

AN ABSTRACT OF THE THESIS OF

Alex Grant Ogg, Jr. for the M.S. in Farm Crops
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Title THE REPRODUCTION AND MODE OF SPREAD OF SAGO PONDWEED
(POTAMOGETON PECTINATUS L.) IN IRRIGATION SYSTEMS

Abstract approved


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Nine experiments were conducted during 1964 and 1965 to study the methods of reproduction and mode of spread of sago pondweed (Potamogeton pectinatus) in irrigation systems.

Frequent treatments of sago pondweed foliage with aromatic solvent greatly reduced the number of tubers in the substratum of an irrigation canal. However, complete eradication of all tubers in one irrigation season was not obtained.

Ecological investigations revealed that sago pondweed was capable of invading and becoming established in new irrigation channels in one season. The propagules responsible for the establishment of new infestations were tubers and plant fragments. Considerable numbers of sago pondweed seed were transported into the new channels by the water, but no seedlings were found. Sago pondweed tubers survived freezing temperatures under field conditions. Subsequent studies in the laboratory showed that sago pondweed tubers could become somewhat hardened or conditioned to withstand freezing temperatures.

Removal of the parent tuber did not affect the survival of

emerged shoots nor did it prevent the growth of excised sprout tips. The main portion of the tuber appeared to have a function other than initiating growth. The study of shoot emergence from tubers indicated a relationship between the weight of the tuber and the depth from which the shoot would emerge. The main portion of the tuber, whose dry weight consists primarily of carbohydrates, may function as a food reserve enabling the shoot to emerge from a considerable depth in the soil.

Sago pondweed seeds that had overwintered in a canal bottom gave 15 percent germination in a greenhouse aquarium after a 50-day period. Seeds that had passed through the digestive tracts of wild-ducks germinated abundantly. However, if the seeds were held in the digestive tract for more than three days, no germination occurred. The storage of sago pondweed seeds at different conditions of temperature and moisture for different periods of time resulted in significant differences in the percentages of germination. Storage of seed in water at 0.0 degrees C. for 12 days resulted in the highest percentage of germination. The seed germination studies indicated that the importance of seed in the spread of sago pondweed in irrigation systems may be underestimated.

THE REPRODUCTION AND MODE OF SPREAD OF
SAGO PONDWEED (POTAMOGETON PECTINATUS L.)
IN IRRIGATION SYSTEMS

by

ALEX GRANT OGG, JR.

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APPROVED:

[REDACTED]

Assistant Professor of Department of Farm Crops

In Charge of Major

[REDACTED]

Head of Department of Farm Crops

[REDACTED]

Dean of Graduate School

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THE REPRODUCTION AND MODE OF SPREAD OF
SAGO PONDWEED (POTAMOGETON PECTINATUS L.)
IN IRRIGATION SYSTEMS

INTRODUCTION

Sago pondweed (Potamogeton pectinatus L.) is a submersed, rooted, perennial, aquatic seed plant. It is one of the most important waterfowl food plants on the North American continent and, therefore, is usually considered a desirable plant in the lakes and streams of waterfowl refuges. On the other hand, sago pondweed is one of the most troublesome weeds in many of the municipal, industrial, recreational, and agricultural waterways of the United States. Its dense, rank growth usually must be controlled in such channels, particularly in those which deliver irrigation water to croplands in the arid portions of the West.

Various mechanical and chemical methods are used to control sago pondweed in irrigation systems. Chaining, dredging, drying, and hand-cleaning are some of the mechanical methods of control. Aromatic solvents, acrolein, and a few other herbicides are used to control sago pondweed chemically. Current mechanical and chemical methods of control have one or more disadvantages. Among these are high cost, toxicity to game fish and other desirable aquatic biota, accumulation of organic decomposition products, and consequent accumulation of plant debris.

Field and laboratory studies on the factors affecting the reproduction of sago pondweed by seed and tubers and field studies on the rate and methods of spread of sago pondweed in irrigation

channels were conducted. A more thorough knowledge of the methods of reproduction and the mode of spread of sago pondweed should aid materially in the development of more satisfactory control measures.

LITERATURE REVIEW

Description of Sago Pondweed

Fassett (1940), Mason (1957), and Muenscher (1944) describe sago pondweed (Potamogeton pectinatus L.) as a wholly submersed, rooted, aquatic plant. The filiformis leaves are alternately placed on stems that are up to two meters long. The stems arise from much branched rhizomes that are tipped by fleshy tubers. The leaves are all submerged, 3-15 cm. long, attenuate, two mm or less in width, and typically one-nerved. The stipules are 15-25 mm long and united to the middle. The flowers of sago pondweed are perfect, and the spikes are conspicuously interrupted by four to six unequally remote whorls of flowers.

Yeo (1965) gives an excellent description of sago pondweed flower and fruit development,

The inflorescence, or spike, was formed on a peduncle at the apex of the leafy shoot. The florets on the immature spike were compressed and enclosed by two leaves. As the florets developed, the peduncle and rachis elongated. The mature floret consisted of four 1-celled pistils with sessile stigmas that extended slightly above four large 2-locular anthers. The surface of the stigma was orange-yellow. Four large sepal-like connective tissues extended under the anthers and protected the florets during early stages of development. An examination of 50 spikes showed that there was an average of 11 florets on each. The pollen was oblong and translucent, with white caps at each end, and buoyant.

Pollination, fertilization, and development of the fruits took place at the water surface. After pollination, the fertilized ovaries swelled. Usually 2 of the 4 pistils developed. The mature fruit, or drupelet, consisted of a white seed with a seed coat, leathery endocarp and mesocarp, and fleshy exocarp.

Fernald (1932) lists P. pectinatus with P. filiformis and P. vaginatus under the water-pollinated subgenus Colegeton. Other workers, such as Arber (1920), Chrysler (1907), and Fassett (1940), state that the spikes of P. pectinatus raise above the surface of the water, permitting wind pollination. Wind pollination in flowing water of irrigation channels would be unlikely to occur. It would seem more probable that the pollen is carried by the water.

At maturity, the floral rachis sinks beneath the surface of the water and the fruits are stripped from the rachis by the force of the current, by floating masses of algae, or by other floating debris. Sago pondweed fruits are approximately two and one-half mm wide and four mm long, are obliquely ovoid, and have a short, wart-like beak. A hinged section, often called a trapdoor, covers an opening on the dorsal side of the fruit. The trapdoor is hinged at the end nearest to the style base and is forced open when the embryo germinates. There are approximately 100,000 sago pondweed fruits per pound (Muenscher, 1944). In the course of this paper, the fruits of sago pondweed will be referred to as seeds, even though it is realized that the actual seed is only a part of the fruit.

Fernald (1932) found that P. pectinatus had highly developed methods of vegetative reproduction, regardless of production of seeds. He pointed out that vegetative propagation was mostly by subterranean tubers, axillary buds, and creeping rhizomes. The axillary buds are located between the stipules and the stem at each node (Chrysler, 1907).

Two forms of sago pondweed grow in the irrigation channels of

eastern Washington. One is robust, has coarse stems, and has wide, acutely-pointed leaves. The other is less robust and has narrow acuminate leaves. Oborn et al. (1954) found that the robust form of sago pondweed was more resistant to herbicides than the slender form.

Gortner (1934) analyzed sago pondweed and found that it was higher in percentage of ash and crude protein, but considerably lower in crude fiber than alfalfa hay. Harper (1939) found that the average nitrogen, phosphorus, and calcium content was higher in P. pectinatus than in common weeds and grasses.

Distribution of Sago Pondweed

The distribution of sago pondweed is world wide. In 1907, Ascherson and Graebner observed that sago pondweed ranged from sea level in England to above 5,000 meters in Venezuela and Tibet (Arber, 1920). Muenscher (1944) reported that P. pectinatus was found in all of six lakes that were located in different regions of the United States. Martin and Uhler (1939) reported that sago pondweed was found in all of the continental states of the United States and in most of the provinces of Canada. Furthermore, these authors stated that sago pondweed was probably the most important waterfowl food plant on the North American continent and represented at least half of the total food value of the genus Potamogeton. Guppy (1894) found that waterfowl may carry the seeds and seed-like fruits of some aquatic plants unharmed in their stomachs. The consumption of propagules of this plant by waterfowl has undoubtedly assisted in

its distribution.

Environmental Factors Affecting the
Distribution and Adaptation of Sago Pondweed

Conditions that favor growth of hydrophytes are an uninterrupted water supply, abundant carbon dioxide, and protection against sudden changes in temperature (Arber, 1920). Blackman and Smith (1911) studied the nature of the relationship between assimilation and the chief environmental conditions of light intensity, temperature, and carbon dioxide supply. They found in every case that the rate of assimilation was governed by one or another of these environmental conditions acting as a limiting factor.

Light Intensity

Pondweeds grow in water a few inches to several feet deep (Martin and Uhler, 1939). These authors reported that in water of average clarity, six to eight feet was considered the maximum depth for successful growth, but in exceptionally clear water, certain species had been found growing at 20 feet or more. Sago pondweed will grow to a depth of 18 feet in large irrigation channels in central Washington.

Arber (1920) states that reflection from the water surface, absorption by the water, and diffusion by certain substances in solution or solid particles in suspension, reduce the amount of light that reaches a submerged shoot. Hutchinson (1957) states that one meter of pure water will absorb approximately 37 percent of the red light, 24 percent of the orange light, seven percent of the

yellow light, two percent of the green light and violet light, and less than one percent of the blue light. Moreover, suspended particles and dissolved material in natural waters will absorb most strongly the blue and violet light.

Bourn (1932) found that P. pectinatus required four percent (300-400 foot-candles) of total radiant energy for growth and development. Hodgson and Otto (1963) grew sago pondweed for four weeks under 25 to 400 foot-candles of light and found progressively more growth at the higher light intensities. Plant weight, branch number, and stolon development increased while shoot length and shoot to root ratio decreased with increasing light. Furthermore, shoot and stolon development were greater under red light than under blue light.

Hydrogen Ion Concentration (pH) and Carbon Dioxide

Moyle (1943) found that Potamogeton pectinatus was a typical hard water plant and was frequently the only inhabitant of calcareous river silts. He stated that P. pectinatus would grow in water that had an alkalinity greater than 150 parts per million, a sulfate ion concentration greater than 50 parts per million, and a hydrogen ion concentration (pH) of 8.4 to 9.2. Moyle also noted that Potamogeton species were more abundant and robust in hard waters than in soft waters.

A great many flowering plants, such as Elodea canadensis, Ceratophyllum demersum, and Myriophyllum, are commonly found in alkaline water (Everson, 1928) because the quantity of assimilable

carbon is much higher in alkaline than in acid water. The concentration of HCO_3 ions, one of the principle carbon sources, decreases with decreasing pH.

Bourn (1932) found that the growth of Naias flexilis, Potamogeton perfoliatus, and Potamogeton foliosus was increased by increasing the concentration of dissolved carbon dioxide from eight to 72 parts per million. However, increasing the concentration of dissolved oxygen retarded the growth of these same species.

Other Factors

Bourn (1932) found that increasing the salt concentration up to 28 percent sea water stimulated the growth of P. pectinatus. At Swanquarter, North Carolina, plants in good health grew in water with a salt content equaling about 44 percent of normal sea water (Martin and Uhler, 1939). Metcalf (1931) observed sago pondweed growing in water with a three and one-half percent salt content. Hodgson and Otto (1963) found that increasing the temperature from 60 to 80 degrees F. under 400 foot-candles of light increased the dry weight of sago pondweed threefold. Bruns (1961-64) stated that the tubers of sago pondweed began growth in early April when the water temperature was 45 degrees F.

Factors Affecting the Reproduction of Sago Pondweed Tubers

Formation and Distribution of Tubers

Subterranean tubers formed at the tips of the branches of the

rhizomes, are the primary method by which sago pondweed overwinters, particularly in irrigation channels that are dry during the winter months (Otto et al. 1958; Yeo, 1965). Yeo (1965) found that tubers developed from specialized nodal and internodal tissues at the ends of branches. Bruns (1961-64) stated that new tubers were produced at the ends of rhizomes eight weeks after emergence of the first shoot from the parent tuber.

Yeo (1965) investigated the numbers and size of tubers in the bottom of an irrigation canal. He found 267 tubers in a volume of soil 18 inches square and 18 inches deep (5,832 cubic inches). Most of the small tubers were found near the surface of the soil, whereas the larger tubers were found at all depths down to 18 inches below the surface. Bruns (1961-64) and Bruns and Comes (1965) found that the number of sago pondweed tubers in the bottoms of heavily infested irrigation channels may be as high as 400 to 600 per cubic foot of soil. In one case 1,580 tubers were found in a volume of soil 12 inches square and 40 inches deep (5,760 cubic inches). The author has found tubers, that were firm and apparently viable, as deep as 57 inches in areas of heavy silt deposition.

Yeo (1965) planted tubers in soil at depths of 2, 6, 12, 18, and 24 inches. All tubers emerged from depths down to 12 inches, but only one tuber emerged from 18 inches and none emerged from 24 inches. He also recorded the number of tubers formed each month. The number of tubers formed increased through July and then decreased slightly during August and rapidly during September. A total of 2,380 tubers were produced by the growth from one tuber in a

six-month period.

Tuber Weight and Carbohydrates

Otto et al. (1958) found that the average dry weight of sago pondweed tubers was about 28 percent of the fresh weight. Hodgson (1963) stated that the average dry weight of sago pondweed tubers having a fresh weight of about two-thirds gram was 19.5 percent, but that the percentage dry weight increased with tuber size. Furthermore, the total carbohydrate content of the tubers was found to be about 74 percent of the dry weight. Hodgson found that the percentage of carbohydrates also increased as tuber size or fresh weight increased and that the major increase in the carbohydrate content was as starch.

The carbohydrate supplies of the tubers were largely depleted during the first 10 to 14 days following planting. The percentage of dry weight of the tubers decreased from 19.5 to 3.5 percent after 30 days growth, whereas the fresh weight decreased only 15 percent during the same period. It was noted that tuber structure remained intact and that the cells of the tuber were filled with water after 18 days.

In about 10 days, the losses in dry weight of the tuber were balanced by dry weight increases in other organs of the newly formed plant and the net weight of the plant began to increase.

Initial Shoots from Tubers

Initial shoots from tubers planted in greenhouse aquaria

possessed only two subterranean nodes (Bruns 1961-64). The first node developed one to one and one-half inches from the tuber. The second node developed at or near the soil surface regardless of the depth at which the tuber was planted. The first aerial shoots and the rhizomes always developed from the second node.

Hodgson (1966) found that the main shoot began to export material to other parts of the plant at about 16 days, which indicated that the shoot was self-sustaining. He further found that the whole plant, less the tuber, doubled in dry weight each four and one-half days.

