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EFFECTS OF FERTILIZERS ON CULTURED SALT

MARSH PLANTS, RUPPIA AND CHARA

by

Scott Marshall Stenquist

A thesis submitted in partial fulfillment of the requirements for the degree

of

MASTER OF SCIENCE

in

Wildlife Biology

Approved:

Maior Professor

Committee Member

Committee Member

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UTAH STATE UNIVERSITY Logan, Utah

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Scott Marshall Stenquist

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ABSTRACT

Effects of Fertilizers on Cultured Salt

Marsh Plants, Ruppia and Chara

by

Scott M. Stenquist, Master of Science

Utah State University, 1974

Major Professor: Dr. Jessop B. Low Department: Wildlife Science

Widgeongrass (<u>Ruppia maritima</u>), muskgrass (<u>Chara sp.</u>), and midge larvae (family Chironomidae) were grown under controlled greenhouse conditions using a solution of Logan tap water, 3000 ppm sodium chloride, and an algicide-fungicide inhibiter. Soil, vegetation, and invertabrates came from a spring-fed marsh in western Utah. Ammonium sulfate, treble superphosphate, and sewage sludge fertilizers were applied to the plants at 5, 10, 20, 25, 50, 75, 100, and 300 pounds per acre equivalents of ammonium sulfate; 5, 25, 50, 75, 100, and 300 pounds per acre equivalents of treble superphosphate; and 5, 25, 100, 200, 300, 400, 500,700, and 1000 pounds per acre equivalents of sewage sludge. Widgeongrass plants had weights below the control at all fertilizer levels. Plant lengths at treatment levels of 10 pounds per acre equivalents ammonium sulfate and 5 pounds per acre equivalents sewage sludge were greater than the control length, but the differences were not significant. Muskgrass plants had weights which were neither significantly above or below the control weight. Muskgrass plants at the 75 pounds per acre rate of treble superphosphate had significantly greater lengths than did the control. The highest chironomid survival rate, 12.6 percent, occurred in the control treatment. Bluegreen algae, <u>Oscilatoria</u> sp., was present in all fertilized treatments.

(97 pages)

INTRODUCTION

Penfound (1952) classifies swamps and marshes according to water quality and water depth: fresh, brackish, or salt, and shallow, intermediate, or deep. Water sources such as streams or springs which feed wetland areas may also be important criterion for marsh classification.

Many western marshes which are managed for waterfowl are stream-fed. Since streams may travel hundreds of miles before emptying into the marsh, inorganic and organic nutrients may be brought into the marshland areas. Ground water and tributary rivers collect and transport dissolved nutrients. Agricultural, industrial, and domestic effluents may also enrich stream waters.

Water which is finally deposited in the marsh bears an accumulation of nutrients, silt, and organic material. Plant growth is rank, and symbiotic invertebrates prosper. Competition between plant species is primarily for space and light rather than for nutrients (Nelson, 1955).

As a result of aboundant plant and invertebrate numbers, waterfowl production is considered good. Along stream-fed systems on the eastern shore of the Great Salt Lake, Jensen and Chattin (1964) pointed out that there may be as many as 60-80 nests per acre in some areas. Williams and Marshall (1938) reported 0.8 nests per acre at Bear River Migratory Bird Refuge. Near the Bear River Refuge on Knudson Marsh, a density of 5.3 nests per acre was recorded (Wingfield, 1951).

Other areas which receive river water also show high waterfowl productivity. On the Blitzen Unit of Malheur National Wildlife Refuge (NWR), Oregon, there were 0.84 nests per acre (Jarvis and Harris, 1971). Gray's Lake NWR is a managed stream-fed marsh of southern Idaho which produces 40 nests per acre along some stream edges. Jensen and Chattin (1964) cited breeding densities per square mile on unmanaged and managed river-fed areas. Twenty breeding pairs per square mile were recorded for the area where the Columbia and Kooteny Rivers originate in British Columbia as well as for the Sacramento and San Joaquin Rivers in California's Central Valley. Managed areas such as Tule Lake and Lower Klamath NWR, however, have supported as many as 256 pairs per square mile.

Spring-fed marshes occur in arid regions of the west. Unlike streams, springs generally do not accumulate inorganic and organic material because of their proximity to the areas they feed. Run-off from surrounding areas supply minimal amounts of nutrients because of scant rainfall. In many cases, springs upwell through saline drylake areas; these waters supply large amounts of dissolved sodium and chloride.

The lack of nutrients, silt, and organic material inhibit plant development. Natural build up of materials through the decay of small plant populations may take hundreds of years. With no inflow of organic

matter and minimal organic build up through decay, old and newly flooded areas of refuges are often barren of vegetation. Where vegetation does exist, it is usually small and poor in quality. Plant associated invertebrates are also often limited.

Waterfowl production on spring-fed areas is generally low. At Clear Lake Waterfowl Management Area (WMA), Bergen (1941) found 0.015 nests per acre. During the same period, Locomotive Springs WMA produced 0.004 nests per acre (Smith, 1941).

Fish Springs National Wildlife Refuge, Utah, is a typical springfed refuge which exhibits a low production phenomenon. McKnight (1969) recorded 0.62 and 0.85 nests per acre for 1967 and 1968. Brood production was 18.56 and 19.2 broods per square mile. Canadian praire potholes regularly produce between 30 to 50 broods per square mile while Malheur and Summer Refuges in Oregon generally have from 300 to 700 ducklings per square mile (Jensen and Chattin, 1964).

Four northern units of Fish Springs contain little vegetation even though water has been present in these impoundments for 8 years; they are essentially barren flooded salt flats. Other central units contain some submergent vegetation, but their density flucuates according to the specific location on the unit.

The marsh soil and water of the refuge are highly saline and comparatively infertile. These conditions are now believed to be responsible for the limited quantity of waterfowl food plants, limited suitable nesting sites, and limited populations of important invertebrate fauna. All of these conditions may have a direct bearing on the waterfowl production of the area.

This project is number four in a series concerning waterfowl populations on spring-fed marshes in western Utah by the Utah Cooperative Wildlife Research Unit. Other projects concerning waterfowl on spring-fed marshes were Bolen (1962), Meyer (1967), and McKnight (1969).

Objectives

Artificial enrichment of Fish Springs NWR will be examined with the introduction of nitrogen, phosphorous, and sewage sludge fertilizers on native aquatic submergent vegetation under greenhouse conditions. Project objectives include:

1. To determine the response of widgeongrass to fertilizer treatment.

2. To ascertain the effect of artificial fertilizers on invertebrates.

3. To propose a management plan for spring-fed marshes where submergent and emergent vegetation is limited.

LITERATURE REVIEW

Land fertilization has been practiced by man for thousands of years. The literature concerning fertilization of man's agricultural crops is numerous, but research about fertilization of wildland is comparatively small. Highlights of range, upland, and aquatic investigations will follow.

Animal populations are dependent on vegetation for food and cover. When vegetation is high in protein and abundant, associated wildlife are in greatest numbers. Fertile soil yields a superior quality of plants. Allen (1954) summarized this point: "Good soil yields the best crops, both in quantity and quality, of practically everything that lives upon them."

Denny (1944) noted that better soil fertility produced larger animals. Small game populations examined according to habitat soil type showed that average weights and average lengths were greater than those from less fertile areas (Crowford, 1950).

Commercial fertilizers have been applied to improve native and introduced plant cover on range lands. Rogler and Lorenz (1957) noted that western wheatgrass showed a darker color and increased growth after application of nitrogen. Under both moderate and heavy grazing, 90 pounds per acre (lbs./ac.) of nitrogen produced the greatest yield which was significantly better than the control.

Four brush species in California were treated with 1 level each of nitrogen, phosphorous, and potassium in pots with soil from their respective collection sites. Fertilizer combinations were control, nitrogen-phosphorous, nitrogen-potassium, phosphorous-potassium, and nitrogen-phosphorous-potassium. Wedgeleaf ceanothus, two species of deerbrush, chamise, and western mountain mahogany responded best to treatments which contained nitrogen. Yield for all four species, expressed as percentages of the nitrogen-phosphorous-potassium treatment, were from 49 to 100 percent with nitrogen and were from 7 to 29 percent with the control (Schultz et al., 1958). Other ranges in California produced significantly better herbage production in both wet and dry seasons when sulfur and nitrogen were applied together as well as sulfur applied singly (Conrad et al., 1966).

Plant cover manipulations through the addition of fertilizer has been done in semidesert grassland communities. Tobosa grass on a semi-desert range was fertilized individually with ammonium sulfate at 60 and 90 lbs./ac. and with treble superphosphate at 13 lbs./ac. The yield of the second replication was significantly better than the control except for the 13 lbs./ac. rate of phosphorous (Herbel, 1963).

Holt and Wilson (1961) used ammonium nitrate and ammonium phosphate in 25, 50, and 100 lbs./ac., respectively, on desert ranges of lehmann lovegrass and Santa Rita threeawn. All treatments increased the total forage significantly above the control. Cattle utilization was

from three to five times higher on the treated plots as compared to the control.

Seeded foothill ranges, seeded mountain ranges, and native mountain ranges in Utah were supplied with ammonium sulfate and treble superphosphate over a period of 7 years (Cook, 1965). Nitrogen was applied on established crested wheatgrass foothill range at **20**, 30, 40, and 60 lbs. /ac. along with 20, 40, and 60 lbs. /ac. of phosphorous, individually and in combination. All individual applications of nitrogen were higher than the control. Forty and 80 lbs. /ac. of nitrogen on established smooth brome grass mountain sites also produced better yields than the control. Native mountain meadows of grasses and forbs were supplied with 80 lbs. /ac. of nitrogen and phosphorous, individually and in combination. Although all treatments increased forage amounts, nitrogen had a greater effect on grasses and forbs together and on grasses alone.

Cook (1965) notes that the carry-over effect of fertilizers on forage yield varied from 0 years (no effect) to 4 years. There appeared to be no correlation between the carry-over effect and the rate or type of fertilizer applied.

In an attempt to rehabilitate big game ranges, nutrients have been artifically applied. Flowering dogwood plots were fertilized individually with muriate of potash, rock phosphate, and nitrate of sodaammonium sulfate. Deer browsed the plots with the nitrate of sodaammonium sulfate more heavily than any other treatment including the

control (Mitchell and Hosley, 1936). Sugar concentrations in the leaves increased proportionally with the application of nitrogen. Mitchell and Hosley (1936) concluded that deer use appeared to be correlated with nitrogen and sugar content rather than with leaf moisture, size, or color.

