELDRIDGE	ND58435
KNUDSON, CURTIS L	1973DOE ENGR RSCH/BOX 20 UND	GRAND FORKS	ND58202
*KOFENKER, WM F	19589079 SLIGO CRK PKWY 1812	SILVER SPRING MD	20901
*KOHANOWSKI, N	1949GEOLOGY DEPT UND	GRAND FORKS ND	58202
KOLLMAN, ALDEN L	1973PROJECT RECLAMATION-UND	GRAND FORKS	ND58202
KOLSTOF RALPH H	1962PSYCHOLOGY DEPT UND	GRAND FORKS	ND58202
+KODR, MICHAEL D	1979BIOLOGY DEPT UND	GRAND FORKS	ND58202
KOPONEN, MARK A	1975BOX 5225 NDSU	FARGO	ND58105
KRAFT DONALD J	1970BEMIDJI STATE COLLEGE	BEMIDJI	MN56601
KRAUS OLEN	1969PHYSICS DEPT UND	GRAND FORKS	ND58202
KRESS WAPPEN D	19583FOGRAPHY DEPT NDSU	FARGO	ND58102
KRUISCHWITZ EARL H	1947431 6TH ST SW	VALLEY CITY	ND58072
KURB WAYNE R	1949CHEM ENG DEPT UND	GRAND FORKS	ND58202
KUCERA HENRY L	1966AGR ENG DEPT NDSU	FARGO	ND58102
LAIRD WILSON M	19411807 WAINWRIGHT DR	RESTON VA	22070
LANA EDWARD D	1957HORTICULTURE DEPT NDSU	FARGO	ND58102
LARSON, GARY F	1978BOTANY DEPT NDSU	FARGO	ND58102
LARSON OMER R	1964BIOLOGY DEPT UND	GRAND FORKS	ND58202
LEITE, MICHAEL R	1977RR 2	MINOT	ND58701
LEPPOLD ROGER A	1970BROOKTRFE PK	HARWOOD	ND58042
LI KAM W	1968MECHANICAL ENG DEPT NDSU	FARGO	ND58102
LIPP WILLIAM V	197295 28TH AVE N	FARGO ND	58102
LINDGORE, LAWRENCE L	1973ANTHRO DEPT UND	GRAND FORKS	ND58202
LOGUE, MARSHALL W	1979DEPT OF CHEMISTRY NDSU	FARGO	ND58105
+LOKEN, GREGORY	19782918 GRINNELL CT #10	ROCKFORD	IL61109
LORENZ RUSSELL J	1962N GREAT PLAINS RES CNTR	MANDAN	ND58554
LOVE, JANET	1958108 BRIARWOOD LN	MARS	PA16046
LOW FRANK N	1964ANATOMY DEPT UND	GRAND FORKS	ND58202
MCCARTHY, RONALD F	19682618 CLOVER DR	GRAND FORKS ND	58201
*MACKICHAN RUTH J	1958MATH OPT UND	GRAND FORKS ND	58202
MADHOK OM P	1967MINOT STATE COLLEGE	MINOT	ND58701
+MAERTENS, THERESA A	1978341 S WEIBLE NDSU	FARGO	ND58102
*MAGNUSSON, ADELYNN M	1951703 S 20TH ST	GRAND FORKS	ND58201
MALAND HARTLEY B	1969LAKE REGION JUNIOR COLLG	DEVILS LAKE	ND58301
+MANSKE, LEWFLLYN L	1978BOTANY DEPT NDSU	FARGO	ND58102

MANZ, OSCAR	1970CIVIL ENGR DEPT UND	GRAND FORKS	ND58202
MARKFELL CLARK	1972MINOT ST COLLEGE	MINOT	ND58701
MARTIN DEWAYNE C H	19622104 7TH AVE NW	MINOT	ND58701
MARWIN RICHARD M	1949MICROBIOLOGY DEPT UND	GRAND FORKS	ND58202
MASON, HARRY	19511602 2ND PL NE	JAMESTOWN	ND58401
MATHSEN, DON	19701011 19TH AVE S	GRAND FORKS ND	58201
MATTHIES DONALD L	1973ANATOMY DEPT UND	GRAND FORKS ND	58202
MATSON, MARVIN P	1971422 17TH ST N	MOORHEAD MN	56560