Effects of Drying and Freezing on Survival of Tubers

Otto et al. (1958) studied the effects of drying on the survival of tubers. These authors found the viability of air-dried tubers remained quite constant for a period of four to five days, but the tubers did not survive air drying beyond nine days. Martin and Uhler (1939) state that only one out of eight tubers that were dried for a month at ordinary room temperatures retained enough vitality to grow.

Winter time air temperatures in many of the Northwest states often drop many degrees below freezing and the ground may be frozen to depths of several inches or even several feet. Otto et al. (1958) observed that tubers of sago pondweed do not survive temperatures of 0 to -20 degrees F., but tubers did survive at 32 degrees F. Yeo (1965) stated that tubers exposed to temperatures of 32 degrees F. and lower were killed. The author has observed that tubers

frozen in canal soil germinate and grow when placed in greenhouse aquaria. This information, coupled with the fact that canal soil does freeze during the winter, indicate that there is some mechanism operating in field conditions which protects the tuber and enables it to withstand freezing conditions.

Reducing Tuber Production

Yeo (1965) studied the influence of defoliation on the development of tubers. He found that removing foliage reduced their numbers and that removing the foliage twice was more effective than removing it once. The greatest reduction in the number of tubers produced occurred when the foliage was removed twice in August. A period of four weeks elapsed between the dates the foliage was removed.

Otto, Bartley and Garstka (1963) investigated the effect of repeated treatments with aromatic solvents on the production of tubers. Their study indicated that treatment of the foliage with aromatic solvents significantly reduced the number and size of tubers. Two treatments reduced the number more than one treatment, but a third treatment did not reduce numbers further. In all tests, regrowth apparently developed only from rhizome meristematic tissue, and not from tubers that had never been vernalized.

Germination of Sago Pondweed Seeds

Seed Viability

Records of the occurrence of Potamogeton seedlings in nature

appear to be rare. As reported by Muenscher (1936), Maguire and Wann found large numbers of Potamogeton pectinatus seedlings growing on a mucky river bottom. Seeds washed upon the shore in early spring were dried out and non-viable, but those still in the water germinated in abundance in water cultures in the laboratory. McAttee (1917) stated in his discussion of the propagation of wild-duck foods, that P. pectinatus could be grown easily from seeds. In 1894, Sauvageau obtained seedlings of P. pectinatus from seeds which had been in laboratory cultures less than one year (Muenscher, 1936). Moore (1915) succeeded in germinating a small percentage of overwintered seeds of P. pectinatus. Fischer, in 1907, found that seeds of P. pectinatus and other aquatics could be kept for several years in culture without germinating, but if fermentation was allowed to set in, germination soon began (Muenscher, 1936). He concluded that seeds of aquatic plants possess dormant embryos and a chemical stimulus was necessary to bring about germination.

Crocker (1907) obtained considerable germination of P. pectinatus seeds when the seeds were gathered green and put into distilled water at 23 degrees C. However, if seeds were allowed to ripen in the cold water of ponds and then placed in water at 23 to 29 degrees C., they would not germinate. When ripe seeds were sterilized and the seed coats ruptured, 51 percent germination was obtained. Crocker pointed out that in the seeds of P. pectinatus the limiting or excluding of water or oxygen by the seed coat (pericarp) may have more to do with inhibition of germination than the dormancy of the embryo, as assumed by Fischer. Muenscher (1936) found that P.

pectinatus seeds stored dry or in water at room temperature did not germinate, while those stored in water at one to three degrees C. for two to six months gave 17 to 38 percent germination. The results of Muenscher's work indicated that the apparent scarcity of viable seeds and seedlings in certain species of Potamogeton was not due to the inability of these species to produce viable seeds. Yeo (1965) obtained 100 percent germination of sago pondweed seeds when the trapdoor was opened and the embryo exposed.

Arber (1920) and Crocker (1938) state that the seeds of many water plants are dormant when harvested, but a few months in water just above freezing often ripens the seeds and breaks the dormancy. These findings were consistent with those of Muenscher (1936).

Delayed Germination

Guppy (1897) found that the seeds of most aquatic plants had a considerable delay in germination, and in most species the germination of a single crop was distributed over two or more years. Arber (1920), in discussing the delayed germination of aquatics, stated that the sprouting of seed may be deferred until the third, fourth, or fifth year. From the studies by several investigators on the subject of delayed germination, Arber concluded that the delay in germination occurred only if the seeds were continuously immersed in water. The seeds of many aquatics are protected both by pericarp and testa, which may be of value in enabling them to resist the rotting effect of prolonged submergence (Arber, 1920).

Effects of Drying on Germination

Arber (1920) states that seeds of many water plants germinate promptly after being subjected to a period of drying. The reason for increased germination following preliminary drying may possibly be that drying gives rise to some cracking of the seed coats (Arber, 1920).

Guppy (1897) seems to have been the first to investigate the effect of drying on the viability of Potamogeton seeds. He concluded that the seeds of most species of this genus retain their power of germination for several months after drying. Yeo (1965) found that storing sago pondweed seeds dry at 36 degrees F. for three months resulted in 29 percent germination, while of those overwintering on a canal bottom, 14 percent germinated. Germination decreased when seeds were stored wet at 36 degrees F. for three months, or either wet or dry at 70 degrees F. for three months. Furthermore, the frequency of alternate wetting and drying did not influence germination. Chapman (1964) found that one lot of sago pondweed seed was nine percent viable, although it had been stored dry for ten years. The viability of another lot of seeds that had been stored dry for two years ranged from 86 to 91 percent over four tests.

Other Factors Affecting Germination

Guppy (1897) found that all of the seeds taken from the stomachs of wild ducks germinated in a short time. He obtained 60 percent germination of Potamogeton natans seeds which had passed

through the digestive system of a domestic duck, whereas only one percent of seeds which were held in the water from which the duck had fed, germinated. Arber (1920) stated that a speeding-up of germination occurred if the seeds of aquatic plants had passed through the alimentary canal of a bird. Arber concluded that this could be the result of some disintegrating chemical or mechanical action exerted on the seed coats. Contradictory to the findings of Guppy and Arber, Chapman (1964) found that passage through the alimentary canal of wild ducks greatly retarded the germination of sago pondweed seeds.

Guppy (1897) stated that fish similarly prepared the seeds for germination. He stated that Darwin, in his "Origin of Species" and other authors, had noted the occurrence of Potamogeton seeds in the stomachs of fresh-water fish.

Arber (1920) found that freezing may also assist germination by its effect on the outer covering of the seed. However, Guppy (1897) did not find an increase in germination when seeds of Potamogeton species were frozen in ice or mud. He did find that darkness favored the germination of Potamogeton natans. Yeo (1965) found that swirling P. pectinatus seeds with abrasive sand for 30 minutes gave zero percent germination, while 60 minutes gave 14 percent germination.

Anatomical and Morphological Adaptations of Submersed Aquatics

The important anatomical and morphological adaptations of plants to the submersed aquatic environment are the reduction of

protecting, supporting, and conducting tissues and the development of air chambers the full length of the plant, providing for aeration and buoyancy (Oborn et al. 1954; Eames and MacDaniels, 1947; Weaver and Clements, 1938).

Stems and Leaves

The structure and function of the epidermis of the stems and leaves of submersed aquatic plants differ greatly from those of plants in an aerial habitat. In such aquatics, the epidermis is not protective, but absorbs gases and nutrients directly from the water. The epidermal layer of the typical aquatic plant has an extremely thin cuticle, and the thin cellulose walls permit ready absorption from the surrounding water (Eames and MacDaniels, 1947). The epidermis of many submersed aquatic plants contain chloroplasts and may form a considerable part of the photosynthetic tissue, especially when the leaves are very thin (Schenck, 1886 and Sauvageau, 1891).

The leaves of many submersed aquatic plants are greatly reduced in size and thickness (Costantin, 1886). Furthermore, some are finely divided so that a relatively large area of leaf surface is in contact with the water (Eames and MacDaniels, 1947). Such species as Potamogeton trichoides and P. pectinatus have very narrow submersed leaves which are linear in form and tender and translucent in texture.

The axes of submersed aquatic plants need not support much weight because their branches and leaves contain gas-filled inter-cellular spaces and are buoyed in water (Arber, 1920). The stems of

submersed aquatic plants are usually long and slender and have a poorly developed vascular region (Weaver and Clements, 1938). The mechanical need for developing vascular bundles to secure rigidity no longer exists, and consequently, the cortex occupies a much larger area in proportion to the stele. This arrangement increases the photosynthetic activity of the plant and compensates for the reduced light under water.

The leaves and stems of most species of submersed rooted aquatic plants are rapidly desiccated when exposed to air. The rapid desiccation is due to the lack or deficiency of cuticle or protective tissue, to the adaptation of root structures for anchorage rather than absorption of water, and to the lack of sufficient xylem conducting tissue to move water rapidly into the stems and leaves (Oborn et al., 1954).

Chrysler (1907) found that in the internodes of P. pectinatus, the xylem was represented only by a central cavity surrounded by parenchyma and phloem. A division of the stele into three areas was more or less evident at the nodes.

Roots

The roots of submersed aquatic plants are greatly reduced in size, poorly or not at all branched, and in some genera (e.g. Ceratophyllum) the roots have entirely disappeared. In submersed aquatic plants the function of root anchorage is of greater importance than the function of absorption (Oborn, et al., 1954; Arber, 1920). Arber (1920) states that plasticity is a marked feature of

aquatic plants.

Aerating System

Chambers and passages filled with gases are common in the leaves and stems of submersed plants (Arber, 1920; Eames and MacDaniels, 1947). Gas chambers are large, usually regular, intercellular spaces extending through the leaf and often for long distances through the stems. The gas chambers not only give buoyancy to the organs in which they occur, but also provide an internal atmosphere for the plant. In these spaces the oxygen given off in photosynthesis is apparently stored and used again in respiration, and the carbon dioxide from respiration is held and used in photosynthesis (Eames and MacDaniels, 1947).

STUDIES ON THE REDUCTION OF SAGO
PONDWEED STANDS AND TUBER PRODUCTION

The persistence of sago pondweed tubers in canal substratum is of prime importance in developing satisfactory measures for control of this troublesome aquatic weed in irrigation systems. For example, if it could be shown that sago pondweed tubers survive for only one year after they are produced, then extensive chemical treatments for one season to prevent new tubers from being formed might be a practical method of control

Methods and Materials. The Prosser West Branch Canal, Sunnyside Valley Irrigation District, located in the Yakima Valley in south central Washington, was selected to determine the effect of different frequencies of chemical treatments on the reduction of sago pondweed tuber numbers and foliage.

The Prosser West Branch Canal was approximately 1.5 meters wide and .3 meters deep. The rate of flow was seldom more than 1.5 or 2.0 cubic meters per second and the velocity was approximately 1.0 kilometers per hour.

Sample plots in the canal were the full width of the channel and 4.5 meters long. Buffer zones between plots were 6 meters long.

On March 17, 1965, counts of tubers in soil samples 30.5 cm² x 30.5 cm deep taken by 7.6-cm increments, were made from three locations within each plot by washing the soil through a series of screens. The plots were then covered with eight-mesh nylon screens (Figure 1) to prevent unattached, drifting tubers from lodging in the plots



Figure 1. Placing the eight-mesh nylon screen in the bottom of the Prosser West Branch Canal.

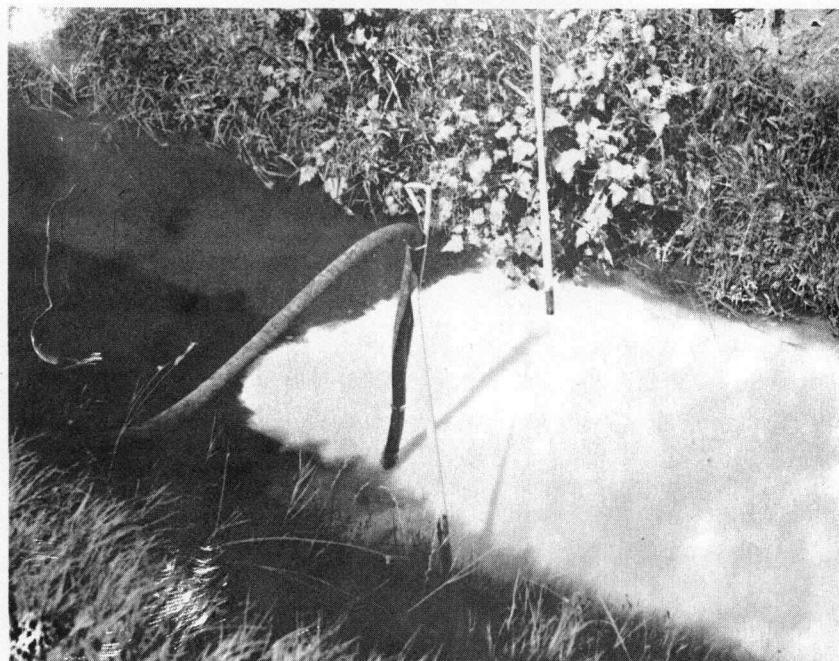


Figure 2. Introduction of aromatic solvents into Prosser West Branch Canal.

and interfering with the experiment.

The Prosser West Branch Canal was treated as follows:

- (1) One plot, located downstream from all other plots, was treated with aromatic solvent every week.
- (2) A second plot, located immediately upstream from the first plot, was treated with aromatic solvent every two weeks. Therefore, the solvent that treated the second plot also treated the first plot.
- (3) A third plot, located immediately above the second plot, was treated with aromatic solvent every four weeks. Thus, when the third plot was treated, the second and first plots were treated by the same solvent.

Aromatic solvent was introduced at the rate of 100 liters per cubic meter per second (cms) in the Prosser West Branch Canal over periods of 29 to 45 minutes. Aromatic solvent is a mixture of cyclic hydrocarbons of either petroleum or coal-tar origin. Therefore, many different mixtures are available and they are sold as aquatic herbicides under various trade names. Specifications on the flash points, distillation ranges, and specific gravities of two widely used aromatic solvents are given by Comes et al. (1963). Aromatic solvent was selected for use in this experiment for several reasons. First, it effectively killed aquatic vegetation within a few hours after treatment and second, it was easy to handle and was readily available from the irrigation project. Because aromatic solvent is insoluble in water, an emulsifier was added at the rate

of one and one-half percent to form a good emulsion of the solvent throughout the water in the canal. A high-volume pump was used to make the introduction of the herbicide. Water from the canal and aromatic solvent from a holding tank were drawn into the pump bell through dual suction hoses, mixed thoroughly, and discharged under pressure into the stream. The plots in the Prosser West Branch Canal were close together and therefore an even dispersion of the aromatic solvent throughout the width and depth of the canal was very important. An even dispersion (Figure 2) was obtained by attaching to the end of the discharge hose a 1.5-meter length of 19 millimeter pipe that had 3.2 millimeter holes drilled in it every 15 centimeters. The pipe was placed across the canal, under the water. This method and equipment was used in all of the treatments.