Experimental plantings of oats, orchard grass, and red clover were made on the Olympic Game Range, Washington, in an attempt to reduce deer and elk damage to agricultural crops on private lands in the area. Ammonium sulfate and ammonium nitrate applications caused the deer and elk to decrease their use of unfertilized private land and unfertilized portions of the game range (Knott, 1956).

Knott (1956) reported similar results of deer use change on an alfalfa field which received superphosphate. Although nearly 90 percent of the deer use was transferred to that portion of the field which received the phosphate nutrients, the fertilized area still produced more forage per acre than did the unfertilized area.

Nitrogen, phosphorous, and sulfur were applied individually and in combination to Douglas-Fir plots at the rate of 100 lbs./ac. each. Deer use, measured by the difference between new shoot length in fenced and unfenced plots, was 10.4, 10.1, and 1.5 for nitrogen, nitrogenphosphorous-sulfur, and control, respectively (Oh et al., 1970). The effect of other fertilizers on the forest environment has been reported (Mustonoja and Leaf, 1965).

Rodents and livestock have also used fertilized vegetation over non fertilized vegetation. Gessel and Orians (1967) reported that

applications of nitrogen and potassium individually and in combination produced improved needle weight and length on Pacific Silver Fir. After fertilization, rodent use increased from 11 to 37 percent on specific treatments as compared to 6 percent on the control.

Smith and Lang (1956) found that nitrogen application to vegetation during the growing season would alter the grazing pressure by livestock. Thomas et al. (1964) and Hooper et al. (1969) felt that livestock use of the range might be more evenly distributed with the use of fertilizer. Cook (1965), however, cautioned that it was still necessary to introduce livestock to the specific fertilized area; they would not seek out new grazing territory.

Rowe (1947) and Bruna (1952) examined cottontail rabbit populations in Missouri and Kentucky, respectively. They reported a correlation between soil fertility and body weight. In order to substantiate or refute these findings, Williams (1964) harvested cottontail rabbit samples in Stoddard County, Missouri. He found that there appeared to be no significant relationship between body weight and soil fertility or soil series.

Further cottontail rabbits investigations were done by Hill (1972). In Alabama, two soil regions which were considered to be the most fertile areas of the state produced significantly larger second-of-the-year litters. Hill (1972) raised cottontails under pen conditions to determine the response of the rabbits to fertilized soils. It was concluded that the pooled litter size of the pen which received 2 tons of lime and 400 pounds of

12-24-24 was not statistically significant from the pooled litter size of the control pens.¹ Larger litters, however, came from pens receiving lime and fertilizer applications; plant size and color were similarly affected (Hill, 1972).

Fertilization effects on plant production have also been examined for upland sites. Miller (1968) recorded the effects of nitrogen and calcium di-hydrogen phosphate on red grouse, mountain hares, and cottontail rabbits which used heather as a food source on the Scottish moor study site. Animal selectivity on various heather plots was measured by counting the number of droppings deposited by each animal. In the winter, the number of feeding grouse was greatest on heather which received the highest amount of nitrogen. There were no differences in feeding selectivity by grouse on nitrogen plots in the summer nor on phosphorous plots in the winter or summer. Miller (1968) did not obtain similar conclusive data for selective feeding by hares or cottontails.

On the light textured soils of southern New Jersey, Toth et al. (1960) observed that burnet produced 149 lbs./ac. and 128 lbs./ac. of seed when treated with 600 and 1000 lbs./ac. of 5-10-10. Further experiments with burnet on the Colliers Mills Public Hunting and Fishing Area, New Jersey, were made by Toth et al. (1964) for 3 years. Treatments were made yearly and consisted of the control, 30 lbs./ac.

¹12-24-24, a standard method of reporting fertilizers in percentages of nitrogen, phosphorous, and potassium, respectively.

of nitrogen as urea, 60 lbs./ac. of nitrogen as urea, 60 lbs./ac. of superphosphate, 100 lbs./ac. of superphosphate, 60 lbs./ac. of muriate of potash, 100 lbs./ac. of muriate of potash, 300 lbs./ac. of 5-10-10, 600 lbs./ac. of 5-10-10, and 1000 lbs./ac. of 5-10-10. Prior to fertilization, the entire areas was limed with dolomitric limestone and 400 lbs./ac. of 5-10-10 was applied.

The rate of 600 and 1000 lbs./ac. of 5-10-10 yielded more seed than other combinations, 247 and 229 lbs./ac. of burnet seed, respectively. This result was identical to that obtained in earlier experiments by Toth et al. (1960). Toth et al. (1960) suggested that rates above the 600 lbs./ac. of 5-10-10 reduced yields because of increased salt injury to plants.

Fertilization projects with ponds have demonstrated that fish respond to nutrient addition. Hogan (1933) found that ponds fertilized with 125 pounds of 18 percent superphosphate produced greater numbers of larger sized largemouth black bass than did ponds which were treated with 125 pounds of 6-8-6 and 250 pounds of cottonseed meal. Meeham (1933) reported similar results.

Treatments using superphosphate, superphosphate and sodium nitrate, and superphosphate and ammonium sulfate produced 134, 156, and 174 lbs./ac. of fish (Swingle and Smith, 1938). Swingle (1939) used ammonium sulfate, acid phosphate, muriate of potash, and limestone for experimental fish pond treatment. Application by hand was made often enough to keep the water green. Fertilized ponds produced 580 lbs./ac. of fish while the control yielded 150 lbs./ac. of fish. Smith and Swingle (1941) concluded that inorganic fertilizers gave the best results of plankton and fish production because the chemicals were more available as compared to organic fertilizers. Patriarch and Ball (1940) working with 100 lbs./ac. of 10-6-4 found that fish production was definitely higher in fertilized ponds.

Marsh studies of Sherwood (1966) and Toth et al. (1972) initiated fertilization of vegetation for waterfowl use. Artificial nesting islands constructed on Seney NWR were limed at 2 tons per acre and fertilized with 300 pounds per acre of 23-28-14 (Sherwood, 1966). Rye, Canadian bluegrass, brome grass, alkali clover, and fescue were sowed after chemical treatment. Geese responded immediately by loafing and preening on the islands. As the grasses developed, the birds grazed on the shoots. Nesting increased 30 percent over the previous year even though there was a decrease in the total number of Canada Geese nesting pairs on the refuge. Fertilization substantially improved the island habitat.

Toth et al. (1972) examined the effects of fertilizing smartweed on an aquatic wildlife management area in New Jersey. Fertilizer combinations consisted of a control, 30 and 60 lbs./ac. of nitrogen as urea, 60 and 100 lbs./ac. of superphosphate, 60 and 100 lbs./ac. of K_2^0 as muriate of potash, 300 and 600 lbs./ac. of 5-10-10, and a combination of 100 lbs./ac. of nitrogen, 100 lbs./ac. of phosphorous, and 100 lbs./ac. of K_2^0 . The fertilizers were applied during the spring of all four study years.

Four year averages showed that the 100 lbs./ac. of nitrogen, 600 lbs./ac. of 5-10-10, and 100 lbs./ac. of the nitrogen phosphorouspotassium combination produced the greatest yields: 48.83, 41.18, and 53.00 lbs./ac., respectively. The 4 year average for the control was 15.73 lbs./ac. All treatments, however, had statistically greater yields over the control during the first and third years; other yields for the second and fourth years could not be analyzed due to insufficient replications. Total nitrogen and crude protein analysis of seeds and tops indicated that fertilization had a minimal effect of changing the nitrogen and protein percentages.

LOCATION AND ORIENTATION

The Fish Springs marsh was recognized as an oasis in the hostle environment of the early west. Indians camped in the area and presumably killed animals that came to the springs to water. The Pony Express built a permanent rest station for its riders at the springs. The first transcontinental telegraph and road were part of the early history of Fish Springs. Bolen (1962) provides a vivid historical narrative of the region.

Five major springs and numerous other minor springs, collectively called Fish Springs, feed the marsh. These springs rise from a fault zone on the east side of the Fish Springs Range. The upwelling waters passes through saline lakebed sediments and is discharged onto the salt desert floor. As a result, the springs have a high sodium and total dissolved solid content.

Fish Springs National Wildlife Refuge is in the cold desert of the Great Basin (Figure 1). Precipitation averages between 6 and 8 inches per year. Temperatures range from the high 90's in the summer to the low teens in the winter. In Juab County, Utah, the refuge remains isolated from urban population centers. The Nevada border is nearly 40 miles west, and Salt Lake City, Utah is 140 miles northeast.

The marsh is surrounded by the Great Salt Lake Desert to the north, the Dugway and the Thomas Mountains to the east, the Fish



Figure 1. Location map of Fish Springs National Wildlife Refuge.

Springs Flat to the south, and the Fish Springs Mountains to the west. The northern salt desert also serves as a military range for the Army's Dugway Proving Ground. Vehicular access to the site is on secondary dirt roads from Dugway or Delta, which are approximately 70 and 80 miles east and southeast of the refuge, respectively.

The original marsh was confined to the springheads and their adjacent sloughs. Avocet Unit contains much of the primary marsh which lies on large natural peat deposits.

Eight other impoundments are man made and consist of ponds located on the silty clays of the Great Salt Lake deposit. After flooding for nearly 14 years, Mallard, Shoveller, Pintail, Harrison, Gadwall, Ibis, Egret, and Curlew pools have limited growths of emergent vegetation (Figure 2). The Avocet area clearly has a greater population of emergents.

Water quantity is a limiting factor in the summer. Evaporation accounts for a water loss averaging 70 inches per year. Water is available for those impoundments near the springs, but the eastern management units of Gadwall, Ibis, and Egret pools dry up. The canals and ditches can not supply enough water for the western and eastern management areas.

MATERIALS AND METHODS

All experimental work was conducted in a university owned greenhouse on campus. Water and air temperatures were regulated with steam heating and evaporative cooling systems. A series of movable glass loovers allowed additional air circulation when needed. During fall and winter, artificial light from fluorescent lamps extended the growing period. Natural light, however, was used exclusively during the spring and summer.

Germination Studies

Muskgrass (<u>Chara sp.</u>) spores required no special collection or germination techniques. Widgeongrass (<u>Ruppia maritima</u>) seeds were collected from adult plants and from bottom soil on the Shoveller Unit of Fish Springs NWR. Before seed sorting, vegetation and soil were separated with wash and rinse cycles. Wet seeds were more visible within the vegetation than dry seeds. Seeds were stratified at 0 and 32^oF. Other investigators also noted that widgeongrass seed required stratification (Joanen, 1964; Gore, 1965; and Meyer, 1967).