MAXSON, GEORGE-ANN	197823355 GOPHER DR NE	BETHEL	MN55005
MCDONALD CLARENCE E	1965CEREAL TECHNOLOGY NDSU	FARGO	ND58102
+MCDONNELL, TIMOTHY	1978ANATOMY DEPT UND	GRAND FORKS	ND58202
+MCKENNA, MICHAEL	1976ECOLOGICAL INST UND	GRAND FORKS ND	58202
MCMAHON KENNETH J	1970BACTERIOLOGY DEPT NDSU	FARGO	ND58102
*MCMILLAN WILLIAM W	1947407 7TH ST W	GRAFTON	ND58237
MELDRJM ALAN H	1957GENERAL IND ENG DEPT UND	GRAND FORKS	ND58202
MELFTHIL, SRI K	1978COLLEGE OF PHARMACY NDSU	FARGO	ND58102
+MERCIL, STEVE	1977114 CHESTNUT ST	GRAND FORKS	ND58201
MESSENGER, THOMAS	1976PHIL DEPT UND	GRAND FORKS ND	58202
METZGER, CHARLES F	19662023 GRIMSRUD DR	BISMARCK	ND58501
MEYER DWAIN W	1970AGRONOMY DEPT NDSU	FARGO	ND58102
MEYER, MAVIS	1978NORTHERN PRAIRIE WILDLIFE	JAMESTOWN	ND58401
MILLER, JAMES E	19644901 SEARLE PARKWAY	SKOKIE	IL60076
MITCHELL E N	1960220 GLENHILL LN	CHAPEL HILL	NC27514
MOLITOR, THOMAS	19781121 2ND AVE S	FARGO	ND58102
MOORE WILLIAM L	1973VALLEY CITY STATE COLLEGE	VALLEY CITY ND	58072
MORAN STEPHEN R	197011315 87 AVE	EDMONTON	AB
MORRISON, WILLIAM W	1972STATE HEALTH DEPT	BISMARCK	ND58501
MOWERY, GARRY B	1979334 FOREST AVE N	FARGO	ND58102
+MURPHY, KATHLEEN A	1979MICROBIOLOGY DEPT UND	GRAND FORKS	ND58202
NATSMITH DONALD P	1958MECH ENGR DEPT UND	GRAND FORKS	ND58202
NALFWAJA JOHN D	1963AGRONOMY DEPT NDSU	FARGO	ND58102
+NEAL, DEAN	1977500 TULANE DR #201	GRAND FORKS	ND58201
NEEL JOE K	1969BIOLOGY DEPT UND	GRAND FORKS	ND58202
NEIDLINGER, TERRY R	19781912 COTTONWOOD ST	GRAND FORKS	ND58201
*NELSON C N	1972NDSU BOTTINEAU BRANCH	BOTTINEAU	ND58318
NELSON DELBERT R	1961218 E QWASSO LN	ST PAUL	MN55112
NELSON DENNIS R	1964MET & RAD RES LAB NDSU	FARGO	ND58102
NELSON, HARVEY K	19672431 RIVIERA DR	VIENNA VA	22180
NICOLLS, KEN E	1973COMM MED DEPT-U OF IOWA	IOWA CITY	IA52240
NIELSON, FORREST H	1974USDA HJMAN NUTR LAB UND	GRAND FORKS	ND58202
+NILL, KIMBALL	1978REFD HALL #123	FARGO	ND58102
NORDLIE ROBERT C	1962BIOCHEMISTRY DEPT UND	GRAND FORKS	ND58202
NYSTUEN PEDER A	1964EXPERIMENT STA NDSU	FARGO	ND58102
O'CONNELL JAMES W	1973535 8TH AVE SW	VALLEY CITY ND	58072
OGAARD, LOUIS	1976AGRIC ECON DEPT NDSU	FARGO	ND58105
OLESON, ARLAND E	1973BIOCHEM DEPT NDSU	FARGO	ND58102
OLSON, JACQUEFLYN K	1978DGE ENGR RSCH CTR BOX 20	GRAND FORKS	ND58202
+OLSON, KAREN	1978800 10TH AVE NW	MINOT	ND58701
ORING LEWIS W	1971BIOLOGY DEPT UND	GRAND FORKS	ND58202