Sunnyside Valley Irrigation District personnel first treated the Prosser West Branch Canal on May 17, at which time some shoots were 89 cm long (Appendix Table 1). This was somewhat later than desired. The density of growth, maximum length of growth, and condition or vigor of plant growth were determined before each treatment. Visual observation of the density and vigor of the growth was made and the maximum length of the growth was determined by actual measurements. Water temperature and turbidity were recorded before each treatment.

After the first season of treatments, tuber counts were again made in the Prosser West Branch Canal by taking soil samples 30.5 cm x 30.5 cm deep by 10-cm increments from three locations within each

plot and washing them through a series of screens (Figures 3 and 4).

Results and Discussion. Weekly observations of the plots are recorded in Appendix Tables 2, 3, and 4. The regeneration that occurred between treatments during May, June, and July on the plot treated every week was sparse, but reached 12- to 25-cm lengths. After August 1, however, little, if any, new growth occurred. On the plot treated every two weeks, regeneration between treatments was moderate to sparse and attained maximum lengths of 3 to 30 cm until early September. No regrowth occurred after September 7. No new growth occurred after September 20 on the plot treated every four weeks. Previous to that time, regrowth between treatments was mostly moderate to dense and reached maximum lengths of 8 to 64 cm. Much of the new growth between treatments originated from the surface or subsurface nodes of surviving stems and from rhizomes, even in the plot treated every week.

Before treatments, tubers were most abundant in the middle of the channel and in the top 8 cm of substratum (Table 1). After treatments, tuber populations were reduced 53 to 87 percent. Few, if any, tubers were found at depths of from 10 to 30.5 cm at the end of the season. The percent reduction of tubers in the one-week and two-week plots was nearly the same, and considerably higher than the 53 percent reduction of tubers in the plot treated every four weeks. Although the number of tubers in the one- and two-week plots was greatly reduced, there were still sufficient tubers remaining to develop a vigorous stand of sago pondweed during the coming year.



Figure 3. Taking soil sample from plot in Prosser West Branch Canal for determination of the population of tubers.

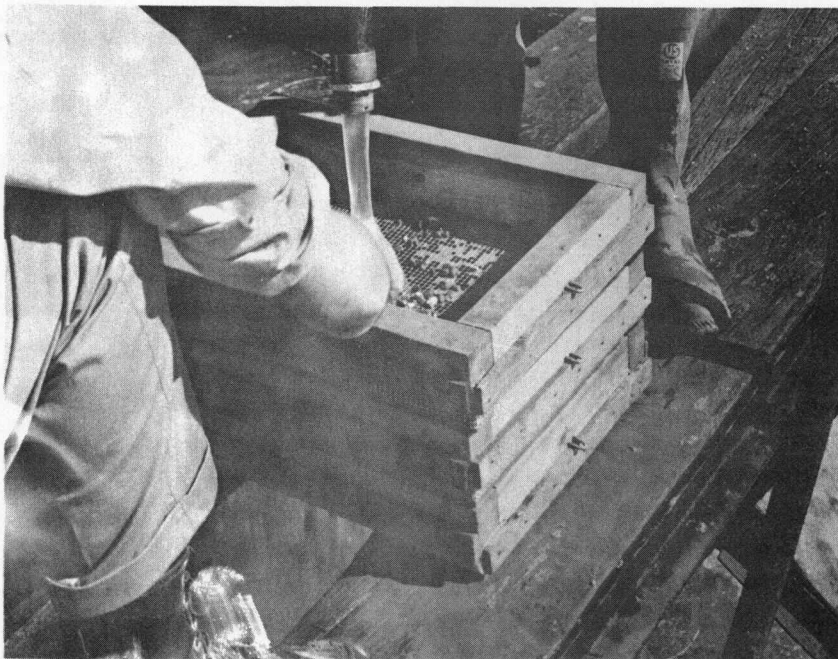


Figure 4. Washing soil samples through a series of screens to separate tubers from soil.

Table 1. Tubers per soil sample 30.5 cm² x 30.5 cm deep in plots before and after treatment with aromatic solvent in the Prosser West Branch Canal during 1965.

Sample location in canal	Samples collected Mar. 17, 1965					Samples collected Nov. 16, 1965			
	Sampling depth (cm)					Sampling depth (cm)			
	0-8	8-15	15-23	23-30.5	Total	0-10	10-20	20-30.5	Total
1-week treatment									
South side	18	4	0	0	22	3	0	0	3
Middle	132	6	3	0	141	24	0	0	24
North side	28	1	0	0	29	16	0	0	16
Total	178	11	3	0	192	43	0	0	43
2-week treatment									
South side	64	16	2	0	82	5	1	0	6
Middle	190	5	4	2	201	24	1	0	25
North side	18	0	0	2	20	8	0	0	8
Total	272	21	6	4	303	37	2	0	39
4-week treatment									
South side	61	8	1	0	70	22	1	0	23
Middle	83	25	3	6	117	57	0	0	57
North side	7	1	0	0	8	11	0	0	11
Total	151	34	4	6	195	90	1	0	91

Aromatic solvent is a contact herbicide that kills the top-growth but not the rhizomes below the soil surface. Sago pondweed tubers are developed at the ends of the rhizomes. Therefore, the possibility exists that some new tubers were formed during 1965, even though topgrowth was usually killed by the periodic solvent treatments. Evidence for this possibility was provided by the fact that nearly all of the tubers present at the end of the season were in the top 10-cm of substratum. In addition, more tubers were present in the four-week plot than in the one- or two-week plots.

One season of frequent treatments with aromatic solvent will greatly reduce the numbers of sago pondweed tubers in irrigation canals. However, complete eradication of the tubers was not obtained. The tubers were either carried over in the substratum for more than one year, or the rhizomes continued to form new tubers even though the topgrowth was removed periodically. Therefore, frequent (one- or two-week intervals) treatments with aromatic solvent would not be practical for control or eradication of sago pondweed in irrigation systems. The four-week treatment would be much more practical from an economic standpoint and would give satisfactory control. After several years of treatments every four weeks, there might be sufficient reduction in the number of tubers to allow control with only one or possibly two treatments per year.

ECOLOGICAL STUDIES IN NEW IRRIGATION CHANNELS

Laterals WB5-G and WB10

Methods and Materials. A study was initiated in 1964 to determine the mode and rate of infestation of sago pondweed in several channels on the Columbia Basin Project in central Washington.

Laterals WB5-G and WB10, located on the Wahluke Slope south of Othello, Washington, were selected for preliminary observations in 1964. Both channels were about 2.5 meters wide. The water in Lateral WB5-G was approximately .5 meters deep and the velocity was 1.6 kilometers per hour. The water in Lateral WB10 was 1.0 meter deep, but the velocity was only 0.8 kilometers per hour. Lateral WB5-G had been receiving water for approximately five years and was heavily infested with sago pondweed. Lateral WB5-G was included in this study for the purpose of determining the changes occurring as a canal matures and is infested with sago pondweed. Lateral WB10 was a newly constructed channel scheduled to begin regular water deliveries in the spring of 1964. This channel had been tested with intermittent flows of water during 1963. The Wahluke Branch Canal, the source of water for Laterals WB5-G and WB10, was heavily infested with Richardson's (Potamogeton richardsonii), horned (Zanichellia palustris), leafy (Potamogeton foliosus), and sago pondweeds.

A site 15.2 meters long and the full width of the channel, was selected in each channel for detailed observations. The appearance of new species, changes in species dominance, time of flowering,

time of seed formation and other observed changes were recorded. Soil samples from the top 15 cm of soil in each site were taken for mechanical analysis and pH determination. Water temperatures were recorded by thermographs throughout the season. Laterals WB5-G and WB10 were treated periodically with aromatic solvent or acrolein by the Bureau of Reclamation.

Results and Discussion. The soil in both channels was loam with a pH of 7.9 in Lateral WB5-G and 8.8 in Lateral WB10. Moyle (1943) found that sago pondweed would grow vigorously at pH values in the range found in these two channels. These data indicate that the soils in both channels were similar and should be capable of supporting an infestation of sago pondweed.

In Lateral WB5-G, a dense stand of sago pondweed emerged from the soil in late April and early May (Appendix Table 5). Although numerous horned pondweed seedlings were observed in May, sago pondweed was the only species observed after July. Flower formation and flowering of sago pondweed occurred in early June and by mid-July seed set was well advanced.

On May 23, a dense stand of horned pondweed and the first sago pondweed plants were observed in Lateral WB10 (Appendix Table 6). The periodic treatments with aromatic solvent and acrolein had killed all of the topgrowth by early August. However, by the end of the irrigation season, a sparse stand of Richardson's pondweed was established in the observation site. No sago pondweed plants were found in the observation site, however, 3 to 5 kilometers upstream, a dense stand with a maximum length of 91 cm was observed.

The water temperature in Lateral WB5-G averaged 17.2 degrees C. and seldom varied more than one or two degrees C. daily. In Lateral WB10, the water temperature averaged 18.3 degrees C. and usually varied five or six degrees C. daily. Lower water velocity and greater distance from the main canal probably accounted for most of the variance in the water temperature at the site in Lateral WB10. The water temperatures were similar enough so that a comparison of plant growth between the two channels could be made.

The preliminary study indicated that sago pondweed could invade and become established in irrigation laterals in one year's time. As in new terrestrial communities, annuals, such as horned pondweed, were the first to become established, but soon gave way to the more persistent perennials such as Richardson's and sago pondweeds. The aquatic herbicide treatments may shorten the time required for this change.

Eltopia Branch Canal

Methods and Materials. The lower end of the Eltopia Branch Canal, located in the southernmost portion of the Columbia Basin Project in south central Washington, was selected for further ecological studies. It was a newly constructed canal and scheduled to receive test water in the late summer of 1964. The canal was approximately 5 meters wide and the water was 1.2 meters deep with a velocity of approximately 0.8 kilometers per hour. The upper end of the Eltopia Branch Canal had been constructed several years previously and had a dense infestation of horned, leafy, and sago pondweeds.

Two sites were selected for detailed observations. One site was in the main channel approximately 0.2 kilometers below the end of the lined section. The second site was located in Lateral EB15. This lateral had many checks and drops which were preceded by areas of slow moving water. It was above the first check and drop that the second site was established. The water in Lateral EB15 was approximately 1.8 meters wide and 0.6 meters deep with a velocity of approximately 0.2 kilometers per hour.

A soil sample was taken from the upper 15 cm of soil in the first site for mechanical analysis and pH determination. Thermographs were used to record the water temperature in both the main channel and Lateral EB15.

The water in the Eltopia Branch Canal was sampled continuously between September 25 and November 3, 1964, for pondweed propagules by means of elongated screen bags (Figure 5). Screen bags were made by removing both ends of a five-gallon (18.9 liters) bucket and attaching a 1.8-meter bag made from ten-mesh nylon screen to one end of the bucket. A rope was attached to the bucket by means of an eye-bolt and the entire screen was suspended in the water from a bridge (Figure 6). One screen was positioned at the surface of the water and the other was placed on the bottom of the channel. The screens were cleaned once a week and the number of sago pondweed seed and tubers in the screens were recorded. Such sampling was continuous throughout the 1965 irrigation season. The Eltopia Branch Canal was treated periodically in 1965 with aromatic solvent or acrolein by the Bureau of Reclamation.

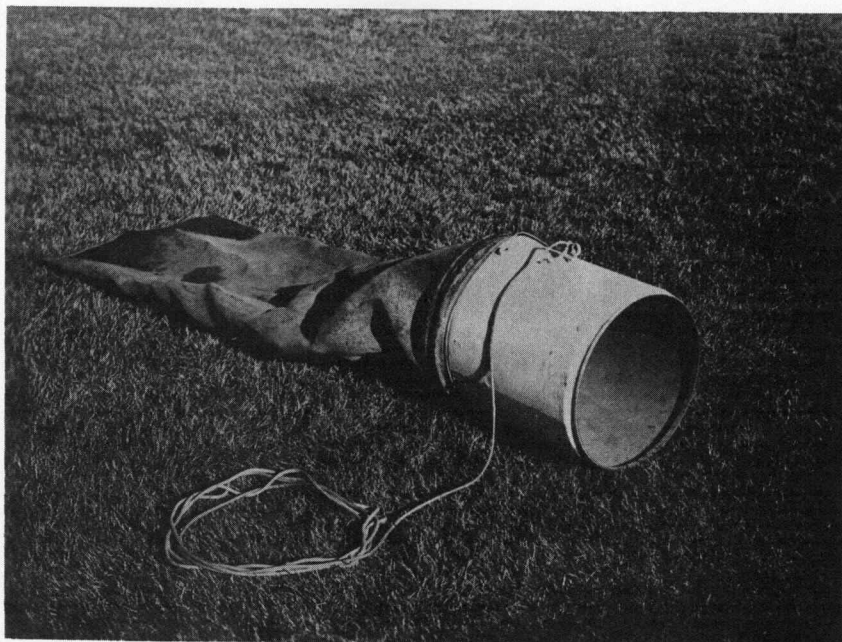


Figure 5. Screen bag used to collect tubers and seed in the Eltopia Branch Canal.

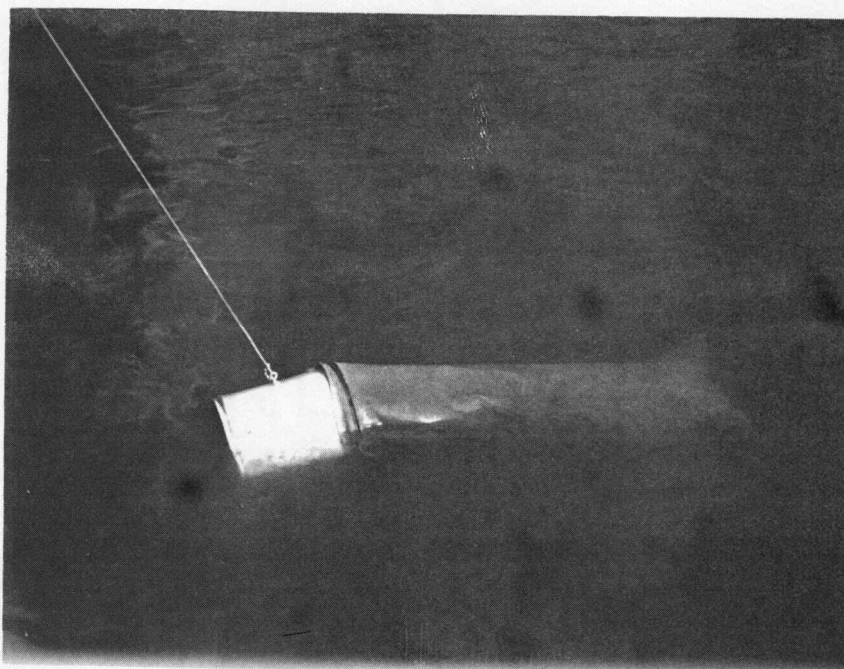


Figure 6. Surface screen in operation in the Eltopia Branch Canal.