The seeds were placed in tap water under natural light. Tap water was added as needed. Seeds were checked daily for radicle penetration through the seed coat; this point was defined to be seed germination.

The count for the germinated and ungerminated seeds took place approximately 10 days after the first sign of germination.

Fertilization Studies

The basic experimental design had rooted aquatics, widgeongrass and muskgrass, growing from potted Fish Springs soil. Pots were submerged in 8 and 20 gallon aquaria and plastic buckets filled with tap water, respectively. Four plants were used for each pot in all experiments. Each treatment had four replications. There were a total of six complete runs. One treatment was assigned to each container in a completely randomized manner.

The initial solution for all experiments was made with Logan tap water plus 3000 ppm sodium chloride, and an algicide-fungicide inhibitor. The addition of 3000 ppm sodium chloride better approached the actual saline conditions on site. Algae and fungi inhibitors were added to the salt solution at the time of mixing. Copper sulfate and a Captan garden fungicide were dissolved in the water at the rate of 0.5 and 20 ppm, respectively. No detrimental effects of these compounds were noted on plant growth or reproduction. Meyer (1967) also observed that the copper sulfate and the garden fungicide chemical produced no negative interactions on the aquatic growth. After the initial solution was added to all containers, only Logan tap water was used to replenish the aqueous solution lost through evaporation (Table 1).

Cation	ppm	Anions	ppm
Calcium	48	Chloride	2
Magnesium	16	Sulfate	9
Sodium	6	Carbonate	0
Potassium	1	Bicarbonate	214
Electrical conductivity:	350 micromho	s per cm.	
Total dissolved solids:	166 ppm		

Table 1. Analysis of Logan City tap water

Soil used for plant growth was collected from the Shoveller Unit at gate structure STR D-3 (Table 2). Efforts were made to collect the soil in one general area and at a constant distance from the soil surface. The sampling site was selected several hundred yards north of the dike to avoid the borrow pit area.

The material was scooped into cans and brought back to the greenhouse. After drying for a minimum of 3 to 4 weeks, the soil was sifted and mixed to insure uniformity. The material was potted into 3 inch pots and allowed to settle before planting.

After germination, widgeongrass seeds were planted. Four seeds per pot were sown. All seeds showed the main green leaf above the soil surface after sowing. Dead and washed out seeds were replanted prior to fertilization. After the fertilizer had been added, the

	Soil	
рН		7.8
Salinity		5400 µmhos/cm.
Phosphorous		38 ppm
Potassium		650 ppm
Nitrogen [Total]		.3 percent
Organic carbon		4.5 percent
Water soluble sodium		32.6 me./1.

Table 2. Analysis of soil and water collected from Shoveller Unit, Fish Springs National Wildlife Refuge, September 1972 and April 1973, respectively

Wa	ter
----	-----

Salinity	4722 µmhos/cm.
Phosphorous	$3 \ \mu g \ PO_4 - P/1.$
Nitrite	$2 \mu g NO_2 - N/1$.
Nitrate	5 μ g NO ₃ -N/1.
Sodium	590 mg/1.
Chlorine	1042 mg/1.
Sulfate	740 mg/l.

seeds were not replaced and were considered in the weekly mortality survey.

Muskgrass sporangia were present naturally in soil; they were not artificially cultured to produce germination. The soil was sifted and mixed prior to potting, and the distribution of muskgrass sporangia was assumed to be completely random throughout all of the pots.

Pelleted forms of ammonium sulfate, treble superphosphate, and sewage sludge supplied the nitrogen and phosphorous which were used during the entire project. The fertilizers were placed on the water surface. Mechanical stirring helped mix the entire solution in each tank. Sewage sludge had been stored outside prior to acquisition and had picked up moisture. The sludge was dried for 2 to 3 days before application (Table 3).

Table 3. Analysis of sewage sludge fertilizer used in experiments, 1972-1974

PH	6.5
Salinity	4200 μ mhos/cm.
Nitrogen [Total]	2.2 percent
Organic carbon	32.2 percent
Potassium [Total]	2100 ppm
Phosphorous [Total]	5.6 percent

Fertilizers were added on an equivalent pounds per acre (lbs./ac.) basis. The concentrations of ammonium sulfate were 5, 10, 20, 25, 50, 75, 100, and 300 lbs./ac. equivalent. Treble superphosphate was applied as 5, 25, 50, 75, 100, and 300 lbs./ac. equivalent. Sewage sludge was added as 5, 25, 100, 200, 300, 400, 500, 700, and 1000 lbs./ac. equivalent.

Ammonium sulfate, treble superphosphate, and sewage sludge had electrical conductivities of 6506, 5169, and 4200 μ mhos/cm., respectively,

Widgeongrass and muskgrass were harvested between 7 and 10 weeks after fertilizer treatment. Leaves, stems, roots, and reproductive structures were removed from the soil and washed; these parts were oven dried for 2 days at 55°C. A Sartorius brand milligram beam balance was used for weighing. The leaf length for each species was measured. The leaf was measured from the root crown to the end of the longest leaf axil. Flowers and seeds were counted on widgeongrass, but were simply noted on muskgrass. The abundance of reproductive structures on muskgrass made counting impractical.

Treatments were checked daily for algae, general growth, and abnormalities. Plant mortality was recorded weekly along with reproductive data. Environmental conditions of temperature, light, and water level were also monitored.

Chironomid larvae were selected as the invertebrate indicator organism. They were collected from the soil and the submergents on the Shoveller area. Ten organisms were introduced into each treatment during the last experiment as a pilot study. There was only one replication.

The laravae were noted during the daily observation but they were only counted for mortality purposes during the harvest. Because of their burrowing habit, it was not feasible to disturb the potted plant for a weekly mortality survey of the Chiranomides. The midges were collected at the end of 10 weeks; they were dried and weighed in the same manner as the vegetation.

RESULTS AND DISCUSSION

Widgeongrass, muskgrass, sago pondweed (<u>potamageton</u> <u>pectinatus</u>), and spiny naiad (<u>Najas quadalupensis</u>) were noticeable submergents found in plant transects on the Mallard and Shoveller Units of Fish Springs NWR. Of these four plants, widgeongrass and muskgrass were the most abundant plants.

Widgeongrass and muskgrass are important at Fish Springs National Wildlife Refuge. Because of their abundance over other aquatics and because waterfowl readily accept them as a food source, widgeongrass and muskgrass would appear to be satisfactory submergent indicators of aquatic fertilization.

Response of Ruppia and Chara to Fertilizers

Plant reproduction and growth

Reproduction and growth of <u>Ruppia</u> and <u>Chara</u> were monitored to determine the plants response to fertilizer treatment.

<u>Mortality.</u> Widgeongrass mortality was classified as physical and chemical. Physical mortality occurred when seedlings were destroyed because of the shallow planting depth. The seed cost and the radicle worked to the top soil layer. The seedlings were moved from the soil to other locations within the tank. Occasionally, the seedlings
were uprooted from the soil when the tap water was added to maintain the water level. The agitation and oscilation of small currents caused the newly rooted plants to loosen in the soil. These two problems were generally confined to the first week after the plants were placed in their environment. All seedlings displaced during this time were replanted with newly germinated seeds. After the first week, most plants were well established and mortality from the physical factor was nearly 0 percent.

Chemical factors were considered responsible for all mortality once the nutrient treatment had been added. Although algae which covered specific widgeongrass stalks may have been responsible for their ultimate death, the algae growth prospered as a result of nitrogen, phosphorous, and sludge treatments.

Plant mortality for widgeongrass was not constant from week 1 to week 10 (Table 4). The highest average mortality, 23.3 percent of the total pots in the experiment, occurred at the end of week 5. The range was from 3.1 to 23.3 percent for week 1 and week 5, respectively.

Widgeongrass plants died at an increasing rate up to and including the fifth week of treatment. The rate decreased beyond the fifth week. The immediate consequence of nutrient enrichment appeared less lethal to widgeongrass plants compared with the prolonged effects of fertilization.

Ruppia plant deaths were categorized according to nitrogen,

			Phos-	Inter-	
	Nitrogen %	Sludge %	phorous %	Action %	Control %
Week l	3.0	4.2	6.3	2.2	0.
Week 2	10.8	7.5	4.6	6.9	0.
Week 3	23.1	13.6	9.0	17.8	2.5
Week 4	21.3	19.4	13.8	23.8	11.2
Week 5	27.5	22. 2	24.4	35.5	6.7
Week 6	25.3	24.2	20.6	27.6	6.3
Week 7	20.3	24.2	27.5	27.1	8.8
Week 8	19.6	24.2	21.9	16.7	6.3

Table 4.	Summary of widgeongrass	mortality by	treatment	group and
	by week for experiments.	1972-1974		

	Widgeongrass mortality	by week
	%o	
Week l	3.1	
Week 2	6.0	
Week 3	13.2	
Week 4	17.9	
Week 5	23.3	
Week 6	20.8	
Week 7	21.6	
Week 8	21.4	

phosphorous, nitrogen and phosphorous interactions, sludge, and control (Table 4). Plant deaths occurred most often under the interaction treatment followed by nitrogen, sludge, and phosphorous. Plants never having been exposed to fertilizer had the lowest mortality. <u>Ruppia</u> plants from fertilized treatments, regardless of whether the nutrient source was organic or inorganic, had the lowest survival rate.

Muskgrass occurred as an invader in all experiments. The <u>Chara</u> plants did not appear to be susceptible to the physical type of mortality. Seedlings which developed from sporangia were firmly anchored in the soil. Water movement did not uproot or loosen immature muskgrass.

Immature muskgrass, which had germinated from spores present in the soil, penetrated the soil surface within 3 weeks after the pots had been emersed. Since these plants had no vegetation exposed to the nutritive treatment, two experiments used adult muskgrass. Adult plants were transplanted from the widgeongrass seed plant trials. The adult plants received the only comprehensive mortality examination.

The adult muskgrass responded negatively to the fertilizer treatments. At the end of the first week of treatment, most <u>Chara</u> plants had died. Dead plants turned white and lost all of their green characteristics. Chlorophyll was depleted, and little turgor pressure existed. Muskgrass, however, in the control remained green. Muskgrass exhibited similar mortality traits in earlier experiments (Teeter, 1963). Mortality during the first week was 66.5 percent for all treatments.