ORTH, JAMES	1970CP 1076 SCHEFFERVILLE	QUEBEC CANADA	
OVERVOLD, CAROL	1979920 3RD AVE S	FARGO	ND58102
OWEN ALICE K	1966BIOLOGY DEPT UND	GRAND FORKS	ND58202
OWEN, JOHN B	1966BIOLOGY DEPT UND	GRAND FORKS	ND58202
*OWEN, SHIREL D	1958800 PANORAMA RD	PANORA IA	59216
OWENS THOMAS C	1970CHEM ENGR DEPT UND	GRAND FORKS	ND58202
+PAKOLA, HARLEY A	1978PHYSIOLOGY DEPT UND	GRAND FORKS	ND58202
PARK, CHUNG S	1979DEPT OF ANIMAL SCI NDSU	FARGO	ND58105
PARMAR, SURENDRA	1977PHYS & PHARM UND	GRAND FORKS ND	58202
PARRILL, CLARK	1977	DUNSEITH	ND58329
PEDERSON A ROBERT	1972414 20TH AVE N	FARGO	ND58102
PEDERSON VERNYL D	1968PLANT PATHOLOGY NDSU	FARGO	ND58102
PEKAS JEROME C	1968MET & RAD RES LAB NDSU	FARGO	ND58102
PEMBLE RICHARD H	1971MOORHEAD STATE COLLEGE	MOORHEAD	MN56560
+PERSKY, BRUCE	1972ANATOMY DEPT UND	GRAND FORKS	ND58202
PERSON, DONALD A	19613022 WINSLOW	HOUSTON	TX77025
PETERKA JOHN J	1968BIOLOGY DEPT NDSU	FARGO	ND58102
PEISTER, PHILIP C	196830 MEADOWLARK LANE	FARGO	ND58102
PORTER ROBERT R	19561703 4TH AVE N	GRAND FORKS	ND58201
POWER JAMES F	19621608 N 19TH ST	BISMARCK	ND58501
PRATT GEORGE L	1961AGRICULTURAL ENGR NDSU	FARGO	ND58102
PRESZLER, DALE A	19781717 E INTERSTATE AVE	BISMARCK	ND58501
PUYFAR, ROBERT L	1968700LOGY DEPT NDSU	FARGO	ND58102
+RAHMAN, AMRANA	1978216 SQUIRES HALL-UND	GRAND FORKS	ND58202
RAND ROGER W	1975542 5TH AVE SW	VALLEY CITY ND	58072
*RATHMANN, FRANZ H	1955NDSU CHEM DEPT	FARGO	ND58102
+RAY, JOHN T	1979201 WALNUT ST	GRAND FORKS	ND58201
RAY PAUL D	1968BIOCHEMISTRY DEPT UND	GRAND FORKS	ND58202
REDMANN, ROBERT E	19641448 N 1900 E	LOGAN	UT84321
REICHMAN GEORGE A	1962306 6TH AVE NW	MANDAN	ND58554
REID JOHN R	1962GEOLOGY DEPT UND	GRAND FORKS	ND58202
REIFF, THEODORE R	19783607 9TH AVE N	GRAND FORKS	ND58201
+REINISCH, JERRY	19773904 UNIV AVE #220	GRAND FORKS	ND58201
+REISKIND, JEREMY	1978GEOLOGY DEPT UND	GRAND FORKS	ND58202

REITEN PALMER J	1957MECH ENGR DEPT UND	GRAND FORKS	ND58202
RICHARDSON, J L	19781245 N 9TH	FARGO	ND58102
RIEDER WILLIAM G	1973MECH ENGR DEPT NDSU	FARGO ND	58102
RIES, RONALD E	1979908 2ND AVE NW	MANDAN	ND58554
RODEWALD RANDOLPH F	1975SCIENCE DIV MINOT ST COLL	MINOT ND	58701
*ROGLER GEORGE A	196290X 459	MANDAN ND	58554
ROPPFEL, ROBERT D	1978314 COTTONWOOD #2	GRAND FORKS	ND58201
ROTHENBERGER, STEVE	J MIDLAND LUTHERAN COLL	FREMONT	NE68025
RUDESILL JAMES T	1958COLL OF CHEM & PHYS NDSU	FARGO	ND58102
RUJSTAN, JERRIL H	197990X 140 DSC	DICKINSON	ND58601