Results and Discussion. The soil in the Eltopia Branch Canal was classified as a silty clay loam and had a pH of 8.4. The water temperature averaged 19.4 degrees C. and seldom varied more than one or two degrees C. daily. All of these values are favorable for active growth of sago pondweed.

In the main channel, infestations of horned and leafy pondweeds were first noted about mid-May (Appendix Table 7). The stand increased rapidly, particularly the horned pondweed, until the aromatic solvent and acrolein treatments were applied in June and July. By mid-August the stands of horned and leafy pondweeds were rather sparse, and scattered plants of sago pondweed were first noted.

These observations agreed with those obtained in Lateral WB10 in 1964. That is, sago pondweed was capable of becoming established in a new irrigation channel within one year. The same general plant succession occurred in the same length of time. The annual pondweeds, horned and leafy, appeared first, followed by the perennial, sago pondweed. At the end of the irrigation season, only sago pondweed remained.

No seed or tubers were collected in the screens from September 25 to November 3 in 1964. In 1965, the transport of sago pondweed seeds was greatest from mid-May to the latter part of June (Table 2). This period corresponded with a sharp increase in the flow of water in the canal. In large canals, such as the Eltopia Branch, seed production usually is most abundant in the shallower waters along the edges and on silt berms or bars. Therefore, higher water tends to dislodge the seeds, which had been produced in such areas the

Table 2. Numbers of sago pondweed seed and tubers collected in screens, Eltopia Branch Canal, 1965.

Date	Water temp. ($^{\circ}$ C.)	Surface screen		Bottom screen	
		Seed	Tubers	Seed	Tubers
3-30-65	--	0	0	0	0
4-6-65	--	4	1	0	0
4-13-65	--	10	0	0	0
4-20-65	13.3	0	0	1	0
4-27-65	17.2	0	0	0	0
5-4-65	15.0	0	0	0	0
5-11-65	17.8	3	0	0	0
5-18-65	16.1	77	5	13	1
5-25-65	16.7	1	0	2	0
6-2-65	20.6	0	0	2	0
6-9-65 ^{a/}	22.2	--	--	--	--
6-16-65 ^{a/}	19.4	--	--	--	--
6-22-65 ^{b/}	21.1	13	0	61	0
6-29-65	20.6	8	0	4	0
7-7-65	23.9	8	0	0	0
7-13-65	22.2	3	0	15	0
7-20-65	22.2	7	0	0	0
7-27-65	22.8	5	1	1	0
8-3-65	23.9	7	1	0	0
8-11-65	25.0	3	3	0	0
8-25-65 ^{c/}	--	8	0	--	--
8-31-65	--	1	0	--	--
9-8-65	20.0	0	0	--	--
9-21-65	17.2	0	0	--	--
9-28-65	16.7	0	0	--	--
10-5-65	16.7	0	0	--	--
10-14-65	15.6	0	0	--	--
10-19-65	13.9	0	0	--	--
Total		158	11	99	1

^{a/}Data was lost.

^{b/}Numerous horned pondweed seed.

^{c/}Samples for 2-week period. Bottom screen deteriorated beyond repair and could not be replaced.

previous year and transport them downstream. Oddly enough, few seeds were collected early or late in the irrigation season. More seeds were collected in the surface screen than in the screen on the bottom. This was expected, since the seeds of sago pondweed float for some time before becoming waterlogged and sinking. The importance of seed in the spread of sago pondweed in irrigation systems may be underestimated. There is considerable movement of seed during the irrigation season and these seeds may be capable of starting new infestations.

Few sago pondweed tubers were collected in the screen bags. However, the screen on the bottom of the canal, where drifting tubers were most likely to be present, often became clogged with silt and sediment thereby restricting water movement through the screen and inhibiting the entrance of tubers. Unattached, drifting tubers in considerable numbers have been observed moving along the bottoms of other canals infested with sago pondweed (Bruns and Comes, 1965). Therefore, the movement of tubers of sago pondweed from the upper to the lower end of the Eltopia Branch Canal was believed to occur near the bottom. Although surface screens functioned properly, only 11 tubers were collected during the irrigation season.

A better method of sampling the water near the bottom of the canal must be designed before reliable statements can be made regarding tuber movement and the importance of this propagule in the spread of sago pondweed in irrigation systems.

In Lateral EB15, the water temperature averaged 20.6 degrees C.

and usually varied three to six degrees C. daily. The low volume and velocity of water in Lateral EB15 undoubtedly accounted for much of the variation. However, temperatures were still favorable for the growth of sago pondweed.

Horned and leafy pondweeds and water plantain (Alisma gramineum) began to appear about mid-July (Appendix Table 8). Shortly thereafter, root formation and new leaf growth was observed on fragments of sago pondweed that had been transported by the water. These fragments appeared to be taking root and becoming established plants. It was believed that these fragments had broken loose from partially killed plants in the upper end of the Eltopia Branch Canal and had floated down the canal to Lateral EB15. The density of the horned and leafy pondweed increased markedly during the summer. However, the acrolein treatments applied by the Bureau of Reclamation in August and September killed many of the horned and leafy pondweeds and at the end of the irrigation season only scattered stands of horned and sago pondweeds were found.

Again the succession of plants followed the same pattern as in Lateral WB10 and the Eltopia Branch Canal. It was apparent from the observations of root formation and new leaf growth on transported plant fragments that this was another mode of spread of sago pondweed in irrigation systems.

SURVIVAL OF SAGO PONDWEED TUBERS
UNDER FIELD CONDITIONS

Methods and Materials. Several authors have reported that sago pondweed tubers are killed by freezing temperatures under laboratory conditions (Otto et al., 1958 and Yeo, 1965). However, little information is available on the temperatures at various depths in canal bottoms and the effect of freezing temperatures on sago pondweed tubers under field conditions. Preliminary observations indicate that sago pondweed tubers can withstand freezing in a natural environment.

A soil temperature study in the bottom of the Roza Main Canal, located a few miles north of Prosser, Washington, was initiated on November 15, 1965. Six thermographs were placed on the north side of the channel in an area with a cover of sago pondweed debris. One thermograph recorded air temperature about 15 cm above the soil surface. The other five thermographs recorded soil temperatures at 0.0, 2.5, 7.6, 15.2, and 30.5 cm depths perpendicular to the surface of the soil. A record of the precipitation and snow cover was obtained throughout the test period.

On November 15, 1965, a soil sample 30.5 cm square and 30.5 cm deep was removed by 10.2-cm layers to determine the vertical distribution of the population of sago pondweed tubers in the area where the soil temperatures were recorded. On February 6, 1966, another soil sample of the same size was removed in layers of 0.0 to 2.5, 2.5 to 7.6, 7.6 to 15.2, and 15.2 to 30.5 cm and screened for tubers to be used in a viability test. Seven tubers from the 0.0 to 2.5 cm

depth, nine from the 2.5 to 7.6 cm depth, ten from the 7.6 to 15.2 cm depth, and ten from the 15.2 to 30.5 cm depth were planted in shallow soil in a greenhouse aquarium to test for viability.

Results and Discussion. The weekly minimum and maximum temperatures for the different depths are given in Table 3. The lowest air temperature was -7.2 degrees C. and the maximum depth of freezing was between 7.6 and 15.2 cm. In this study, maximum soil temperatures decreased to a depth of 7.6 cm and then began to increase. There was very little fluctuation in the soil temperatures at the 30.5 cm depth.

Except for a five-day period from December 24 through December 28, 1965, when the maximum snow depth was 10.2 cm, the area was relatively free of snow cover. Total precipitation during the test period was approximately 4.1 cm.

The soil sample taken in November contained sago pondweed tubers that were distributed throughout the sample. The population of sago pondweed tubers and vertical distribution were as follows:

0.0 to 10.2 cm	17
10.2 to 20.3 cm	21
<u>20.3 to 30.5 cm</u>	<u>15</u>
Total	53

The number of tubers at each of the sampling depths in February, 1966, was as follows:

0.0 to 2.5 cm	1
2.5 to 7.6 cm	9
7.6 to 15.2 cm	13
<u>15.2 to 30.5 cm</u>	<u>13</u>
Total	36

Table 3. Weekly minimum and maximum temperatures in the Roza Main Canal. (°C)

Date	Air		0.0 cm		2.5 cm		7.6 cm		15.2 cm		30.5 cm	
	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.	Min.	Max.
11-22-65	2.8	20.0	4.4	15.6	4.4	12.2	4.4	10.0	6.1	5.6	9.4	10.6
11-29-65	-2.8	12.2	0.0	8.9	0.0	6.1	1.1	6.1	4.4	7.2	7.2	9.4
12-6-65	0.6	11.1	0.6	6.1	0.6	4.4	1.7	5.0	3.9	5.6	6.1	6.7
12-13-65	-3.3	13.9	0.6	2.2	0.0	7.8	1.1	5.6	3.9	6.1	6.7	7.2
12-20-65	-6.7	13.9	--	--	-1.1	2.8	-0.6	2.2	1.7	3.9	4.4	6.7
12-27-65	-7.2	10.0	-5.0	2.8	-1.7	-0.6	-0.6	-0.6	1.7	1.7	3.9	3.9
1-3-66	-5.0	11.7	-2.8	4.4	-1.1	0.0	-1.1	-0.6	1.1	1.7	3.9	3.9
1-10-66	-2.8	10.6	-2.2	8.3	-1.1	5.6	-1.1	3.3	1.7	3.9	3.3	4.4
1-17-66	-2.8	13.3	-2.8	11.7	-0.6	7.8	0.6	4.4	2.8	5.0	3.9	5.0
1-24-66	-6.1	8.3	-3.9	8.3	-1.1	2.8	0.0	2.2	2.2	3.3	3.9	5.6
1-31-66	-4.4	5.6	-2.8	2.2	-1.1	-0.6	-0.6	0.0	1.7	2.2	3.3	3.9
2-7-66	-3.9	14.4	-2.2	11.7	-0.6	6.7	0.0	3.3	2.2	3.9	3.3	3.9

In addition, six more tubers were collected from the 0.0 to 2.5 cm depth in the immediate area of the thermographs and included in the viability test. It was believed that these additional tubers would aid in giving a more valid test of the viability of the tubers from this depth.

Fifty-seven percent of the tubers from the 0.0 to 2.5 cm depth, 56 percent from the 2.5 to 7.6 cm depth, and 80 percent from the 7.6 to 30.5 cm depths germinated and developed plants.

In this study not all of the tubers exposed to freezing temperatures were killed. However, the viability of surviving tubers was lowered from 80 to approximately 56 percent. Obviously, there is a mechanism that provides some protection for tubers in a natural environment. It is possible that tubers undergo hardening in the field as the temperature gradually lowers in the fall. The mildness of the winter prevented the evaluation of the survival of sago pondweed tubers at subzero temperatures.

STUDY OF TUBER HARDENING

Methods and Materials. A laboratory test was initiated in the fall of 1965 to determine if sago pondweed tubers could be conditioned to survive freezing by gradually lowering the temperature over a period of several weeks. Information gained in this test may help explain how tubers can survive freezing under field conditions.

Sago pondweed tubers, weighing approximately 0.5 g apiece, were collected from the bottom of an irrigation canal in mid-October,

1965. Each treatment, performed in triplicate, consisted of three tubers per replicate placed in approximately one-half liter of moist, canal-bottom soil, contained in quart ice cream cartons lined with plastic bags.

In an attempt to simulate the temperature conditions that may occur in the field, the tubers were stored for periods and temperatures as follows:

Treatment number	Storage periods and temperatures (°C)							
	6 wks.	1 wk.	1 wk.	1 wk.	2 wks.	1 wk.	1 wk.	1 wk.
1	7.2							
2	7.2	0.6						
3	7.2	0.6	-1.1					
4	7.2	0.6	-1.1	-3.3				
5 ^a /	7.2	0.6	-1.1	-3.3	-3.3			
6	7.2	0.6	-1.1	-3.3	-3.3	-6.7		
7	7.2	0.6	-1.1	-3.3	-3.3	-6.7	-11.1	
8	7.2	0.6	-1.1	-3.3	-3.3	-6.7	-11.1	13.3

^a/Tubers were held at -3.3°C for an additional two weeks to arrange refrigeration at lower temperatures.

At the end of each period, the appropriate cartons were removed from cold storage and transferred to greenhouse aquaria for germination studies. As a control, the viability of nine tubers from the original supply that had been stored at 7.2 degrees C for four months, was tested.

Results and Discussion. All of the tubers held for six weeks at 7.2 degrees C germinated (Table 4). The gradual reduction in the temperature over a five-week period to -6.7 degrees C resulted in a gradual reduction in the percentage of survival from 100 to 44 percent. No tubers survived at -6.7 degrees C or below. All tubers

held for four months at 7.2 degrees C germinated. Therefore, the differences in the percentage of survival of the treated tubers was real and resulted from the lower temperatures. This study showed that sago pondweed tubers can become hardened or conditioned to freezing temperatures to some degree in the laboratory. However, it is felt certain that sago pondweed tubers exposed in the field to the temperature range of this test would survive. Therefore, there are probably other physiological changes in the tubers in a natural environment, which were not brought about under the conditions of this experiment, that enable the tubers to withstand the colder temperatures.

It was interesting to note that there was some similarity between the percentage of survival of tubers at various temperatures in this test and in the one preceding, which was conducted under field conditions. However, as stated, the mildness of the winter prevented the evaluation of tuber survival in the field at near zero or subzero temperatures. Therefore, no definite comparisons could be made between the two tests at these colder temperatures.

Table 4. Survival of sago pondweed tubers to gradual lowering temperatures.

Treatment number	Minimum temp. (°C)	Survival percentage
1	7.2	100
2	0.6	89
3	-1.1	89
4	-3.3	56
5	-3.3	44
6	-6.7	0
7	-11.1	0
8	-13.3	0

DEPENDENCE OF THE EMERGING SHOOT
ON THE PARENT TUBER

Methods and Materials. The extent of the dependence of new sago pondweed shoots upon the parent subterranean tubers for subsistence was studied by separating shoots from tubers at various times. Tubers were collected from the bottom of Satus No. 2 Extension, Wapato Irrigation Project, in mid-October of 1965 and stored at 7.2 degrees C for approximately ten weeks. Twenty-seven one-quart ice cream cartons were cut to form 9 cm cylinders. A tuber, weighing approximately 0.4 g, was placed near the bottom of each container, which was then filled with soil and submerged in greenhouse aquaria. At 0, 1, 2, 3, 4, 8, 12, and 16 days after emergence, three containers were lifted from the aquaria, a rectangular flap cut in the bottoms, the tubers carefully excised, the flaps closed, and the containers resubmerged. In three containers, serving as controls, the tubers were not removed. The growth of the shoots was observed for a period of 20 days following emergence. Plants, including roots and rhizomes, were then harvested, fresh weights taken, and dry weights were determined by oven drying for 28 hours at 65 degrees C.