During the first 2 weeks after treatment occurred the highest number of muskgrass deaths, 66.5 and 67.8 percent, respectively. Mortality dropped with the remaining successive weeks (Table 5).

Since individual plants were uprooted and replanted in fresh soil, transplant shock may have decreased the vigor of the plants. The salinity of the water changed from 166 ppm in the fresh water to at least 3000 ppm in the control solution. The osmotic difference together with the decreased vigor of the plants may have caused the excessive mortality or zero growth. The converse of this statement appears true: Bernstein (1963) reported that plants often exhibited unusually large growths when removed from a saline solution to a non-saline solution.

Plant deaths were grouped according to their respective nutrient classification. Nitrogen, phosphorous, nitrogen-phosphorous interaction, sludge, and control treatments occurred at least once in each treatment.

Plants of the control group had the lowest average death rate of 10.4 percent. <u>Chara</u> from the interaction effects of nitrogen and phosphorous had the highest plant mortality while plants from the nitrogen and sludge treatments had the second and third highest mortality, respectively. Plants from the phosphorous treatment had the lowest number of plant deaths. Plants in the sludge treatments may have had an excessive mortality rate because they were run in only one of two experiments, and the "average" was not the true average of two experiments (Table 5).

The massive <u>Chara</u> die-off did not occur when immature plants were used for further fertilization tests. Immature vegetation was not

Muskgrass mortality by treatment						
	Nitrogen %	Sludge %	Phos- phorous %	Inter- action %	Control %	
Week l	91.7	100.	62.5	78.4	0.	
Week 2	87.5	100.	56.3	83.0	12.5	
Week 3	20.8	41.7	1 2. 5	70.5	12.5	
Week 4	22.9	0.	6.3	40.9	0.	
Week 5	16.7	0.	0.	40.9	12.5	
Week 6	1 2. 5	0.	0.	12.5	12.5	

Table 5. Summary of muskgrass mortality by treatment groups and by week for experiments, 1972-1974

Muskgrass mortality by week

	· · · · · · · · · · · · · · · · · · ·	
Week l	66.5	
Week 2	67.8	
Week 3	31.6	
Week 4	14.0	
Week 5	8.3	
Week 6	3.4	

grown in a fresh water medium and transferred to fertilized treatments. The young plants were not subjected to the sudden osmotic change from the fresh water to the saline water.

<u>Reproduction.</u> Seed production in all <u>Ruppia</u> treatments was minimal. Immature plants in experiments 4 and 5 produced a total 8 and 12 seeds. The control, salt only, and 5 lbs./ac. ammonium sulfate treatment had widgeongrass plants with seeds in experiment 4. Experiment 5 had seeds in the control, salt only, 5, 10, and 20 lbs.ac. equivalent of ammonium sulfate.

Mature plants were raised from seed in the control solution for 10 weeks before they were exposed to treatments. At 10 weeks of age, the plants were removed from the control solution and subjected to experimentation. Plants in the 100 lbs./ac. treatment of ammonium sulfate had eight seeds while the plants of the 5 lbs./ac. treatment of ammonium sulfate had two seeds. Immature plants were grown in the same tanks using identical treatments, but no seeds were produced from immature plants.

Plants at the 5 lbs./ac. ammonium sulfate rate were the only ones which consistently produced seed in three of four experiments where the 5 lbs./ac. treatment of ammonium sulfate nitrogen was present. While the total number of seeds ranged from two to four, plants from the treatment failed to produce seed in only one experiment. Immature widgeongrass was used in the 5 lbs./ac. ammonium sulfate treatment.

Plants from ammonium sulfate concentrations of 10 lbs./ac. and 20 lbs./ac. yielded seeds in experiment 5, but no other plants in the experiments with the 10 and 20 lbs./ac. concentration produced seed (Table 6).

Seed numbers, weights, and lengths were compared between fertilized and unfertilized treatments. The average seed number and corresponding weights were highest on the control treatments. The

Treatment ^a	Experiment	Number	Ave.Weight	Ave. Length
100,0	7	8 ^b	1.5434 g.	10.25 in.
5,0	7	2 ^b	0.2671	5.38
salt	5	3	0.0964	5.63
control	5	4	0.0959	5.56
5.0	5	2	0.0951	11.25
10,0	5	2	0.1120	5.00
20,0	5	1	0.0394	3.00
5,0	4	4	0.3052	6.94
control	4	4	0.0959	5.56

Table 6. Summary of widgeongrass reproduction for all fertilization trials

^awhere treatment concentrations are expressed as lbs./ac. equivalent of ammonium sulfate and treble superphosphate, respectively.

b denotes widgeongrass plants which were greater than 10 weeks old.

greatest average widgeongrass length occurred on fertilized treatments. The specific averages of seed number, weight, and length were 3.7 seeds, 0.1559 g., and 6.8 in.; and 3.2 seeds, 0.092 g., and 6.97 in. for control and fertilized treatments, respectively, which yielded seed.

Seed numbers, weights, and lengths were compared between immature and adult vegetation. Immature vegetation consistently had the lowest average number of seeds, weights, and lengths. Immature widgeongrass averaged 2.3 seeds, 0.1379 g., and 4.8 in. while the adult widgeongrass had an average of 5.0 seeds, 0.9053 g., and 7.8 in.

Setchell (1946) found that widgeongrass produced seeds from 8.5 to 11.5 weeks of age when conditions were favorable. Seeds appeared at 7 weeks of age on immature plants of 6 weeks after fertilization. Mature plants had seeds at 23 weeks of age or at 8 weeks past treatment. Mayer (1967) concluded that the greatest reproduction occurred in 6 week old <u>Ruppia</u> plants rather than 8 or 12 week old plants. He suggested that the production at 8 weeks may have been due to experimental design.

Bernstein and Hayward (1958) suggested that fruit and seed production occurred when vegetative growth was restricted. Immature plants which were subjected to ammonium sulfate concentrations over 5 lbs./ac. failed to produce consistant quantities of seed. Adult plants from the 5 and 100 lbs./ac. concentrations of nitrogen produced seed in the adult plants. Neither immature or adult plants produced seed with treble superphosphate or sewage sludge applications.

Minimal seed production may have been the result of increased osmotic pressure and of the artificial greenhouse environment. Enzymes required to initiate and set seed may have been negatively affected by the osmotic pressure and increased concentrations of certain ions may have affected enzyme systems within the plant.

Muskgrass of the Characese are classified as algae even though the plants are rooted in the mud and silt substrate. The plants reproduce sexually by means of anteridia and cogonia. These two microscopic structures were produced on all stalks, but no attempt was made to classify or enumerate the structures on any treatments. Fertilization had no observable effect on the reproductive ability of the <u>Chara</u> plants except where the plants were killed. Additional work with muskgrass

reproduction would be needed to demonstrate the specific effects of fertilization on anteridia and cogonia.

<u>Growth.</u> Widgeongrass and muskgrass were subjected to fertilizer treatments of ammonium sulfate, treble superphosphate, and sewage sludge. Concentrations of fertilizers were applied on a pound per acre equivalent basis to each treatment. As a means of simplification, the ordered pair notation (x_1, x_2) will designate x_1 lbs./ac. of ammonium sulfate and x_2 lbs./ac. of treble superphosphate. The notation y_1 will indicate y_1 lbs./ac. of sewage sludge. For example, the 50,75 notation would indicate 50 lbs./ac. of ammonium sulfate together with 75 lbs./ac. or treble superphosphate; the 10 treatment would signify 10 lbs./ac. of sewage sludge.

Widgeongrass dry weights were significantly different from treatment to treatment. The F statistic was 3.75 at & = .05 with a tabular value of 1.39. The widgeongrass from the control treatment had the greatest mean weight, 0.3243 g., compared with all fertilized treatments. Widgeongrass weights and lengths are presented in Table 7.

Plants in the ammonium sulfate levels of 5, 10, 20, and 25 lbs./ac. had lower weights than did the control, but these weights were not significantly different from the control. Widgeongrass exposed to dosages of ammonium sulfate above 25 lbs./ac. had significantly lower dry weights. One exception, however, to this generalization occurred in the 100,0 lbs./ac. treatment. The treatment mean was 0.3062 g.; plants from that treatment had an unusually high dry weight because one experiment

Treatment ¹	Weight	Length	Treatment Replications
Control lbs./ac.	.3243 g.	5.80 in.	6
5,0	.1705	4.83	4
10,0	. 2214	6.45	4
20,0	.1822	5.63	4
25,0	.1430	4.85	5
/50,0	0. *	0. *	2
75,0	.0645 *	3.61*	4
100,0	.3062	3.86	9
300,0	0. *	0. *	1
0,5	.1425	4.92	4
0,25	.0165 *	2.54	9
0,50	. 0293 *	3.75	2
0.75	. 0169 *	2.90	5
0.100	. 0255 *	2.81	4
0.300	. 0055 *	2. 22	2
25. 25	0. *	0. *	- 2
25, 75	. 0316*	4.33	4
25,100	. 0148 *	3.19	3
25,300	0006*	94*	2
50,25	0 *	• / •	2
50, 2 5	0450 *	2.56	6
50,100	0036 *	1 72*	2
20,100 /50 300	0 *	1. 12 ¹	2
75 25	0 *	0.*	2
75,25	0. *	0, *	2
75,100	0. *	0. *	2
75,100	0.*	0.*	2
100 25	0.*	0. *	2
100,25	U. ↑ 0027 ÷	U. * 1 20*	
100,75	.0021*	1.04	2
(100,100		U. *	1
100,500	.0010 *	• JJ* 4 - J2	4
5 25	. 2157	0.22	4
25 1.00	. 0012*	5.54	4
200	.0002 *	5.00	4
200	U. *	U. *	<u> </u>
300	. 0978*	4.99	D
400	.0005 *	. 22*	2
500	.0096 *	. 72*	2
700	.0930*	3.97	6
1000	.0695 *	4.28	4

Table 7. Widgeongrass weights and lengths by treatment means

¹where x_1 , x_2 is the lbs./ac/ equivalent of ammonium sulfate and treble superphosphate and where y_1 is lbs./ac. equivalent of sewage sludge. *Significant at & = .05. $P \leq P$ used adult plants. In that experiment, the adult plants of the 100 lbs./ac. of ammonium sulfate produced an increased amount of biomass over the control of that treatment. The dry weight differences found in that experiment are discussed in a later section. Without the influence of the adult plant weights, the treatment mean would have been 0.0587g. This means is significantly lower than the control treatment mean.