*SAIKI A K	1949PATHOLOGY DEPT UND	GRAND FORKS ND	58202
SANDAL PAUL C	1955AGRONOMY DEPT NDSU	FARGO	ND58102
*SANDS F H	1946COLL OF CHEMISTRY NDSU	FARGO ND	58102
SARABUN CHARLES C JR	197384 WELSH TR RD 307	NEWARK DE	19711
SARGFANT ALAN R	1972N PRAIRIE WILDLIFE RES	JAMESTOWN	ND58401
SAUMUR JEAN H	1975PATHOLOGY DEPT UND	GRAND FORKS ND	58202
SCATTOLINI, RICHARD	1973GEOLOGY DEPT UND	GRAND FORKS	ND58202
SCHIFFR, PAUL	19603 SHIRLEY LANE	WOODSIDE	CA94062
+SCHIMMELPFENNIG, D	1978618 BOYD DR	GRAND FORKS	ND58201
SCHNEIDER FREDERICK	1973SOC & ANTHRO DEPT UND	GRAND FORKS ND	58202
SCHORBERT, HAROLD	197800E ENGR RSCH BOX 20 UND	GRAND FORKS	ND58202
SCHULZ JOHN T	1960ENTOMOLOGY DEPT NDSU	FARGO	ND58102
SCORY DONALD R	1968BOTANY DEPT NDSU	FARGO	ND58102
SEABLOOM ROBERT W	1962BIOLOGY DEPT UND	GRAND FORKS	ND58202
+SEARS, SHEILA	1977MRR LAB NDSU	FARGO ND	58102
+SEILER, GERALD J	1978210 WALDRON HALL NDSU	FARGO	ND58102
+SEPE, FRANK	1978PHYSIOLOGY DEPT UND	GRAND FORKS	ND58202
SERIF, JEROME R	197890X 1747	JAMESTOWN	ND58401
SEVERSON ARTHUR L	1970U S BUREAU OF MINES UND	GRAND FORKS	ND58202
SEVERSON D F	1949CHEMICAL ENGR DEPT UND	GRAND FORKS	ND58202
SEVERSON ROLAND G	1958CHEMISTRY DEPT UND	GRAND FORKS	ND58202
SHELTON, DAVID R	197830X 5195 NDSU	FARGO	ND58102
SHELVER, WILLIAM	1979COLLEGE OF PHAR NDSU	FARGO	ND58105
SHUBERT L ELLIOT	1974BIOLOGY DEPT UND	GRAND FORKS ND	58202
SHUFY WILLIAM C	1974CERFAL CHEM & TECH NDSU	FARGO ND	58102
SIBBITT L D	1946CERFAL TECH DEPT NDSU	FARGO	ND58102
SILVERMAN LOUIS B	19572524 OLSON DR	GRAND FORKS	ND58201
SINGH, SHINA P	1978PHYSIOLOGY DEPT UND	GRAND FORKS	ND58202
SLEEPER BAYARD D	1952BACTERIOLOGY DEPT NDSU	FARGO	ND58102
*SMITH, GLENN S	19301115 N 14TH ST	FARGO	ND58102
SMITH HARPY C	195880X 145	SAWYER	ND58781
*SNOOK, THEODORE	1954ANATOMY DEPT UND	GRAND FORKS	ND58202
SOMERVILLE MASON H	1974MECH ENGINEERING UND	GRAND FORKS ND	58202
SOURY ARMAND M	1973CHEMICAL ENGR UND	GRAND FORKS ND	58202
STANISLAD, JOSEPH	1979ENG & ARCH-NDSU	FARGO	ND58102
*STAPFHER GEORGE W	19543605 JAFFA DR	SARASOTA	FL33575
+STARCKS, THOMAS	1977BIOLOGY DEPT UND	GRAND FORKS	ND58202
STATLER GLEN D	1970PLANT PATH DEPT NDSU	FARGO	ND58102
STENBERG VIRGIL I	1961CHEM DEPT UND	GRAND FORKS	ND58202
STEWART JAMES A	1960CHEMISTRY DEPT UND	GRAND FORKS	ND58202
STINNETT, HENRY D	1978PHYSIOLOGY DEPT UND	GRAND FORKS	ND58202