Because all shoots survived, an additional study was conducted using six tubers, each weighing approximately 0.3 g. The entire sprout was excised from three of the tubers (Figure 7). Only the upper 2 cm of the sprout was removed from the other three tubers (Figure 9). The excised sprouts and portions of sprouts were planted at a depth of 1.5 cm in soil in greenhouse aquaria.

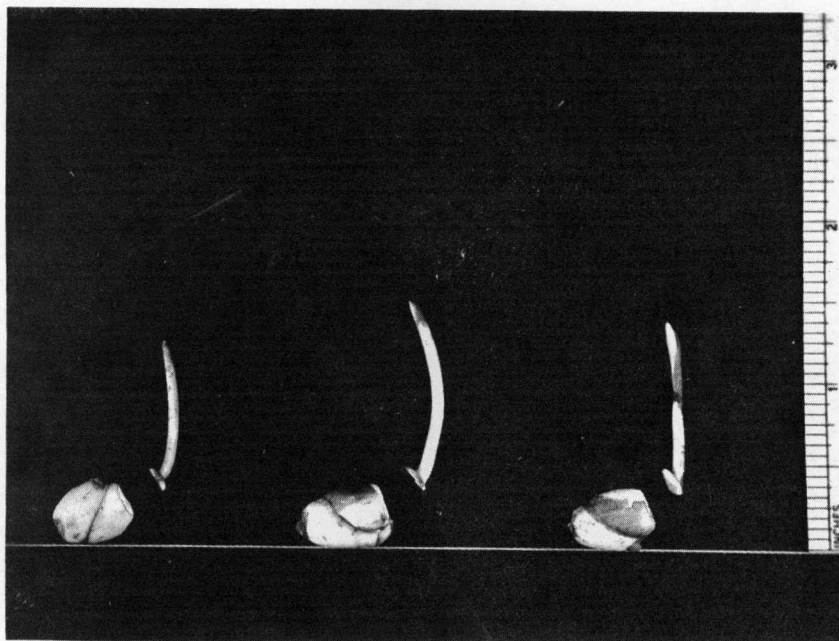


Figure 7. Sago pondweed tubers with sprouts excised. Only the sprouts were planted.

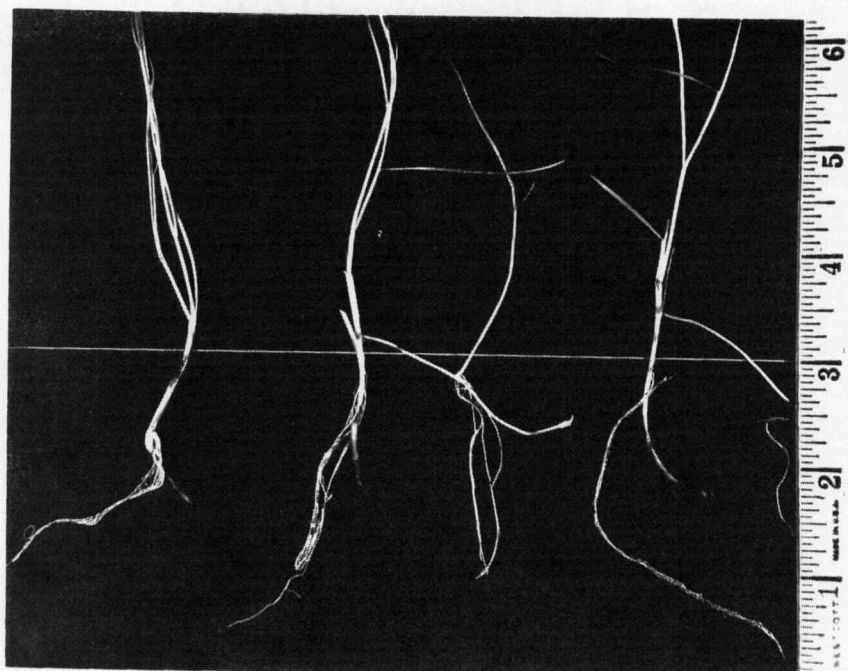


Figure 8. Plants that developed from the excised sprouts.

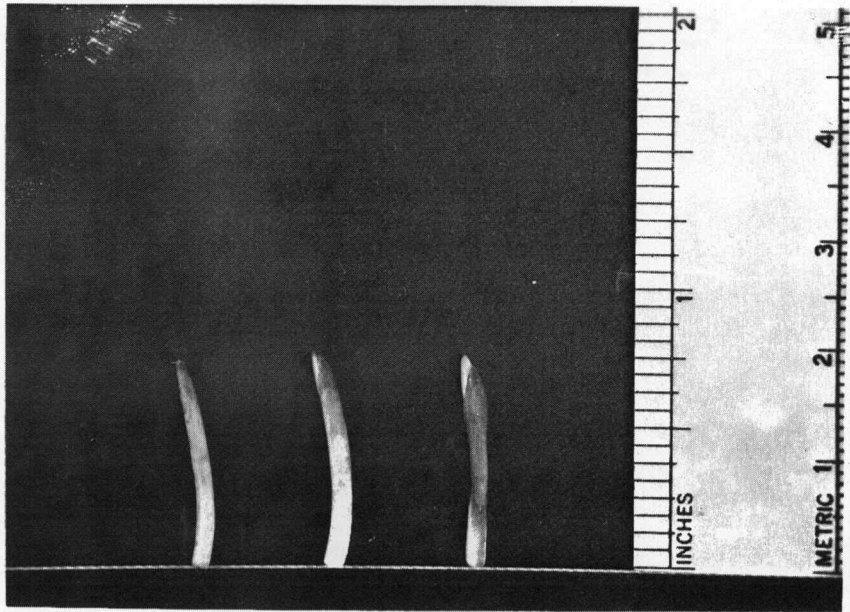


Figure 9. Excised sprout tips 2 cm long.

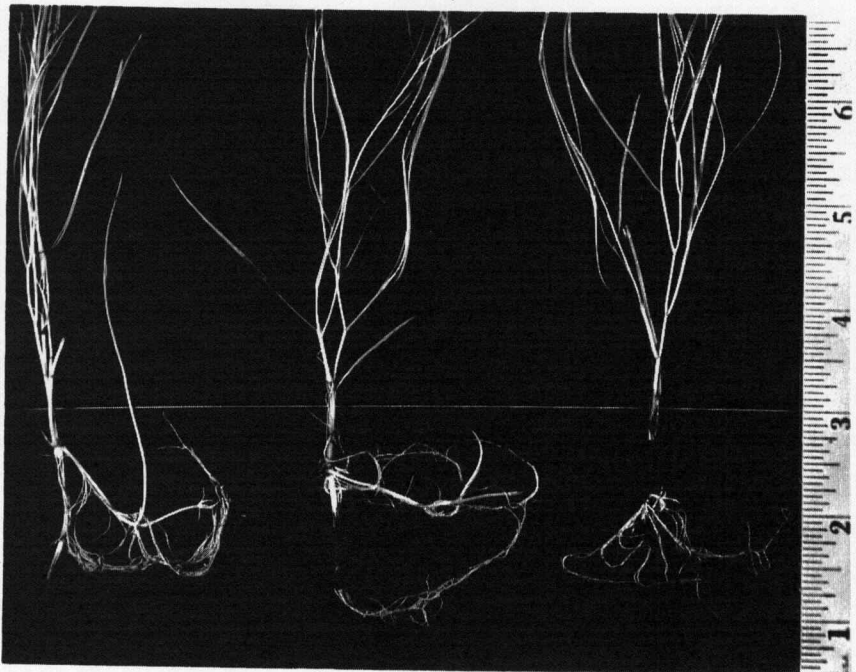


Figure 10. Plants that developed from excised sprout tips.

Shoots that emerged were observed for a period of 20 days. As before, plants, including roots and rhizomes, were then harvested and fresh and dry weights taken.

Results and Discussion. Plants developed and grew from all shoots regardless of when the parent tuber was removed (Table 5). However, the dry weights of the excised plants were less than the dry weights of the controls. Vigorous plants developed and grew from the excised sprouts and sprout-tips (Figures 8 and 10). The main portions of the tubers were not necessary for the initiation of growth of sago pondweed. Therefore, even if the sprout-tips were removed from the tuber, it is still capable of producing a plant. The main portion of the tuber has a function other than initiating growth. One probable function is as a food reserve, enabling the shoot to emerge from considerable depth in the soil.

Table 5. Weights of sago pondweed plants harvested 20 days after emergence.

Treatment	Fresh ^{a/} Wt. (g)	Dry ^{a/} Wt. (g)
Tubers removed the day of shoot emergence.	1.1056	0.0615
Tubers removed 1 day after shoot emergence.	1.1582	0.0703
Tubers removed 2 days after shoot emergence.	0.7954	0.0378
Tubers removed 3 days after shoot emergence.	0.8328	0.0511
Tubers removed 4 days after shoot emergence.	0.7065	0.0312
Tubers removed 8 days after shoot emergence.	1.2147	0.0827
Tubers removed 12 days after shoot emergence.	1.7734	0.1107
Tubers removed 16 days after shoot emergence.	2.1447	0.1316
Tubers excised, only sprout planted.	1.7538	0.1016
Only 2 cm of sprout planted.	2.3700	0.1392
Untreated checks	2.7821	0.1768

^{a/}Weights were the total of the three plants in each treatment.

STUDY OF THE EMERGENCE OF SHOOTS FROM TUBERS

Series I

Methods and Materials. Sago pondweed tubers were collected from the bottom of Satus No. 2 Extension, Wapato Irrigation Project, in mid-October of 1965. The tubers were graded into three weight classes, i.e., approximately .15 g, .30 to .50 g, and 1.0 g. Tubers from each weight class were then planted in triplicate at depths of 15, 23, and 30 cm in soil in greenhouse aquaria. Three tubers, one from each weight class, were planted at the same depth in one glass aquarium.

At the end of 51 days, aquaria in which shoots did not reach the surface of the soil were emptied to determine whether the tubers had not germinated or whether the tubers had germinated but the shoots had not reached the surface. Ungerminated tubers were then placed in shallow soil in other greenhouse aquaria to test for viability.

Results and Discussion. In the large weight class, shoots emerged from 100 percent of the tubers planted at the 15- and 23-cm depths, but shoots emerged from only 33 percent at the 30 cm depth (Table 6). In the .30 to .50 g class, shoots emerged from 67 percent of the tubers at the 15 cm depth, none from the nine-inch depth, and only 33 percent from the 30 cm depth. Shoots emerged from only 33 percent of the tubers weighing approximately .15 g at the 15- and 23-cm depths. None of the shoots emerged from the 30 cm depth. As was expected, there was a correlation between the depth from which

shoots would emerge and the weight of the tubers. Not only did a greater percentage of the shoots of larger tubers emerge, but they also emerged from greater depths than did the shoots of small tubers. Therefore, a mechanical method of killing sago pondweed tubers in the upper 15 or 23 cm of soil would not be an effective method of control because of the ability of the larger tubers to emerge from depths of 30 cm or more.

In nearly all cases, the shoots either reached the surface or the tubers did not germinate. Moreover, subsequent plantings proved that the ungerminated tubers were viable. The factor or factors that trigger germination of tubers under such conditions are unknown. The presence or absence of certain quantities of gases, such as carbon-dioxide or oxygen, may be a factor affecting germination of tubers.

Table 6. Depth of emergence of sago pondweed shoots from tubers planted in greenhouse aquaria, October, 1965.

Average tuber weight (g)	Depth planted (cm)	Percentage of emergence
1.15	15	100
1.05	23	100
1.08	30	33
0.50	15	67
0.30	23	0
0.29	30	33
0.17	15	33
0.15	23	33
0.15	30	0

Series II

Methods and Materials. The same method was used in Series II as in Series I, except that three additional depths (8, 38, and 46 cm) were included. The smallest weight class (.15 g) was not planted deeper than 30 cm, based on the belief that no shoots would emerge from tubers at a depth greater than 30 cm.

Three and seven-tenths liter glass jars were used to contain tubers at the 8 and 15 cm depths. Special aquaria made from 10-cm black plastic drain pipe were used for the other four depths (Figure 11). The pipe was cut at the appropriate lengths, capped, inverted, and filled with the required amount of soil.

Aquaria in which shoots did not emerge were examined after 51 days, as in Series I.

Results and Discussion. Shoots emerged from all of the tubers planted at the 8-cm depth (Table 7). Shoots from 100 percent of the tubers of the .50 and 1.0 g weight class and from 67 percent of the tubers of the .15 g weight class emerged from the 15-cm depth. Sixty-seven percent of the tubers of the 1.0 g class, 100 percent of the .50 g class, and 33 percent of the .15 g weight class produced shoots from the 23-cm depth. No shoots emerged from tubers planted 30, 38, or 46 cm deep.

Examination of the tubers after the 51 days revealed that most of the shoots that did not emerge had germinated, but had died. Usually the shoots grew towards the side of the container before turning and growing towards the surface. Furthermore, the shoots had

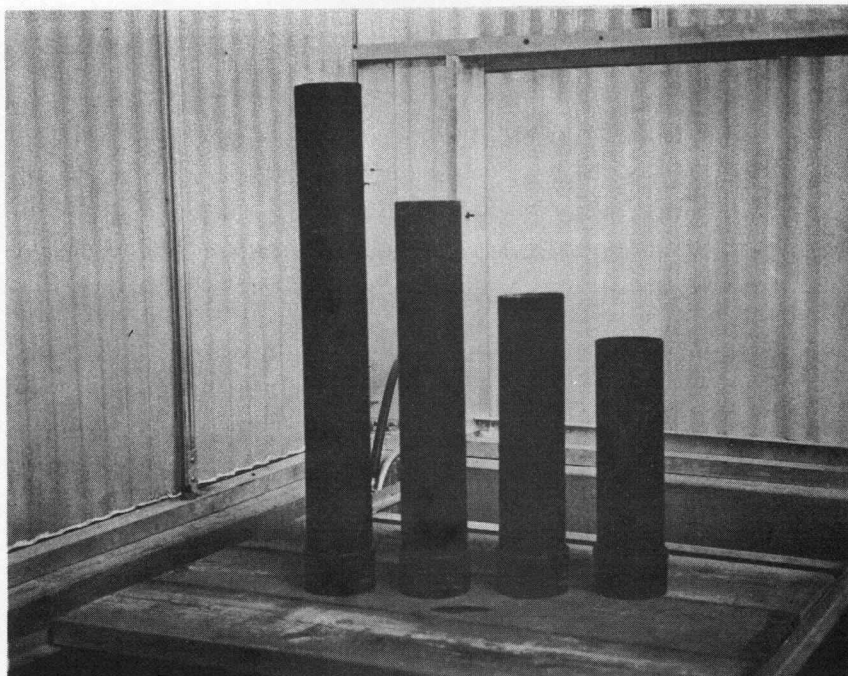


Figure 11. Aquaria of plastic drain pipe used in the study of shoot emergence.

not grown smooth and straight as expected, but were kinked and twisted (Figures 12 and 13). The tips of many of these shoots were flaccid and soft. Because the shoots that behaved in this unusual manner were from tubers planted in the plastic pipe aquaria, the possibility exists that some toxic substance on or in the plastic pipe was leached into the soil and killed the growing tip. However, several shoots did emerge from tubers that were planted in the plastic pipes. Because the exact cause of this unusual growth pattern is unknown, it is not possible to make conclusive statements concerning this test.