Most phosphosphorus treatments had plants which produced dry weights that were significantly below the weight of the control. Plants from the 5 lbs./ac. treatment of phosphorous did not weigh significantly lower than the control weight, and the weight was 0.1405 grams compared to 0.3243 grams of the control. Treatments above the 5 lbs./ac. level were harmful to Ruppia production.

<u>Ruppia</u> plant dry weights were always significantly lower than the control when nitrogen and phosphorous as ammonium sulfate and treble superphosphate were applied together. Unlike nitrogen and phosphorous applied singly, plants from the interaction treatment never approached the control weight. The interaction effects on widgeongrass weight appeared to be most detremental of all fertilizer groups.

Sewage sludge treatments were not as harmful as nitrogen and phosphorous fertilizers to widgeongrass weights. Greater amounts of sludge generally could be applied on a pound per pound basis compared with ammonium sulfate or phosphate with severe weight loss (Table 7).

Ruppia plant lengths were significantly different from mean to

mean for all treatments. The F statistic with & = .05 and a tabular value of 1.39 was 5.2. The length in the control was 5.80 inches.

Plants from nitrogen treatments had significantly lower lengths than the control in the 50 and 300 lbs./ac. treatment, but plants from the 10 lbs./ac. ammonium sulfate treatment had a greater stalk length compared to the control.

<u>Ruppia</u> from ammonium sulfate treatments were less than the control mean, but these lower average lengths were never statistically different. There appeared to be no general trend with widgeongrass lengths as ammonium sulfate concentrations increased.

Plants from phosphorous applications did have decreased lengths. No lengths were greater than or equal to the control. Except for the 25 lbs./ac. treble superphosphate treatment, widgeongrass lengths decreased as treble superphosphate was increased.

Plants from interaction treatments of nitrogen and phosphorous had significantly lower lengths above the 75, 25 lbs./ac. treatment rate of ammonium sulfate and treble superphosphate, respectively, compared to the control. The mean widgeongrass length showed a decreasing trend above the 75, 25 lbs./ac. treatment. Plant length from concentrations in the 25 and 50 lbs./ac. of ammonium sulfate together with varying rates of phosphorous were not consistent. Plants from the treatments had mean lengths which were all lower than the plant length from the control.

Vegetation of sewage sludge treatments had shorter stalk lengths

than did the control except for the 5 lbs./ac. treatment of sewage sludge. At the 5 lbs./ac. rate, the length was 6.82 inches which compares with 5.80 inches of the control; the differences was not significant. Vegetation from concentrations of 200, 400, and 500 lbs./ac. of sludge had significantly shorter lengths than the control, but the vegetation from the 700 and 1000 lbs./ac. rate of sludge was not statistically significant.

The least square difference (LSD) test on widgeongrass weight and length measurements are summarized in Table 7. There were no <u>Ruppia</u> plants from any treatments which had significantly greater weights or lengths over the weight and length of the control vegetation (Figures 3-7).

Muskgrass dry weights were statistically different among treatments, and the F statistical value was 4.8247 with an & = .05. The tabular value was 1.39. No plants from fertilized treatments had statistically better weights than the control weight. The control plants had a treatment mean weight of 0.0776 grams (Table 8).

Muskgrass plants from nitrogen treatments produced the greatest mean weights below the 50 lbs./ac. ammonium sulfate. The weights were not statistically greater than the control. Plants from the ammonium sulfate treatments above the 50 lbs./ac. level had lower weights when compared to the control. However, the weights were not significantly lower than the control weight of 0.0776 grams.

Muskgrass from treble superphosphate concentrations of 5, 25, 75, and 100 lbs./ac. had greater weights than did the control, but these

l Treatment	Weight	Length	Treatment Replications
	~ - - - /		a , and a first a first and a first a first and a first a first and a first a firs
Control lbs./ac. 5,0	.0776 g. .1985	2.02 in. 3.34	4 4
10,0	.1501	2.47	4
20,0	.1991	3.11	4
25,0	.1294	2.36	5
50,0	.0198	1.41	2
75,0	.0484	1.91	4
100,0	.0754	2.90	9
300,0	.0049	. 63	1
0,5	.1225	3.02	4
0,25	.0848	2.95	9
0,50	.0744	2.94	2
0,75	.1438	4.40 *	5
0,100	.1153	2.19	4
0,300	0611	1.34	2
25, 25	.1454	1.10	2
25,75	.0954	2.44	4
25,100	.1007	2.52	3
25,300	.1024	1.69	2
50,25	.0621	1.19	3
50,75	.0964	2.69	6
50,100	.0906	1.44	2
50,300	.0258	.84	2
75,25	. 0216	. 71	2
75.75	.0051	.38	2
75,100	.0223	.94	2
75,300	.0503	1.66	2
100,25	.0075	. 56	1
100,75	.0029	. 31	2
100,100	. 0061	.88	1
100,300	. 0232	. 59	2
5	.1098	2.31	4
25	.1168	2.05	·· 4
100	.1154	3.08	4
200	.0047	.72	2
300	. 0864	2.75	6
400	. 0173	1.13	2
500	.0262	1.47	2
700	.0131	1.00	2
1000	.1077	2.20	4

Table 8. Muskgrass weights and lengths by treatment means

¹ where x_1 , x_2 is lbs./ac. equivalent of ammonium sulfate and treble superphosphate and where y_1 is lbs./ac. equivalent of sewage sludge. *Significant at & = .05. weights were not significant. Plants from treatments of 50 and 300 lbs./ac. of superphosphate had 0.0704 and 0.0611 grams of dry weight whereas the control weighed 0.0776 grams. No indentifiable pattern occurred among the mean treatment weights of Chara.

At the rates of 25, 75; 25, 100; 50. 75; and 50, 100 lbs. /ac. equivalent of ammonium sulfate and treble superphosphate, respectively, the plants had a greater weight than the control. Muskgrass from all treatments above the 50, 100 lbs. /ac. level, including the 25, 300 and 50, 25 lbs. /ac. treatments, had lower weights than the control.

Plants of the sewage sludge treatments did not yield statistically different weights as compared to the control, but the plants of the 5, 25, 100, 300, and 1000 lbs./ac. concentrations of sewage sludge had weights greater than the control.

In two separate ammonium sulfate applications, the plant weights statistically approached the weight of the control. Plants from the control weighed 0.0776 grams while plants from the 5 and 20 lbs./ac. of ammonium sulfate treatments yielded 0.1985 and 0.1991 grams. Statistical tests using the LSD are presented in Table 8.

Lengths of muskgrass were statistically different among treatment mean lengths. The F statistic was 4.5103 with a tabular value of 1.39 at & = .05. Chara plants of the 75 lbs./ac. treble superphosphate treatment had a significantly greater length than the control, 4.40 inches compared with 2.02 inches, respectively.

Muskgrass lengths from nitrogen treatments were greater than

the control for concentrations less than 50 lbs./ac. of ammonium sulfate. Vegetation of ammonium sulfate applications greater than 50 lbs./ac. had lengths which were below the control mean length except for the 100 lbs./ac. treatment. The lengths of the 100 lbs./ac. treatment was 2.90 inches. The 300 lbs./ac. application resulted in a plant length of 0.63 inches which was well below the 2.02 inch control. No plants had lengths statistically significant above or below the control.

<u>Chara</u> plants responded to treatments with decreased lengths from the treatments of 5 to 50 and from 100 to 300 lbs./ac. of treble superphosphate. Plants from the 75 lbs./ac. treatment of superphosphate were an exception to the decreasing mean length trend, and plants from the treatment were significantly longer in lengths than the control, 4.40 and 2.02 inches, respectively. The 5, 25, 50, and 100 lbs./ac. treatment of treble superphosphate had plants which were longer than the control, but the lengths were not statistically significant.

Muskgrass from nitrogen and phosphorous interactions at 25,75; 25,100; and 50,75 lbs./ac. of ammonium sulfate and treble superphosphate, respectively, had 2.44, 2.52, and 2.69 inches of vegetation. Although these growths were greater than the 2.02 inch control length, they were not significantly different from the control. The other treatments had lengths which were below the control.

Plant lengths were greater than the control for five separate sewage sludge treatments. Plants from treatments of 5, 25, 100, 300, and 1000 lbs./ac. of sewage sludge had 2.31, 2.05, 3.08, 2.75, and

2.20 inches of vegetation. No muskgrass vegetation from sludge treatments had significantly better lengths than the control.

<u>Chara</u> mean treatment weights and lengths were evaluated with the LSD mean comparison test (Table 8). There were no plants from any fertilizer treatment that had significantly greater dry weights than the control, but plants of the 75 lbs/.ac. treatment of treble superphosphate had a significantly longer length as compared to the control (Figures 3-7).

Adult <u>Chara</u> plants were in the 75 lbs./ac. treatment of phosphorous. Without the influence of these mature plants, the length was 4.02 compared with the 2.02 length of the control. While plants from the 75 lbs/.ac. treatment had plants which were greater than the control, the difference was not significant without the influence of the adult plants. The adult plants will be discussed more fully in the following section.

<u>Mature Growth.</u> Widgeongrass and muskgrass production were measured as immature plants (less than 10 weeks old) and as mature plants (greater than 10 weeks old). Mature widgeongrass and muskgrass were cultured in a control solution for 15 weeks prior to fertilization; the mature plants were 25 weeks old at harvest.

The mature plant data is presented in Table 9. Widgeongrass weights were significantly better than the control for 20 and 100 lbs./ac. ammonium sulfate. The weights were 0.4624 and 1.5434 grams while the control weight was 0.2369 grams. The plant length at the 100 lbs./ac.

	Widged	ongrass	Muskgr	ass
Treatment	Weight	Length	Weight	Length
control lbs./ac. 5,0	.2369 g. .2671	7.00 in. 5.38	.1736 g. .3399	2.36 in. 2.69
10,0	. 2457	6.13	. 2604	2.00
20,0	.4624 *	9.88	.4279 *	3.44
25,0	.3146	6.63	. 2344	2.63
100,0	1.5434 *	10.25 *	. 0335	5.38
0,5	.1016	5.38	. 2209	2.31
0,25	.0250 *	2.44 *	.0172	2.38
0,75	.0453	4.19	.1235	5.81 *
50,75	.1365	3.49	.0660	1.63
5	.2107	5.56	.1381	1.94
25	.1144	4.88	. 2384	2.25
100	.1027	5.25	.1775	3.81
300	. 2037	7.88	. 2318	2.56
1000 .	. 2421	8.88	. 2391	3.50

Table 9. Adult widgeongrass and muskgrass weights and lengths by treatment means

¹ where x_1 , x_2 denotes lbs/ac. equivalent of ammonium sulfate and treble superphosphate, respectively, while y_1 denotes lbs./ac. equivalent of sewage sludge.