STRAKS, RALPH D	6714 NORTHWEST DR	DES MOINES	IA50322
+STRIGGL, PAUL M	1978ROUTE #5	BISMARCK	ND58501
SUGIHARA JAMES M	1965GRADUATE SCHOOL NDSU	FARGO ND	58102
SUMMERS LAWRENCE	1951CHEMISTRY DEPT UND	GRAND FORKS	ND58202
SWANSON GEORGE A	19671727 4TH AVE NE	JAMESTOWN	ND58401
SWANSON RICHARD J	1972507 3RD ST CT	WEST FARGO	ND58078
+TAYLOR, ROBERT	1978NDSU BACTERIOLOGY DEPT	FARGO	ND58102
THACKER EDWARD J	196411 WOODCREST DR	FARGO	ND58102
THOMPSON CLARENCE E	19751526 COTTONWOOD ST	GRAND FORKS ND	58201
THOMPSON, JOAN	1977NDSU BOX 15 STEVENS	FARGO	ND58102
THOMPSON MICHAEL B	19702208 CRESCENT DR	MINOT	ND53701
TILTON JAMES F	1966ANIMAL SCI DEPT NDSU	FARGO	ND53102
TIMIAN ROLAND G	1954PLANT PATH DEPT NDSU	FARGO	ND58102
TIMPE, RONALD C	1973MAYVILLE STATE COLLEGE	MAYVILLE	ND58257
TODD ROBERT G	1962DICKINSON STATE COLLEGE	DICKINSON	ND58601
+TOWARNICKY, MICHAEL	1978715 N 40TH ST #3061	GRAND FORKS	ND58201
TWEED, DANIEL J	1978	MADDOCK	ND58348
VANALSTINE JAMES B	1975DIV OF SCIEMATH UNIV MN	MORRIS MN	56267
VAN DEUSEN JAMES L	1975USES SHELTERBELT LAB	BOTTINEAU ND	58318
VENETT, JAMES R	PLANT PATH-NDSU	FARGO	ND58102
VENNES JOHN W	1957MICROBIOLOGY DEPT UND	GRAND FORKS	ND58202
VINCENT MURIEL C	1957COLL OF PHARMACY NDSU	FARGO	ND58102
VOLESKY, ALLEN F	197800E ENGR RSCH BOX 20 UND	GRAND FORKS	ND58202
WAHTOLA, CHARLES H	1970457 PARK AVE	PEWAUKEE	WI53072
WALLER JAMES R	1971MICROBIOLOGY DEPT UND	GRAND FORKS	ND58202
WALSH ROBERT G	1969MINOT STATE COLLEGE	MINOT	ND58701
WANCK WALLACE J	1965RT 1 BOX 307	HEMIDJI	MN56601
+WARD, GREGORY T	197824 9TH AVE N #4	FARGO	ND58102
*WARDNER, C ARTHUR	19583518 CHERRY ST	GRAND FORKS	ND58201
WASTVEDT, ROBERT D	19701720 4TH AVE E	WILLISTON	ND58801
WATFEL, ALBERT A	19791071 W 5TH ST	DICKINSON	ND58601

WEISSER WILBUR D	1957PHYSICS DEPT UND	GRAND FORKS	ND58202
WELLS, ERNEST A	1977BIOLOGY DEPT MSU	BOZEMAN	MT59715
WERTH RICHARD G	1964CONCORDIA COLLEGE	MOORHEAD	MN56560
+WETSCH, JOHN R	1977BOX 277	KILLDEER	ND58640
*WHEELER GEORGE C	1924DESERT RES INST UNIV NV	RENO	NV89507
WHITE GLYNDON	1972BOX 1394	JAMESTOWN	ND58401
WHITMAN WARREN C	1950BOTANY DEPT NDSU	FARGO	ND58102
WICKS ZENO W	1974POLYMRS & COAT DPT NDSU	FARGO ND	58102
WIEDERANDERS R E	1968HARMON PARK CLINIC	WILLISTON	ND58801
*WIIDAKAS WILLIAM	1946AGRONOMY DEPT NDSU	FARGO ND	58102
WILLIAMS NORMAN D	1965AGRONOMY DEPT NDSU	FARGO	ND58102
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