Table 7. Depth of emergence of sago pondweed shoots from tubers planted in greenhouse aquaria, December, 1965.

Average tuber weight (g) ^{a/}	Depth planted (cm)	Percentage of emergence
0.88	8	100
0.88	15	100
0.98	23	67
1.05	30	0
1.16	38	0
1.29	46	0
0.05	8	100
0.47	15	100
0.50	23	100
0.47	30	0
0.46	38	0
0.46	46	0
0.13	8	100
0.12	15	67
0.13	23	33
0.14	30	0

^{a/}Values given are the average of three replicates with one tuber per replication.

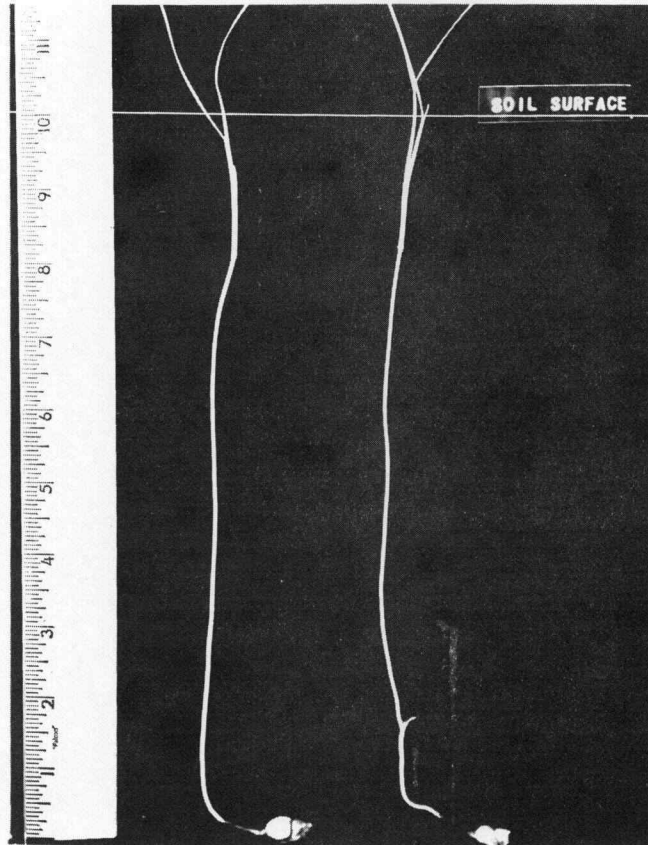


Figure 12. Growth form of shoots from tubers planted in a glass aquarium.

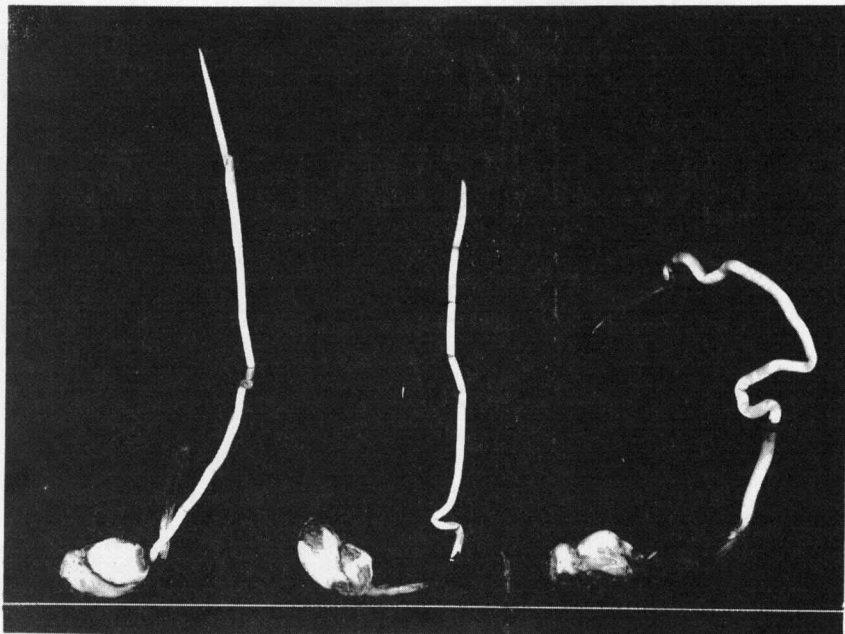


Figure 13. Growth form of shoots from tubers planted in plastic pipe aquaria.

GERMINATION OF SAGO PONDWEED SEED
IN A GREENHOUSE AQUARIUM

The recorded occurrence of sago pondweed seedlings in nature is rare. However, the abundance of seed that is produced would indicate that seed may be an important mode of spread of sago pondweed in irrigation systems. Yeo (1965) obtained 14 percent germination of sago pondweed seeds that had overwintered in a canal bottom.

Methods and Materials. On October 6, 1965, 100 seeds of sago pondweed, that had overwintered in a canal bottom, were dropped into a greenhouse aquarium, the bottom of which was covered with about 5 cm of top soil. A thermograph was used to record the water temperature. Periodic observations were made and the number of germinated seeds was recorded.

Results and Discussion. Within five days after seeding, all seeds had settled to the soil (Appendix Table 9). Sixteen days after seeding, the first seedling was noted. Fifteen percent of the seeds germinated during the 50-day period of the test. The development of the seedlings is shown in Figures 14 and 15. Water temperature during the test period ranged from a minimum of 15.5 degrees C. to a maximum of 25.5 degrees C. However, the temperature generally ranged between 21 and 24 degrees C.

The results of this test were very similar to those obtained by Yeo (1965). The number of seeds that germinated would indicate that seed could be important in distribution and that seedlings should occur in irrigation channels.

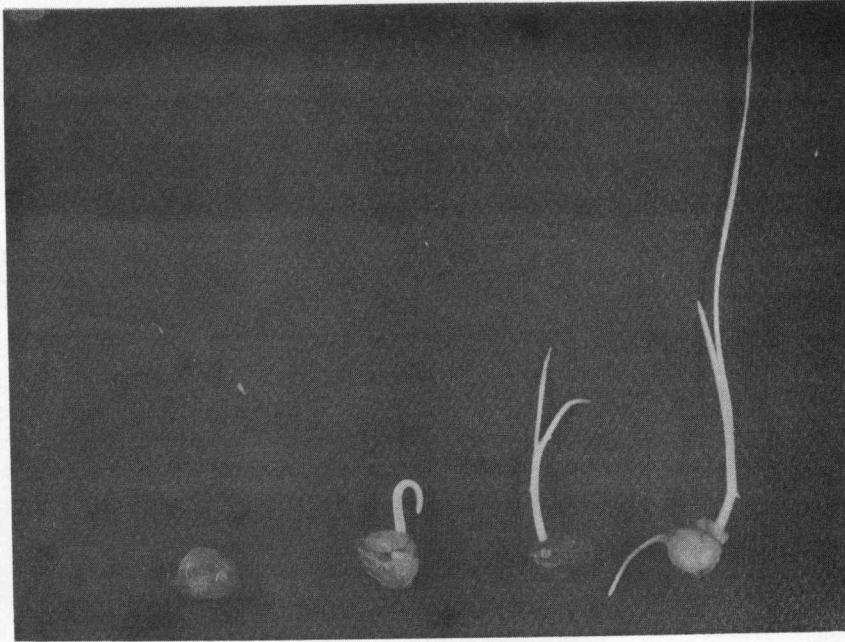


Figure 14. Stages in the development of sago pondweed seedlings.

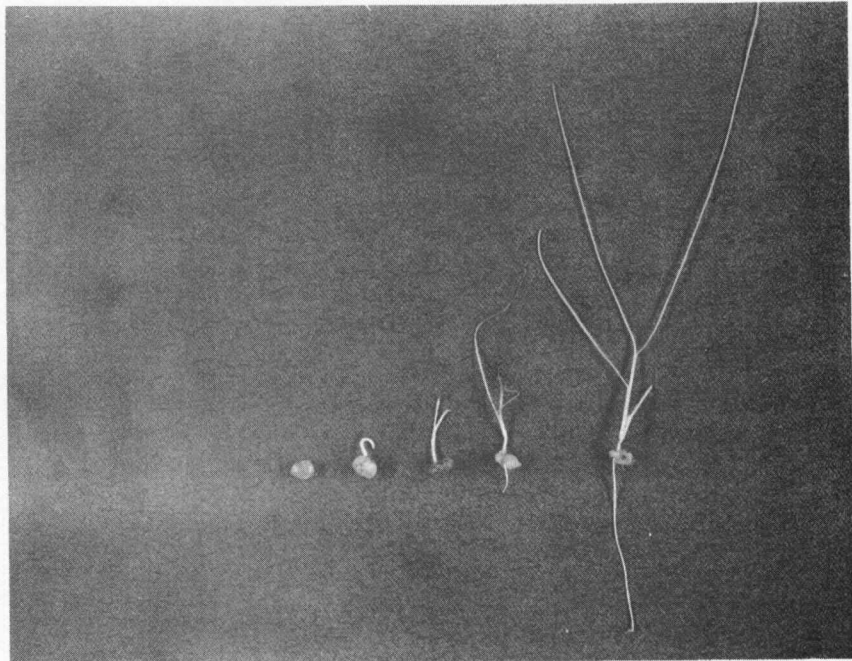


Figure 15. Stages in the development of sago pondweed seedlings.

THE EFFECT OF PASSAGE THROUGH THE DIGESTIVE TRACTS
OF WILD DUCKS ON GERMINATION OF SAGO PONDWEED SEEDS

There are several contradictory reports of the effect of the passage through the digestive tracts of waterfowl on the germination of sago pondweed seeds. Guppy (1894) found that waterfowl carried the seeds and seed-like fruits of some aquatic plants unharmed in their stomachs. On the other hand, Chapman (1964) concluded that passage through the digestive tract of wild ducks retarded the germination of sago pondweed seeds.

Methods and Materials. Five wild mallard ducks (Anas platyrhynchos) were obtained from the National Waterfowl Refuge at Othello, Washington. Three of the ducks were drakes and two were hens. The ducks were placed in small individual cages and given food and water (Figure 16). Metal trays were placed under the cages to collect the droppings. The ducks were allowed to become adjusted for about two weeks before the test began.

Two hundred sago pondweed seeds were placed in the food can in each cage. After 24 hours, the seeds not eaten were collected, counted, and placed in the germinator to serve as controls. The droppings were screened every day to collect the seed that had passed through the ducks. The use of 32-mesh nylon screen and light water pressure facilitated the screening process. All of these seeds were placed in the laboratory germinator. Collection of droppings continued until no seeds were found in the droppings for three consecutive days.

All seeds were held in a germinator, with a temperature cycle

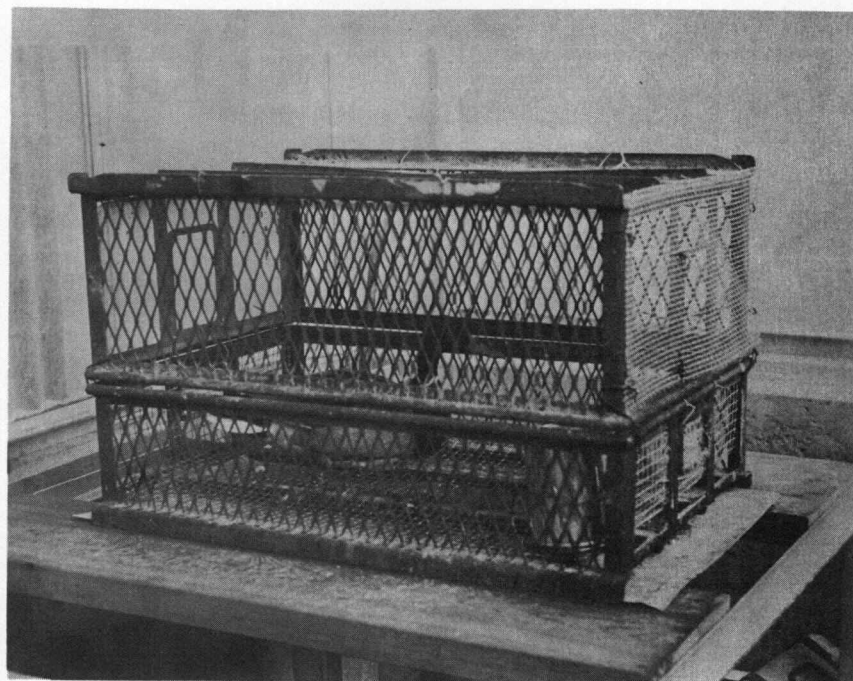


Figure 16. Figure shows the type of cage used to contain wild ducks fed sago pondweed seed. Metal trays were placed under cages to collect droppings.

of nine hours at 30 degrees C. alternating with 15 hours at 20 degrees C. for 28 days.

Results and Discussion. The average of 35.2 percent (Table 8) germination of the sago pondweed seeds that were held in the digestive tracts of the wild ducks for one day was slightly more than six times the average percentage of germination of the untreated checks. The percentage of germination declined with length of time that the seeds had remained in the digestive tract of the ducks. No germination occurred from seeds that had been held longer than three days.

During the first day or two after the seeds were eaten, there was apparently only enough mechanical and chemical action on the seed to break down the hard exocarp and rupture the seed coat. Longer exposure of the seed to these forces resulted in injury and death to the embryo.

This test indicated that waterfowl can be an important factor in the spread of sago pondweed. They could ingest the seed of sago pondweed and carry them many miles before passing the seed, thus providing propagules for new infestations of this plant.

Table 8. Germination of sago pondweed seeds that passed through the digestive tracts of wild ducks.

Duck number	No. of days After ingestion	No. of seeds placed in germinator	Percentage of germination
1	1	27	22.2
2	1	55	25.5
3	1	79	39.2
4	1	14	28.6
5	1	38	60.5
			Average 35.2
1	2	17	35.3
2	2	3	0.0
3	2	3	0.0
4	2	6	16.7
			Average 13.0
1	3	7	0.0
2	3	6	16.7
4	3	15	0.0
5	3	2	0.0
			Average 4.2
3	4	1	0.0
4	4	17	0.0
			Average 0.0
4	5	3	0.0
			Average 0.0
1-CK	0	27	0.0
2-CK	0	84	6.0
3-CK	0	65	3.1
4-CK	0	40	12.5
5-CK	0	83	7.2
			Average 5.8

GERMINATION OF SAGO PONDWEED SEED
IN THE LABORATORY

A study was initiated on February 11, 1966, to determine the effects of freezing and thawing during storage in water and soil at different temperatures on the germination of sago pondweed seeds. The storage conditions and temperatures were selected to simulate those that might occur in a natural environment.