*Significantly different from their respective control at the & = .05 level.

ammonium sulfate level was significantly greater than the control length, 10.25 and 7.00 inches.

The response of widgeongrass to the 100 lbs./ac. ammonium sulfate treatment in the limited adult experiment is observed in the comparison of the combined treatment weight of <u>Ruppia</u> plants. While the weight of plants from the 100 lbs./ac. ammonium sulfate level was not significantly greater than the combined mean control weight, the effect of the mature plants from the 100 lbs./ac. rate accounts for the increased average weight (Tables 7 and 9).

Muskgrass weights were significantly better than the control for the 20 lbs./ac. ammonium sulfate treatment where the mean weight was 0.4279 grams and .1736 grams for the control. Plants from the 75 lbs./ac. treble superphosphate treatment had a significantly better average stalk length over the control, 5.81 and 2.36 inches. The response of muskgrass in the limited adult experiment is observed in the combined comparison of treatment weights and lengths of muskgrass. The length of the adult plants at the 75 lbs./ac. rate were significantly greater than the length of the control (Tables 8 and 9).

Vegetation may have been heavier for the widgeongrass and muskgrass and longer for widgeongrass because of increased plant tolerance to salinity. Immature vegetation was more subject to weight loss and appeared to be less tolerant to osmotic pressure change. Mayer (1967) suggested that immature widgeongrass suffered greater mortality and

weight loss in saline studies. Similar results were reported for sago pondweed by Teeter (1963).

Older widgeongrass and muskgrass plants would be expected to have a greater biomass. Mature plants had additional time to produce vegetation.

<u>Hoagland's Solution</u>. Hoagland's solution is a basic plant nutritive media used by plant physiologists. Hoagland and Arnon (1950) developed the solution which was designed to be a balanced nutrient solution for plant growth. A solution of tap water, half-strength Hoagland medium, and 3000 ppm sodium chloride were added to tanks where four replications of immature and mature widgeongrass and muskgrass were present. Widgeongrass and muskgrass were cultured and monitored as previously described.

Immature widgeongrass and muskgrass in both the non-algicide and the algicide treatments suffered extensive mortality, and, at the end of 10 weeks, all plants were dead. The weights and lengths for each treatment were 0 due to the 100 percent mortality.

The adult widgeongrass and muskgrass in the two treatments were more tolerant to treatment than were the immature widgeongrass and muskgrass. Widgeongrass had 0 percent mortality while muskgrass had 25 percent mortality in the treatment with the algicide. The two species, however, were all dead in the treatment without the algicide,

Adult widgeongrass had a weight of 0.0668 grams, and the adult muskgrass had a weight of 0.0400 grams. Widgeongrass and muskgrass lengths were 5.00 and 1.88 inches, respectively. These mean treatment weights and lengths were for the Hoagland's solution which contained the algicide.

The weights and lengths of <u>Ruppia</u> plants and <u>Chara</u> plants suggest that the plants showed no measurable response to the macro-micro elements in the Hoagland's solution versus the nutrient-free control solution (Table 10).

Potassium, calcium, magnesium, zinc, copper, molybdenum, and iron were present in the Hoagland's solution; these nutrients were not contained in the ammonium sulfate, treble superphosphate, or sewage sludge treatment to any large extent. The submergents, however, did not show positive improvement in weights or lengths as a result of the exposure to the nutrients in the Hoagland's solution.

The results of the Hoagland's solution experiment suggest that widgeongrass and muskgrass were not growth inhibited in the basic ammonium sulfate, treble superphosphate, and sewage sludge treatments by an absence of specific macro-micro elements.

Ion Toxicity and Osmotic Damage. Many plants are intolerant of increasing salt concentrations in their environment. Ammonia, nitrite, nitrate, and phosphate ions may be considered toxic to plant and bacteria populations when present in large concentrations. Wadleigh et al. (1947), Hayward and Spurr (1944), and Robinson (1930) concluded that the effect of certain toxic ions along with osmotic pressure differences caused plant mortality.

A control treatment and a 50, 75 lbs. /ac. treatment of ammonium sulfate and treble superphosphate, respectively, were applied to widgeongrass and muskgrass vegetation and cultured to determine whether toxic amounts of cations or anions were present in the fertilization experiments. Soil, water and plant materials were analyzed after 1, 4, and 10 weeks of exposure to treatment. Procedures and methods used in culturing the two treatments were identical to previous procedures and methods.

Amonia concentrations, as a result of amonium sulfate applications, varied from 2.2 ppm for week 1 to 1.0 ppm for week 10 for the water in the control treatment. The highest reading was 2.2 ppm of ammonia in the control whereas the 50,75 lbs./ac. ammonium sulfate and treble superphosphate, respectively, had the highest amount of ammonia, 4.4 ppm, during week 1. The ammonia decreased from week 1 to week 4, but increased from week 4 to week 10. Ammonia concentrations in the water did not appear to be toxic to widgeongrass or muskgrass because of the minimal amounts which were present.

Duisberg and Buehaer (1954), for example, observed that ammonia concentrations less than 260 ppm did not decrease plant weights. Germination occurred in experiments when the ammonia level was below 450 ppm.

Ammonia and nitrite ions interact under conditions of high pH. Nitrogen loss occurs as a result of volatization, Wahhab and Uddin (1954) indicated that an interaction was present only when high amounts of nitrate and ammonia (10 mg NO₂ and 100 mg $NH_4/100$ g. soil,

		IMMATURE		
	Widgeo	ngrass	Muskg	rass
Treatment	Weight	Length	Weight	Length
Control	.0856 g.	4.09 in.	.2088 g.	2.71 in.
Nitrogen	.0792	3.10	.1048	1.88
Phosphorous	. 0299	2.23	.0700	3,10
N-P Inter.	.0193	1.93	. 9797	3.02
Sludge	. 0475	2.65	. 0625	2.09
Hoagland's + angicide	0.	0.	0.	0.
Hoagland's	0.	0.	0.	0.

Table 10. Average widgeongrass and muskgrass weights and lengthsaccording to treatment group in Hoagland's solution

۸	n	ΤT	τ	TT.
ъ	$\boldsymbol{\nu}$	U	┶	1

Widgeongrass		Muskgrass		Muskgrass	
Weight	Length	Weight	Length		
.2369 b.	. 7.00 in.	.1756 g.	2.38 in.		
. 7269	7.65	. 2592	3.43		
.0573	4.00	.1050	3.50		
.1365	3.44	.0660	1.63		
.1747	6.49	. 2050	2.81		
. 0668	5.00	. 0400	1.88		
0.	0.	0.	0.		
	Widgeo Weight . 2369 b. . 7269 . 0573 . 1365 . 1747 . 0668 0.	Widgeongrass Weight Length . 2369 b. 7.00 in. . 7269 7.65 . 0573 4.00 . 1365 3.44 . 1747 6.49 . 0668 5.00 0. 0.	Widgeongrass Muskg Weight Length Weight . 2369 b. 7.00 in. .1756 g. . 7269 7.65 .2592 . 0573 4.00 .1050 . 1365 3.44 .0660 . 1747 6.49 .2050 . 0668 5.00 .0400 0. 0. 0.		

respectively) were present under desicated soil conditions. Soils were never dried during the present study nor did ammonia concentrations ever reach 100 mg/100 g. soil. A significant reduction of nitrogen through the nitrite and ammonia interaction did not occur.

Nitrite may increase to toxic proportions under alkaline soil conditions as a result of denitrification. The process of denitrification is stimulated by high pH, high ammonia, high temperatures, and analysistic arearobic conditions or waterlogging (Chapman and Liebig, 1952). Chapman and Liebig (1952) found a maximum of 46 ppm nitrite when ammonium sulfate was applied at 871 lbs./ac. Nitrates ranged from 22 to 320 ppm. Nitrite toxicity appeared to decrease with increased nitrates and to increase with increased ammonia (Bingham et al., 1954).

Nitrite concentrations were not measured, but the concentrations of nitrate in mud were below 0.1 ppm during week 1, 4, and 10 for both the control and the 50,75 lbs./ac. treatment of ammonium sulfate and treble superphosphate, respectively, except for week 10 in the control where the nitrate was 0.6 ppm. Low nitrate and ammonia concentrations would indicate that no substantial amounts of nitrites occurred in any treatment under the arearobic waterlogged conditions. Plant mortality was not due to nitrite toxicity.

Nitrates ranged from less than 0.1 ppm to 0.6 ppm in mud, water, and plant material. In the mud, less than 0.1 ppm nitrates were evident in the 50,75 lbs./ac. treatment for week 1, 4, and

10 and in the control for week 1 and 4. During week 10, mud in the 50,75 lbs./ac. treatment had reached 0,6 ppm nitrates.

Nitrate levels in the water were below 0.2 ppm for all weeks in the control and the 50, 75 lbs. /ac. treatment. Analysis for nitrates in the widgeongrass and muskgrass material was limited to the 10th week because plants from week 1 and week 4 did not provide enough vegetative material for testing. Plant material from the control and the 50, 75 lbs. /ac. treatment of ammonium sulfate and treble superphosphate, respectively, produced less than 0.1 ppm nitrate at 10 weeks of age. These rates are well below the nitrate levels which are considered to be toxic. Bain and Chapman (1940) felt that the concentrations of nitrate between 210 and 350 ppm may have been deleterious to avocado plants. Nitrates were considered less harmful to plant growth compared with ammonia in other studies (Viets, 1965).

Phosphates in the mud were different from the control and the 50,75 lbs,/ac. treatment. The mud in the control had 12, 12, and 14 ppm phosphate during week 1, 4, and 10, respectively. The 50, 75 lbs./ac. treatment yielded mud with a similar phosphate response. The values for week 1, 4, and 10 were 29, 29, and 34 ppm. Phosphates in the water solution were below 0.1 ppm for all periods in the control treatment. The values of 1.5, 5.1, and 9.0 ppm phosphate showed an increase in phosphate from week 1 to week 10 at the 50, 75 lbs./ac. level.

Longeragan and Asher (1967) reported that 0.9 percent of

phosphate in the dry weights of lupin and clover was toxic to those plants. The phosphorous percentage in dry matter of yellow lupins was 2.2 and 1.98 percent (Warren and Benzain, 1959). The plants died because of the phosphorous toxicity. Forsberg (1964) reported that <u>Chara</u> plants showed decreased growth at 15-30 µg phosphorous/1.

<u>Ruppia and Chara had 0.29, 1.08, and 2.26 percent phosphorous</u> in dry plant matter at the 50,75 lbs./ac. treatment level at week 1, 4, and 10. These phosphorous percentages approached toxic tolerance limits of other plants. At higher levels of phosphorous use, the tolerance limits were probably exceeded. It appears that the effects of phosphorous accumulation within roots, stems, and leaves of widgeongrass and muskgrass were toxic above the 75 lbs./ac. rate of phosphorous application.

Sulfate and methane gas were normal by-products of organic decomposition in marsh systems. Large concentrations of both substances are normally present, and most aquatic plants appear unaffected by them. Kretchmer et al. (1953) felt that sulfate in the soil had a minimal effect on the absorption of sulfate or other ions into the plant. The problem of sulfate toxicity from the addition of the ammonium sulfate fertilizer seemed remote because the plants were exposed to concentrations of sulfate in the natural environment. Sulfates were not considered toxic in the present experiment.

The ionic concentration of ammonia, nitrite, nitrate, and sulfate were not considered toxic in fertilizer experiments. There was evidence, however, to conclude that phosphorous applications over 75 lbs./ac. might have formed toxic phosphate concentrations. Figures 8 and 9 and Table 13 and 14 in appendix summarize the data. Table 11 gives values found in water samples.

Salinity decreases the ability of plants to obsorb nutrients. Wadleigh and Gauch (1942) found that plants exposed to increasing saline solutions failed to absorb nutrients. Unassimilated nitrogen, soluble organic nitrogen, and protein nitrogen were less in plants from saline solutions as compared with plants from the control. Other experimental results suggested that smaller amounts of phosphate and total nitrogen were assimilated by the plants in saline media versus the plants in the control treatments (Gauch and Wadleigh, 1942). Large masses of calcium, sodium, and potassium were contained in plants from salt treatments, but the plants from the control treatments had smaller amounts of calcium, sodium, and potassium.

Since fertilizers are actually salts, additional fertilizer applications increase the osmotic pressure and salinity of the solution. Other studies have shown that widgeongrass, saga pondweed, hardstem and alkali bulrush, and cattial responded negatively to increased osmotic pressure (Teeter, 1963; Kaushik, 1963; and Mayer, 1967).

The osmotic pressure change in varying saline solutions has been responsible for decreased plant growth (Wadleigh and Gauch, 1942; Hayward and Spurr, 1944; Wadleigh et al., 1947; and Bernstein and Hayward, 1958).





lbs./ac. of ammonium sulfate and treble superphosphate, respectively.



Figure 8. Phosphorous and nitrate values present in soil samples during weeks 1, 4, and 10 for control and 50, 75 lbs./ac. of ammonium sulfate and treble superphosphate, respectively.



Figure 9. Phosphorous and nitrate values present in plant samples during weeks 1, 4, and 10; and week 10, respectively, for control and 50, 75 lbs./ac. of ammonium sulfate and treble superphosphate, respectively.

I.

Treatment	Week	Ammonia	Ph o spho rous	Nitrate
**************************************		ppm	ppm	ppm
	1	2.2	<.1	< .1
Control	4	.8	< .1	. 2
	10	1.0	<.1	< .1
	1	4.4	5.1	< .1
50,75 ^a	4	. 8	1.5	< .1
	10	1.1	9.0	.1

Table 11.	Ammonia,	phosphorous,	and nitrate	concentrations	in	water
	after fertil	ization for two	treatments			

^awhere the 50,75 treatment represents 50 and 75 lbs./ac. of ammonium sulfate and treble superphosphate, respectively.

b weeks after fertilization.

Plastids or mitochandria may fail to adjust to osmotic pressure and cause plant death (Bernstein, 1961). It is possible that if the plants do make the osmotic adjustments within the cells, it is at the expense or the reduction of overall plant growth. Bernstein (1963) also suggested that osmotic pressures may affect enzymes within the plant system. Enzymes affecting reproduction, growth, or maintenance metabolism might be altered by the osmotic pressure. Plant mortality may be the necessary consequence of enzyme alterations.

Algae Bloom

A dramatic change in nearly all fertilized treatments was noted directly after nutrient material was added (Figures 10, 11). Blue green algae developed in the tanks within 48 to 96 hours after treatment. Substantial algal populations were established in 90 percent of the tanks at the end of 168 hours post treatment.

The filamentous bluegreen alga, <u>Oscilatoria</u> sp., occurred at all concentrations of nitrogen, phosphorous, and sewage sludge levels. It occupied two related habitats; the algae had positioned itself at different depths in the tanks or had attached itself to plants, pots, or tank sides.

The algal density was classified into four groups: none, light, medium, and heavy. The low or light density was used when the tops of the pots were visible when the algae was present. The pot bottoms were visible under medium alga concentrations. At the heavy classification, no pots were visible through the water; this density classification was identical to the "pea soup" conditions characteristic of eutrophified lakes.

In order to determine the complete biomass resulting from fertilization, the water from each treatment was strained through filter paper using the millipore filter technique. Algae blooms were collected. Unfortunately, the filtrate contained silt and other soil particles along with the algae. The foreign material would have added weight to the algal sample, and the true total biomass would have been inaccurate.

Algae from other experiments were not collected for the total biomass, and the data from the initial millipore filtering was discarded.

Several methods were incorporated into the experimental runs to decrease or to minimize the concentration of <u>Oscilatoria</u>. A solution of copper sulfate and Captan was added to all experiments except for a pilot experiment. This solution did not significantly reduce the alga. The blooms appeared on tanks regardless of whether the copper sulfate and Captan solution was added.

Aeration together with copper sulfate and Captan solution was applied to the last experiment. The presence of <u>Oscilatoria</u> was not reduced, but the algal blooms on the water surface were less frequent as compared to the non-aerated experiments. The bloom decrease probably was the result of water movement in the tanks. This fragmented the <u>Oscilatoria</u> before it could accumulate into massive concentrations. Table 12 shows the algae concentrations for experiment 1, 2, and 6 where no copper sulfate and Captan were used, where copper solfate and Captan were used, and where copper sulfate and Captan were used with aeration.

The algae present in each tank underwent rapid density changes from week to week. The algal population change showed predictable patterns. Strands of algae developed on the tank bottom and became attached to the displaced soil and to the pot bottoms. Through sexual and asexual division, new growths were produced. The filamentous algae was able to increase its range into the widgeongrass the muskgrass

Treatment	E	xperiment Number	
	1	2	3
Control ^A	$light^B$	$light^C$	$light^{D}$
0,25	medium	light	light
0,75	medium	medium	medium
25,0	e	light	light
50,75	е	light	light
100,0	light	light	light
100,25	heavy	heavy	e
100,75	medium	heavy	е
100,100	light	heavy	е

Table 12. Algae concentrations at the end of treatment periods for threefertilizer experiments on widgeongrass and muskgrass

A where the treatment concentration is expressed as lbs./ac. equivalent of ammonium sulfate and treble superphosphate, respectively.

^Bwhere no copper sulfate and Captan were used.

C where copper sulfate and Captan were used.

^Dwhere copper sulfate and Captan were used with aeration.

^ethe particular concentration of ammonium sulfate and/or treble superphosphate not used in experiment. stalks and leaves. Additional reproduction of the algae was visible on the water surface in the form of an algal bloom.

Effect of Artificial Fertilizers on Invertebrates

Chironomid invertebrate indicators

Chironomid invertebrates were deposited into treatments with specific fertilizer concentrations to ascertain the effect of artificial fertilizers on invertebrates.

Mortality. The midge larvae were held in the same containers as <u>Ruppia</u> and <u>Chara</u> samples. The larvae were introduced to the environment 1 week prior to fertilization. The two plant species were available in an immature and mature stage. Four pots of immature vegetation and four pots of mature vegetation provided a constant source of detritus and living plant material to the chironomids. Widgeongrass and muskgrass plants together with the silt on the container bottom and in the plant pot were used as a substrate for the larvae. Each tank was continuously aerated, and 10 larvae were applied to each treatment. There was only one replication.

The larvae disappeared within 24 hours after their introduction. While some body parts were visible on the silt surface or in the vegetation, most of the larvae had apparently burrowed into the silt. The absence of chironomids from treatments was the result of either death or maturity. Dead midges may have settled in the silt at the bottom of the tank. Decaying chironomids were subject to evisceration, and dismembered larvae could have been confused with other debris. Although all debris and water was filtered through a screen with a diameter of approximately 0.25 cm., no dead midges were observed on the surface or the bottom of the tanks, or on the debris.

Some chironomids did not survive the trip from the collection site to the laboratory. The freshly killed invertabrates floated to the surface of the holding tanks. The red cases remained on the surface for nearly 1 week before the material was discarded. If the artificially induced larvae had died in the treatment tanks, their red cases should have been visible in the tanks.

Oldroyd (1965) determined that cannibalism was common in midge populations. Viable and inviable larvae may have been prayed upon by individuals within the same population. The presence or absence of individuals at the examination time may have been related to a preypredator relationship rather than the result of fertilization.

The invertebrates were present in the larval stage throughout the entire study. Jamnback (1954) suggested that the life cycle of midges including a 4 week larval stage. Chironomids remained in a 12 week larval stage in other studies (Fellton, 1940). Usinger (1956) concluded that the larval stages varied from specie to specie in the Chironomidae, and the exact amount of time needed for larvae development was unknown.

The absence of midges in the treatments during the sampling period can be attributable to death, cannibalism, or maturity. The

term mortality as used here will refer to the absence of midges from specific treatments regardless of the cause of disappearance.

Visable chironomid larvae were present in 6 of 18 treatments at the end of the 10 week study. Dead or partially dead larvae were never present in any treatment at any time. Only the live larvae were present at the tenth week. Sixty percent or more of the larvae in each treatment were not present at the end of the study. Chironomids from the saltonly treatment had the lowest mortality rate of 60 percent. Larvae in the control had 90 percent mortality. In two other treatments, algicide and plain water, 100 percent of the individuals died. Larvae in fertilized treatments had mortality rates between 80 and 100 percent. The lowest rate, 80 percent, occurred in the 75 lbs./ac. treble superphosphate treatment.

<u>Growth.</u> The greatest midge weight was 0.0032 g. in the 20 lbs./ac. ammonium sulfate rate, and the lowest midge weight was 0.0001 g. in the 5 lbs./ac. sludge treatment. There appeared to be no relationship between average larvae weight and the concentration of ammonium sulfate or treble superphosphate fertilizer.