Methods and Materials. The seeds used in the study were collected on October 17, 1965, from plants growing in Lateral EL 63.8 on the Columbia Basin Project in central Washington. The seeds were placed in dry storage at room temperature until the test began. A South Dakota blower was used to remove the trash and light seeds before the 50-seed lots were counted out and placed in small petri dishes. Four 50-seed lots were subjected to each treatment. The seeds were stored for periods of three, six, nine, twelve, and fifteen days in water or wet soil at temperatures as follows:

- (A) In water at 20 degrees C.
- (B) In water at 0 degrees C.
- (C) In water at -18 degrees C.
- (D) In water at alternating temperatures of -18 and 20 degrees C.
- (E) In wet soil at alternating temperatures of -18 and 20 degrees C.

In the two treatments continuously cycled between -18 and 20 degrees C. the seeds were held for nine hours at -18 degrees C. and then for 15 hours at 20 degrees C.

Four 50-seed lots of dry stored seed were placed in the

germinator at the end of each three-day period to serve as controls. At the end of each three-day period, four 50-seed lots of seed from each treatment were placed in the germinator for 30 days. The temperature in the germinator ranged from 30 degrees C. during the day to 20 degrees C. during the night for the first 15 days of the test. Only one seed germinated during the first 15 days. At that time, a review of Chapman's (1964) work indicated that a constant temperature of 27 to 29 degrees C. resulted in highest percent germination for this species. It was decided to maintain the germinator at a constant 30 degrees C. for the remainder of the test. This resulted in the following temperature conditions for the five storage periods:

3 days storage	15 days at 20-30°C. + 15 days at 30°C.
6 days storage	12 days at 20-30°C. + 18 days at 30°C.
9 days storage	9 days at 20-30°C. + 21 days at 30°C.
12 days storage	6 days at 20-30°C. + 24 days at 30°C.
15 days storage	3 days at 20-30°C. + 27 days at 30°C.

The seeds were observed and the number of germinated seeds recorded at least once every three days during the 30 days incubation period.

The data were transformed by using the $\arcsin\sqrt{\text{percentage}}$ and analyzed statistically as a factorial analysis for a completely randomized design. Duncan's new multiple range test was used to tell differences between storage condition and storage period means (Steel and Torrie, 1960).

Results and Discussion. The average percentages of germination of sago pondweed seeds stored under various conditions and for

different periods of time are given in Table 9. These percentages ranged from a high of 62.5 percent for seeds stored in water at 0 degrees C. for 12 days to a low of 1.0 percent for seeds stored in water at alternating temperatures of -18 to 20 degrees C. for 15 days. The effect of storage conditions and storage periods on the percentage of germination and the interaction of these two variables are shown in Figure 17. When seeds were stored in water at 0 degrees C., there was a marked increase in percentage of germination as the storage period was lengthened from three to six days. The percentage germination was drastically reduced when the storage period was extended beyond nine days for seeds stored in water or wet soil at alternating temperatures of -18 to 20 degrees C. Generally, there was a gradual decrease in the percentage of germination as the storage period was increased to 15 days for seeds stored in water at -18 or 20 degrees C. and for seeds stored dry at room temperature.

The statistical computations were made with transformed data because the percentages of germination had wide variation. Although there was considerable interaction between the two variables of storage conditions and storage periods, the analysis of variance (Appendix, Table 10) indicated there was a significant difference in means at the 1 percent level. The original data and not the transformed data are shown in Table 9. Storage of sago pondweed seeds in water at 0 degrees C. resulted in a significantly higher percentage of germination than the other storage conditions. Storage of seeds in water at -18 and 20 degrees C. and dry at room temperature,

Table 9. Average percentage of germination after a 30-day incubation period of sago pondweed seeds stored at different moisture and temperature conditions for different periods of time.

Storage condition	Percent Germination					Average
	Days of storage					
	3	6	9	12	15	
0°C. in H ₂ O	24.5 ^{a/}	49.0	51.5	62.5	55.5	48.5 a ^{b/}
-18-20°C. in H ₂ O	33.0	48.5	54.5	3.0	1.0	28.0 b
-18-20°C. in soil	22.0	34.5	46.5	2.0	3.0	21.6 bc
-18°C. in H ₂ O	19.5	19.0	9.5	12.0	4.5	12.9 cd
Dry room temp.	21.0	12.5	8.0	7.5	5.5	10.9 d
20°C. in H ₂ O	22.0	13.0	6.5	4.5	3.0	9.8 d
Average	23.7 a ^{b/}	29.4 a	29.4 a	15.3 b	12.1 b	

^{a/}Each average is the mean of four lots of seed.

^{b/}Averages in the column or row followed by the same letter do not differ significantly at the 1 percent level as determined by Duncan's new multiple range test.

resulted in a significantly lower percentage of germination than the remaining storage conditions. There was no significant difference in the average percentage of germination of sago pondweed seeds stored for three, six, and nine days (Table 9). However, when the storage period was extended to 12 or 15 days, there was a significant decrease in the average percentage of germination.

Figure 17 clearly illustrates the result of this test. Storage of sago pondweed seed in water just above freezing resulted in the highest percentage of germination. This information should be of vital importance to waterfowl management for the establishment of new stands of this important waterfowl food plant. Although this

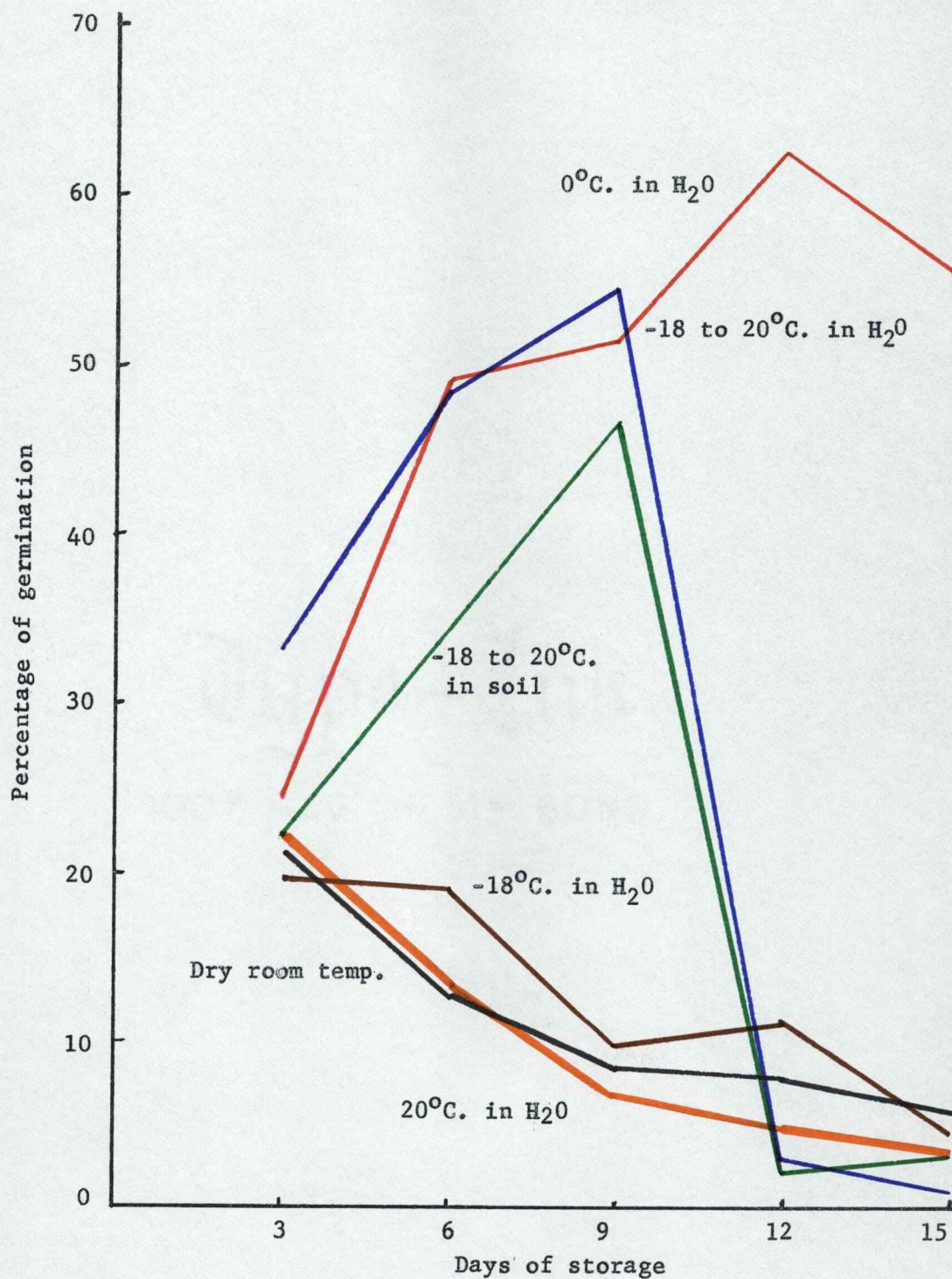


Figure 17. Germination of sago pondweed seeds stored at different temperatures in water and wet soil for different periods of time.

type of seed conditioning could occur in lakes and rivers, it is not likely to occur in irrigation systems that are dry during the winter months. Alternating freezing and thawing of water or wet soil would be more likely to occur in irrigation canals. Figure 17 shows that the percentage of germination under these two conditions increased as the storage time increased from three to nine days. However, after 12 days of storage, the percentage of germination had decreased to very low levels. This decrease might be due to rupturing of the seed coat by the repeated freezing and thawing after nine days, resulting in destruction of the embryo when it imbibed water and was again subjected to freezing temperatures. If this was indeed the case, it may explain the apparent lack of sago pondweed seedlings in irrigation canals. On the other hand, 15 percent germination of seeds that have overwintered in a canal bottom has been reported. Further investigation of the effects of alternating freezing and thawing on the germination of sago pondweed seeds may explain these findings.

SUMMARY AND CONCLUSIONS

A series of experiments were conducted during 1964 and 1965 to provide a better understanding of the reproduction and mode of spread of sago pondweed (Potamogeton pectinatus) in irrigation systems. The field experiments were conducted in canals on several irrigation projects in south central Washington. The laboratory studies were conducted at the Irrigated Agriculture Research and Extension Center at Prosser, Washington.

Nine experiments were completed. Field experiments included studies on the reduction of stands of sago pondweed and tuber production by chemical treatments, ecological investigation of colonization in new irrigation channels, and a study of the survival of sago pondweed tubers following freezing temperatures under field conditions. Laboratory and greenhouse studies used sago pondweed tubers to test for hardening of tubers to freezing temperatures, for emergence of shoots from tubers at different depths in the soil, and for the dependence of shoots on the parent tuber. Studies were also conducted determining the percentage of germination of seeds placed in greenhouse aquaria, of seeds that had passed through the digestive tracts of wild-ducks, and of seeds that had been stored at different temperature and moisture conditions for different periods of time. The following results were obtained:

1. One season of treatments at one- or two-week intervals with aromatic solvent greatly reduced the number of sago pondweed tubers in irrigation canals. However, complete eradication of all tubers

in one season was not obtained. The tubers were either carried over in the substratum for more than one year, or the rhizomes continued to form new tubers even when the topgrowth was periodically removed. The chances of new tubers being produced would probably have been greatly reduced if the treatments had been started in mid-April instead of mid-May.

2. Sago pondweed proved capable of invading and becoming established in new irrigation channels in one season. The aquatic annuals were the first to appear in the spring, but by the end of the irrigation season the perennial aquatics, such as sago pondweed, were well established. There was evidence that aquatic herbicides may actually shorten the time required for this succession. The propagules responsible for the establishment of new infestations were tubers and plant fragments.

Considerable numbers of sago pondweed seed were transported into the new channels by the water, but no seedlings were found. A field study involving the planting of seeds in a controlled channel should provide information on the true or relative importance of this propagule in the spread of sago pondweed.

3. Sago pondweed tubers survived freezing temperatures under field conditions. However, the percentage of survival in the laboratory decreased as the temperature decreased below freezing. Apparently, there was some mechanism that provided a degree of protection for sago pondweed tubers in a natural environment. The mild winter prevented the evaluation of the ability of tubers to survive the cold temperatures that normally occur. A study

conducted during a cold winter might reveal more information on the protection mechanism.

4. Laboratory studies showed that sago pondweed tubers could become hardened or conditioned to freezing temperatures to some degree. There was a gradual reduction in the percentage of survival as the temperature decreased to -6.7 degrees C. No tubers survived at -6.7 degrees C. or lower.

5. Survival of the emerged shoot was not affected by the time of removal of the parent tuber. Furthermore, excised sprout tips grew and developed vigorous plants. The results indicated that the tuber was not necessary for the initiation of growth. One possible function could be as a food reserve enabling the shoot to emerge from a considerable depth in the soil.

6. The study of shoot emergence indicated a relationship between tuber weight and the depth from which the shoot would emerge. In one test series there was a decrease in percentage of emergence as the weight of tuber was reduced from approximately 1.0 g to 0.15 g. In the second test series, interference by unknown factors caused equivocal results.

Further study is needed on the relationship between tuber weight and depth from which the shoots will emerge. In the laboratory study conducted, the soil was of uniform texture from top to bottom. However, in canal bottoms the soil is layered with zones of different textures. Tuber shoots can probably grow more easily in coarse, loose soils than they can in fine, tight soils. The use of coarse textured soil or layered soil of different textures to

simulate natural conditions might alter the results appreciably.

7. Seeds of sago pondweed that had overwintered in a canal bottom gave 15 percent germination after 50 days in a greenhouse aquarium.

8. The passage of sago pondweed seeds through the digestive tracts of wild-ducks greatly enhanced the germination. However, if the seeds were held in the digestive tract more than three days, no germination occurred. During the first day or two after ingestion there was apparently only enough mechanical and chemical action on the seed to break down the hard exocarp and rupture the seed coat. Longer exposure of the seed to these forces probably resulted in injury and destruction of the embryo.

9. Storage of sago pondweed seeds in water at 0.0 degrees C. greatly increased the percentage of germination over that of seeds stored in water at -18 and 20 degrees C. or dry at room temperature. The percentage of germination of sago pondweed seed stored in water or wet soil at alternating temperatures of -18 and 20 degrees C. increased as the storage period increased to nine days. A further increase in the storage period resulted in a rapid decrease in the percentage of germination. Although the recorded occurrence of sago pondweed seedlings in nature is rare, a high percentage of germination could result if the proper conditions occurred.

A considerable amount of error might have been introduced into this test by changing of germination temperatures 15 days after the test began. A better technique would have been to place the seeds in storage at different times, thus enabling all seeds to be

removed from storage at the same time. In this way, all seeds would have been exposed to the same conditions of germination.