The treatments were grouped into nitrogen, phosphorous, sewage sludge, and control experiments. The average weights of midges in the nitrogen, phosphorous, sludge, and control were 0.0018, 0.0043, 0.0001, and 0.0033 grams; 4.00, 6.67, 2.00, and 12.60 percent of the total chironomids were present in the nitrogen, phosphorous, sludge, and control treatments at the end of 10 weeks.
Chironamids had a higher survival rate when provided with no fertilizer (Figure 12 and Table 15 in Appendix). Provost (1958) suggested that chironamids should occur in greatest numbers when exposed to inorganic nutrients. The fertilizers appeared to be toxic to the chironomids larvae, but further experimentation with more replication should be initiated.

Scatella aquatic flies

During the course of the experiments, minute insects were found on the water surface. These winged insects seemed particularly abundant on those treatments which had surfaced algal blooms, but some insects appeared on treatments which never did have substantial blooms. The winged invertebrates were true flies in the family Ephydridae.

The <u>Scatella</u> sp. were found in all three phases of development: larvae, pupa, and adult. The larva and pupa cases floated on the water surface, and large rafts of these cases were found on the ends and the sides of all containers. The adults seemed to skim on the water surface and then took to the air when disturbed. The adults spent a majority of their time on the islands of algae where they rested and fed (Figure 13).

The <u>Scatella</u> flies were not artificially introduced into the experiments. Since the <u>Scatella</u> are algae consumers, they are not dependent on submergent and emergent aquatic plant growths for food. Fluctuating populations of <u>Ruppia</u> and <u>Chara</u> did not appear to influence the number of aquatic flies.



Figure 12. Dry weight for chironomid larvae according to fertilizer group and specific treatment, respectively.

0,25

5

Salt

Control

Grams

Weight

20,0

25,0

67

Treatment

The addition of fertilizer promoted substantial crops of bluegreen algae. The <u>Scatella</u> invertebrates were most abundant on treatments where the <u>Oscilatoria</u> was present; fertilization increased the presence of the bluegreen algae habitat for the Scatella flies.

Management Implications for Fertilizer Use

Ammonium sulfate, treble superphosphate, and sewage sludge fertilizers under the present experimental levels of fertilizers and salinity should not be applied to marsh impoundments. This conclusion is based upon results from fertilization tests in the controlled atmosphere of the greenhouse with the Logan tap water, 3000 ppm sodium chloride, and algicide-fungicide solution. <u>Ruppia</u> and <u>Chara</u> lengths and weights are not significantly increased after artificial fertilization with the experimental fertilizers and salinity levels used in this study.

Habitat manipulations

Peat has been suggested as a source of energy for plant growth (McKnight, 1969). Past experiments with peat as an organic matter source for cereal grains failed at Fish Springs. Although the cereal grains failed to increase their growth after peat application, submergent and emergent plants might respond positively to peat. Both plant forms appear to be doing well on the Avocat Unit of Fish Springs NWR where natural peat deposits are present.

Portions of many managed state and federal refuges in the west

are non-productive waterfowl and aquatic shorebird areas. Black (1972) evaluated a long term furrowing project on Bear River Migratory Bird Refuge near Brigham City, Utah. Furrows were dug on the salt flat, and the soil was mounded on one side of the trench. Older furrows had more vegetation and yielded greater numbers of waterfowl and aquatic shorebird nests (Black, 1972). Since the furrows at Bear River have provided habitat for game and non-game species where no habitat previously existed, further experiments on other marshes appear justified.

Nelson (1955) reported that sewage and other organic material helped to produce excellent growths of emergent marsh plants. Research with emergent vegetation should be conducted with organic nutrients; a combination experiment using the artificial environment of the greenhouse and using the natural environment of spring-fed marshes would provide additional insight on the effects of fertilizers on marsh vegetation.

SUMMARY

Ammonium sulfate, treble superphosphate, and dried sewage sludge fertilizers were added as dried material to the growth media of widgeongrass (<u>Ruppia meritima</u>) and muskgrass (<u>Chara sp.</u>). The plants were raised artificially from seed and sporangia collected from Fish Springs National Wildlife Refuge in western Utah. Soil was also collected from the refuge.

Wideongrass and muskgrass were rooted in pots of marsh soil, and the pots were in an aqueous solution in the tanks. The solution was Logan tap water plus 3000 ppm sodium chloride and an algicide-fungicide inhibitor. The aqueous solution, however, was not chemically identical to the water found at the refuge.

Six full replications were completed with the growth media and different levels of fertilizers. Four plants were used for each pot in all experiments, and each treatment had four replications. Treatments were arranged in a completely randomized manner. Experiments were conducted under an artificial environment in the greenhouse.

Chironomid invertebrates were introduced in a pilot project. Invertebrates were under the same environmental conditions as were the widgeongrass and muskgrass. Since the chironomids were exposed to a limited number of fertilizer trials with one replication, minimal amounts of tolerance and growth information were obtained.

The following salient points were drawn from the study:

1. Widgeongrass plants weighed less than the control plants in all fertilized treatments. The weight of <u>Ruppia</u> plants ranged from 0.0005 g. at the 5 lbs./ac. equivalent of sewage to 0.3243 g. at the control rate. Plants from the following levels were significantly lower than the control: 50, 75, and 300 lbs./ac. equivalent of ammonium sulfate; 25, 50, 75, 100, and 300 lbs./ac. equivalent of treble superphosphate; and 25, 100, 200, 300, 400, 500, 700, and 1000 lbs./ac. equivalent of sewage sludge. <u>Ruppia</u> from the levels of 25, 50, 75, and 100 lbs./ac. equivalent of ammonium sulfate together with the level of 25, 75, 100, and 300 lbs./ac. equivalent of treble superphosphate had significantly lower weights compared to the control.

2. Widgeongrass plant lengths from fertilized treatments were smaller than the control for most treatments. Plants from ammonium sulfate applications of 50, 75, and 300 lbs./ac. equivalent, sewage sludge applications of 200, 400, and 500 lbs./ac. equivalent, and ammonium sulfate-treble superphosphate applications of 25, 25; 25, 300; 50, 25; 50, 100; 50, 300; 75, 25; 75, 75; 75, 100; 75, 300; 100, 25; 100, 75; 100, 100; and 100, 300 lbs./ac. equivalent, respectively, had significantly smaller lengths than the control. However, plants with treatments with 10 lbs./ac. equivalent of ammonium sulfate and 5 lbs./ac. equivalent of sewage sludge produced greater lengths than the control. The lengths of vegetation were 6.45, 6.22, and 5.80 inches, respectively.

3. Muskgrass plant weights ranged from 0.0029 g. at 100 lbs./ac. equivalent ammonium sulfate with 75 lbs./ac. equivalent of treble superphosphate to 0.1991 g. at 20 lbs./ac. equivalent of ammonium sulfate. No plants had weights which were significantly above or below the control weight of 0.0776 grams.

4. Muskgrass plants generally had smaller lengths than the control as fertilizer concentrations increased. However, <u>Chara</u> from the treatment of 75 lbs./ac. equivalent of treble superphosphate had a significantly greater length than the control, 4.40 and 2.02 inches. The difference was significant.

5. Widgeongrass and muskgrass plants were cultured in a solution of Logan tap water, 3000 ppm sodium chloride, and half-strength Hoagland's solution. Weights and lengths of both plants exposed to the Hoagland's solution were well below the weights and lengths of both plants exposed to the control. The results suggest that widgeongrass and muskgrass were not growth inhibited by an absence of specific macro-micro elements in the ammonium sulfate, treble superphosphate, and sewage sludge fertilizer experiments.

6. The survival rate of chironomid larvae invertebrates in fertilized and unfertilized treatments was low. Chironamids in the control had the highest survival rate, 12.6 percent, but larvae from the treble superphosphate treatments had the greatest weights, 0.0043 g.

7. Bluegreen algae, <u>Oscilatoria</u> sp., appeared in all treatments within 24 hours after fertilizer enrichment. Algal populations

reproduced, bloomed, died, and decayed throughout the period of study. Flies, <u>Scatella</u> sp., from the family Ephydridae appeared on the algal blooms.

8. Additional nutrient experiments should be initiated using water from the study location together with the soil from the same site. This would reduce the effect of the artificial chemical bias of the water found in this study. With the water and soil coming directly from the study site, the nutrient experiments with invertebrates, submergents, and emergents would better approximate the actual on-site conditions.

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APPENDIX

Week ^b	Phosphorous ppm	Nitrate ppm
1	14	<.1
4	12	<.1
10	12	.6
1	29	<.1
4	29	<.1
10	34	<.1
	Week ^b 1 4 10 1 1 4 10	Week ^b Phosphorous ppm 1 14 4 12 10 12 1 29 4 29 10 34

Table 13. Phosphorous and nitrate values present in mud samples for two fertilizer treatments

^awhere 50,75 treatment denotes 50 and 75 lbs./ac. equivalent of ammonium sulfate and treble superphosphate, respectively.

bweeks after fertilization.

Treatment	Week ^b	Phosphorous %	Nitrate ppm
	1	.19	с
Control	4	. 37	с
	10	. 31	<.1

	1	. 29	с
50,75 ^a	4	1.08	с
	10	2.26	<.1

Table 14. Phosphorous and nitrate values present in plant samples for two fertilizer treatments

^awhere 50,75 treatment represents lbs./ac. equivalent of ammonium sulfate and treble superphosphate, respectively.

^bweeks after fertilization.

^c no samples were evaluated.

Fertilizer Group		
Treatment	Dry Weight	
Control	.0033 g.	
Nitrogen	. 0018	
Phosphorous	.0043	
Sewage Sludge	. 0001	

Table 15.	Dry weights for chironomid larvae according to fertilizer
	group and specific fertilizer treatment

_ 1	Specific Fertilizer Treatment	
Treatment	Dry Weight	
Control lbs. /ac.	.0004 g.	
Salt	. 0015	
5,0	0.	
10,0	0.	
20,0	.0032	
25,0	.0004	
100,0	0.	
0,5	0.	
0,25	.0022	
0,75	0.	
5	0.	
25	0.	
100	0.	
300	0.	
1000	0.	

where x_1 , x_2 denotes lbs./ac. equivalent of ammonium sulfate and treble superphosphate, respectively, and where y_1 denotes lbs./ac. equivalent of sewage sludge.

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