The results of this study have been more effective in pointing out the complexity of developing new and improved control methods, than in finding a satisfactory control for sago pondweed. The various methods of reproduction have many special adaptations that can adjust to adverse environmental conditions. The ability of tubers to survive freezing, to emerge from considerable depth in the soil, and to persist in the soil from year to year and the ability of shoots from tubers to survive and grow even after the main portion of the tuber has been removed indicates the scope of the adaptations.

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APPENDICES

Appendix Table 1. Data on introductions of aromatic solvents,
Prosser West Branch, 1965.

Canal data				Treatment data		
Flow (cfs)	Water temp. (°C)	Water turbidity	Date	Interval (wks.)	Rate	
					Liters/cm	Minutes
6	15.6	Murky	5-17-65	4	100	30
5	15.0	Murky	5-24-65	1	100	45
5	15.0	Murky	6-1-65	2	100	33
5	16.1	Murky	6-7-65	1	100	35
6	16.7	--	6-14-65	4	100	30
5	18.9	Murky	6-21-65	1	100	34
5	16.7	Murky	6-28-65	2	100	34
5	21.7	Murky	7-6-65	1	100	31
7	17.8	Murky	7-13-65	4	100	30
5	21.1	Sl. murky	7-19-65	1	100	31
5	20.0	Sl. murky	7-26-65	2	100	29
5	21.1	Sl. murky	8-2-65	1	100	31
7	22.2	Sl. murky	8-10-65	4	100	30
5	21.7	Clear	8-16-65	1	100	31
5	20.0	Sl. murky	8-23-65	2	100	29
5	15.6	Clear	8-30-65	1	100	30
7	15.6	Clear	9-7-65	4	100	30
5	16.7	Clear	9-13-65	1	100	29
5	12.8	Clear	9-20-65	2	100	32
5	15.6	Clear	9-27-65	1	100	30
5	14.4	Clear	10-4-65	4	100	29

Appendix Table 2. Observations of the plot in the Prosser West Branch treated every week with aromatic solvent, 1965.

Date	Stand	Max. length (cm)	Color	Remarks
5-14-65*	Sparse	89	Normal	Algae along banks.
5-24-65*	--	--	--	Observation notes lost.
6-1-65*	Sparse	25-30	Br. green	Basal leaves sloughed.
6-7-65*	Sparse	13	Normal	Basal leaves flaccid; some stem injury.
6-14-65*	Sparse	15	Normal	Mostly regrowth
6-21-65*	Sparse	15	Br. green	Mostly regrowth; some old stems.
6-28-65*	Sparse	20	Br. green	All new growth.
7-6-65*	Sparse	25	Br. green	Growth vigorous.
7-13-65*	Sparse	15-20	Normal	Regrowth from old stems; rhizomes and tubers.
7-19-65*	Sparse	13	Normal	Growth very sparse and spindly.
7-26-65*	Sparse	18	Normal	Growth extremely sparse but vigorous.
8-2-65*	Sparse	5	Normal	Only 2 shoots.
8-9-65*	None	--	--	No pondweed shoots. Algae present.
8-16-65*	None	--	--	No shoots or algae. No algae.
8-23-65*	None	--	--	No pondweed shoots.
8-30-65*	Sparse	20	Normal	Only 3 shoots.
9-7-65*	None	--	--	No pondweed shoots.
9-13-65*	None	--	--	No pondweed shoots.
9-20-65*	None	--	--	No pondweed shoots.
9-27-65*	None	--	--	No pondweed shoots.
10-4-65*	None	--	--	No pondweed shoots.
10-11-65	None	--	--	Water temp. 12.2°C. No shoots.
10-18-65	None	--	--	No pondweed shoots.

*Treatment dates

Appendix Table 3. Observations of the plot in the Prosser West Branch treated every 2 weeks with aromatic solvent, 1965.

Date	Stand	Max. length (cm)	Color	Remarks
5-14-65*	Sparse	89	Normal	Algae along banks.
5-24-65	--	--	--	Observation notes lost.
6-1-65*	Moderate	25-30	Normal	Initial growth not completely killed.
6-7-65	Moderate	30	Fair	Lower leaves sloughed, some stem injury.
6-14-65*	Moderate	30	Fair	Color normal to brownish-green.
6-21-65	Moderate	15	Normal	Some older plants surviving.
6-28-65*	Moderate	4	Fair	Color normal to brownish-green.
7-6-65	Sparse	10-13	Normal	Older stems deleaved, rotting, and sloughing.
7-13-65*	Moderate	25-30	Fair	Surviving older stems brownish-green.
7-19-65	Sparse	3-5	Normal	Some older plants surviving.
7-26-65*	Sparse	13-15	Fair	Color brownish-green to near normal.
8-2-65	Sparse	3-5	Normal	Old remaining stems brown and flaccid.
8-9-65*	Sparse	23	Normal	Plants not vigorous, algae present.
8-16-65	None	--	--	No pondweed shoots, no algae.
8-23-65*	Sparse	30	Fair	Plant vigor fair.
8-30-65	Sparse	13	Normal	Plant vigor fair.
9-7-65*	Sparse	13	Normal	Growth extremely sparse, but vigorous.
9-13-65	None	--	--	No pondweed shoots.
9-20-65*	None	--	--	No pondweed shoots.
9-27-65	None	--	--	No pondweed shoots.
10-4-65*	None	--	--	No pondweed shoots.
10-11-65	None	--	--	Water temp. 12.2°C. No shoots.
10-18-65	None	--	--	No pondweed shoots.

*Treatment dates.

Appendix Table 4. Observations of the plot in the Prosser West Branch treated every 4 weeks with aromatic solvent, 1965.

Date	Stand	Max. length (cm)	Color	Remarks
5-14-65*	Sparse	89	Normal	Algae along banks.
5-24-65	--	--	--	Observation notes lost.
6-1-65	Moderate	25-30	Normal	Initial growth not completely killed.
6-7-65	Moderate	48	Normal	Growth vigorous.
6-14-65*	Moderate	61	Fair	Color normal to brownish-green.
6-21-65	Moderate	15	Normal	Some older plants surviving.
6-28-65	Moderate	30-41	Fair	Color brownish-green.
7-6-65	Dense	38	Fair	Color brownish-green to normal.
7-13-65*	Dense	--	Normal	Growth nearing water surface.
7-19-65	Dense	10-13	Normal	Some older plants surviving.
7-26-65	Moderate	25	Normal	Nearly normal vigor.
8-2-65	Dense	43	Normal	Normal vigor.
8-9-65*	Dense	64	Normal	Filamentous algae present.
8-16-65	Moderate	10-13	Normal	Normal vigor. No algae.
8-23-65	Moderate	30-33	Fair	Vigor fair.
8-30-65	Sparse	25-28	Fair	Vigor fair.
9-7-65*	Moderate	46-51	Normal	
9-13-65	Sparse	5-8	Fair	Vigor fair.
9-20-65	Sparse	10	Poor	Vigor lacking.
9-27-65	None	--	--	No pondweed shoots.
10-4-65*	None	--	--	No pondweed shoots.
10-11-65	None	--	--	Water temp. 12.2°C. No shoots.
10-18-65	None	--	--	No pondweed shoots.

*Treatment dates.

Appendix Table 5. Observations of changes in plant growth in Lateral WB5-G. Columbia Basin Project, 1964.

Date	Observations
4-2-64	Placed thermograph in channel; soil sample was taken; examination of the upper 15 cm of soil revealed that sago pondweed shoots were about to emerge from the soil.
4-24-64	Many sago pondweed shoots about to emerge from the soil.
5-1-64	Sago shoots 3 cm long were noted.
5-8-64	Dense emergence of sago pondweed shoots. Maximum length 8 cm. Numerous horned pondweed seedlings also.
5-15-65	Maximum plant length was 10 cm. Color was brownish-green to normal.
5-23-64	Dense stand having maximum length of 56 cm.
6-2-64	First bud formation noted.
6-12-64	First blooming of flowers of sago noted.
7-17-64	Observed plump seed on several plants.
8-3-64	Sago was only species observed. Recent aromatic solvent and acrolein treatments had injured the plants considerably.
9-18-64	Thin stand of sago 8 to 13 cm in length.

Appendix Table 6. Observations of changes in plant growth in Lateral WB10. Columbia Basin Project, 1964.

Date	Observations
4-2-64	Placed thermograph in channel; channel had water in it. Soil sample had been taken at an earlier date. Pondweed residue of unidentified species had been observed previous to canal watering.
5-8-64	Sparse stand of horned pondweed along right bank was observed. 5 cm maximum length.
5-15-64	Thin to moderate stand of horned pondweed. Maximum length was 10 cm. Color was normal.
5-23-64	Dense stand of horned pondweed having a maximum length of 51 cm. Scattered plants of sago pondweed were also found. Maximum length of sago was 86 cm. It was noted that both the horned and the sago were fine stemmed. Color was normal.
6-26-64	Horned pondweed was recovering from recent aromatic solvent treatment and was about 13 cm long. Dense stand. Also observed scattered plants of sago and Richardson pondweed. Richardson pondweed was in the bud stage and about 64 cm long.
8-3-64	No pondweeds observed. Recent aromatic solvent treatment was 100 percent effective in removing plant growth.
9-18-64	No horned or sago pondweed was observed. A sparse stand of Richardson pondweed having a maximum length of 38 cm was observed. Two to three miles upstream, a dense stand of sago pondweed of 91 cm maximum length was observed.

Appendix Table 7. Observations of changes in plant growth in the Eltopia Branch Canal, 1965.

Date	Max. length (cm)	Remarks
3-24-65	--	Water sl. murky; little surface debris.
3-30-65	--	Water sl. murky; little surface debris.
4-6-65	--	Water sl. murky; little surface debris.
4-13-65	--	Water sl. murky; little surface debris.
5-11-65	13-25	Horned and leafy pondweeds present.
5-18-65	15-20	Moderate stand of horned pondweed.
6-2-65 ^{a/}	56	Dense stand of horned pondweed. Some injury.
6-9-65 ^{a/}	--	Growth slumping and injury evident.
6-22-65	--	Growth sloughing and floating downstream.
6-29-65	--	Initial stand stripped of leaves.
7-7-65	--	Initial stand 95% removed. Some re-growth.
7-13-65	8-10	Initial stand 100% removed. Regrowth.
7-22-65	8	Sparse stand of horned pondweed. Large numbers of sago leaves and stem pieces present.
7-27-65	5-8	Horned pondweed sparse; sago leaves and stems present, but not rooted.
8-11-65	--	Canal 99+% free of pondweeds. Two severely injured sago plants found.
8-25-65	76	Sparse stands of horned, leafy, and sago pondweed; sago in deeper water.
12-3-65	--	Scattered plants or small stands of sago pondweed in check site.

^{a/} Aromatic solvent treatments made by the Bureau of Reclamation on May 24, 1965 and June 9, 1965.

Appendix Table 8. Observations of changes in plant growth in Lateral EB-15, 1965.

Date	Max. length (cm)	Remarks
5-4-65	--	No pondweeds; 8 to 10 cm layer of muck in bottom of the channel.
5-11-65	--	No pondweeds were found.
5-18-65	--	No pondweeds were found.
5-25-65 ^{a/}	--	No pondweeds were found.
6-2-65	--	No pondweeds; algae present on substratum.
6-22-65 ^{a/}	--	No pondweeds were found.
6-29-65	--	No pondweeds were found.
7-7-65	--	No pondweeds; algae abundant.
7-13-65	5-8	Horned and leafy pondweed sparse; algae present; canal bottom mucky.
7-22-65	--	Numerous small plants of leafy and sago pondweed; plants nearly rootless and appear to be growing from plant pieces; color normal. Alisma and filamentous algae present also.
7-27-65	--	Pondweeds still small; scattered alisma plants; filamentous algae.
8-11-65	--	No pondweeds; scattered alisma; algae present.
8-25-65	76	Dense stand of leafy and horned pondweed; plant color and vigor normal; algae present.
8-31-65	--	Dense stand of leafy and horned pondweed. Growth near surface of the water.
9-8-65	--	Dense stand of leafy and horned pondweed; upper portions of plants brownish and appeared chemically injured; lower portions green.
9-21-65	38	Growth severely injured; color and vigor poor.
9-28-65	3	Horned and leafy pondweed regrowth had normal color and vigor.
12-3-65	--	No pondweeds found in observation site, however, scattered stands of sago and horned pondweed were found 400 meters downstream.

^{a/} Aromatic solvent treatments made by the Bureau of Reclamation on May 24, 1965 and June 9, 1965.

Appendix Table 9. Observations on the germination of sago pondweed seeds in a greenhouse aquarium, 1965.

Date	Minimum temp. (°C.)	Maximum temp. (°C.)	% Germ. accumulative	Remarks
10-6-65	--	--	--	100 sago seeds placed in aquarium.
10-11-65	--	--	--	All seeds settled to the bottom.
10-12-65	17.2	21.1		
10-13-65	19.4	24.4	--	--
10-14-65	19.4	22.8	--	Some seeds appear to be swelling.
10-15-65	19.4	23.3	--	--
10-16-65	15.6	21.1	--	--
10-17-65	18.9	23.3		
10-18-65	18.3	21.7	--	No germination observed.
10-19-65	21.1	23.9	--	--
10-20-65	19.4	23.9	--	--
10-21-65	21.1	25.0	--	--
10-22-65	20.0	24.4	1	1 seedling found.
10-23-65	21.7	25.0	1	--
10-24-65	21.7	25.6	3	3 seedlings found.
10-25-65	21.7	--	3	--
10-26-65 ^{a/}	--	--	3	--
10-28-65	--	--	3	--
10-29-65	--	--	6	6 seedlings observed.
11-12-65	23.3	23.9	8	8 seedlings observed.
11-13-65	22.8	23.3	8	--
11-14-65	22.8	23.3	8	--
11-15-65	22.8	24.4	10	10 seedlings observed.
11-16-65	22.2	23.9	12	12 seedlings observed.
11-17-65	22.8	23.3	12	--
11-18-65	22.8	23.3	12	--
11-19-65	22.8	23.9	12	--
11-20-65	22.8	23.9	12	--
11-21-65	22.2	23.3	12	--
11-22-65	21.1	23.3	12	--
11-23-65	21.1	22.2	12	--
11-24-65	22.2	22.8	12	--
11-25-65	22.8	23.9	12	--
11-26-65	22.2	23.3	15	15 seedlings observed.

^{a/}The daily temperature recorder was removed on October 26 and replaced with a weekly temperature recorder on November 12, 1965.

Appendix Table 10. Analysis of variance of data concerning germination of sago pondweed seed.

Source of variation	Degrees of freedom	Sum of squares	Mean squares	F test
Treatments	(29)	(24,812)		
Storage conditions	5	10,292	2,058.3	70.0**
Storage period	4	5,387	1,346.8	42.8**
Condition X period	20	9.203	460.1	15.7**
Within treatments	90	2,644	29.4	
Total	119	27,528		