# SEASONAL GROWTH OF <u>CERATOPHYLLUM DEMERSUM</u> L., <u>ELODEA NUTTALLII (PLANCH) ST.JOHN</u> AND <u>POTAMOGETON PECTINATUS</u> IN EXPERIMENTAL PONDS

Report of a two-month research period at the CABO, September-October 1988

by

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Appendix I-III

#### SAMENVATTING

Dit rapport behandelt de seizoensgebonden groei van drie soorten ondergedoken waterplanten. Het is gebaseerd op gegevens over de eerste drie oogsttijdstippen van het eerste jaar van een experiment waarin groei en hergroei vermogen worden bestudeerd over een tweejarige periode.

De seizoensgebonden groei van vegetatietypen van <u>Ceratophyllum demersum</u> L., <u>Elodea nuttallii</u> (Planch.) St. John en <u>Potamogeton pectinatus</u> L., gekweekt in experimentele vijvers, werd beschreven in relatie tot licht en koolhydraatreserves. De grote verschillen in biomassa ontwikkeld door planten behorend tot dezelfde soort, maar gekweekt in twee verschillende vijvers, werden vooral toegeschreven aan verschillen in de hoeveelheid licht die de planten vroeg in het groeiseizoen bereikte. Van de drie soorten vertoonde <u>P. pectinatus</u> de kleinste seizoensafhankelijke veranderingen in biomassa.

De suikergehaltes van alle planten bedroegen ca. 2 tot 4 % van het asvrij drooggewicht, met een maximum midden in de zomer. Zetmeel was de belangrijkste koolstofreserve bij alle soorten. De zetmeelgehaltes van <u>C. demersum</u> en <u>E. nuttallii</u> namen vroeg in het groeiseizoen sterk toe en stabiliseerden vervolgens op een zomerpeil. De zetmeelgehaltes van <u>P. pectinatus</u> vertoonden een vergelijkbaar patroon als de suikergehaltes en ook de waarden van zetmeelen suikergehalte waren vergelijkbaar.

## SUMMARY

The present report describes the seasonal growth of three species of submerged macrophytes. It is based on data on the first three harvests in the first year of an experiment in which the growth and regrowth potential are studied during a two-year period.

Seasonal growth of <u>Ceratophyllum demersum</u> L., <u>Elodea nuttallii (Planch.)</u> <u>St.John</u> and <u>Potamogeton pectinatus</u> L. stands cultivated in experimental ponds was described in relation to light and carbohydrate reserves. Large differences in biomass formation between plants of the same species but cultivated in two different ponds were mainly explained by differences in light availability early in the growth season. Of the three species, <u>P. pectinatus</u> showed the smallest seasonal changes in biomass.

The sugar contents of the plants of all three species were about 2 to 4 % AFDW, showing a maximum value at mid-summer. Starch was the major carbon reserve in the three species. The starch contents of <u>C. demersum</u> and <u>E. nuttallii</u> increased strongly early in the growth season and stabilized subsequently at the mid-summer level. In case of <u>P. pectinatus</u>, the starch content tended to follow pattern and values of the sugar content.

## INTRODUCTION

Submerged aquatic macrophytes may influence the shallow aquatic ecosystems in which they grow in various ways. They affect the water chemistry as a result of uptake and/or excretion of nutrients, inorganic carbon and organic matter. In agricultural areas macrophytes may also affect the water transport in water courses. In these areas the aquatic vegetation usually consists of a few common species with a high summer biomass, increasing the flow resistance in the water course and causing, consequently, a less efficient land drainage and water supply (Best et al., 1986). Weed cutting is required to prevent these interactions and knowledge of the growth and regrowth potential of the macrophytes would be a useful tool in management practice.

A research programme has been launched by the Centre of Agrobiological Research (CABO), Wageningen, to investigate growth of submerged aquatic plants and their regrowth potential after periodical removal of the aboveground biomass.

In this study growth of aquatic macrophytes is defined as the increase in ash-free dry weight of the plants' structural material. Growth is assumed to be governed by temperature, light or both (Barko & Smart, 1981; Best, 1981) because most nutrients are readily available in water courses in agricultural areas due to crop fertilization. Carbon (CO<sub>2</sub> or bicarbonate), however, may act temporarily as limiting factor, its availability being regulated by inorganic carbon concentration, pH and water velocity. Low concentrations of bicarbonate may occur, for example, during phytoplankton blooms. Because the biomass of plants is composed largely (over 50 %) by carbon, carbon is generally considered a suitable parameter for estimating biomass and for other growth characteristics. Growth occurs when the energy fixed during photosynthesis exceeds the energy required for maintenance of the plant. Excess of energy may be stored as carbon reserves and depends on insolation (50 % photosynthetically active), temperature, plant species and age.

The present report describes the seasonal growth of three species of submerged macrophytes. It is based on data on the first three harvests during the first year of a regrowth experiment with a total duration of two years. The data concern the biomass and carbohydrate reserves of three common submerged macrophyte species.

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### MATERIALS AND METHODS

## Plant material

The regrowth experiment includes 3 different plant species: <u>Ceratophyllum</u> <u>demersum</u> L., <u>Elodea nuttallii (Planch.) St.John</u> and <u>Potamogeton pectinatus</u> L. These species occur frequently in water courses in Dutch agricultural areas.

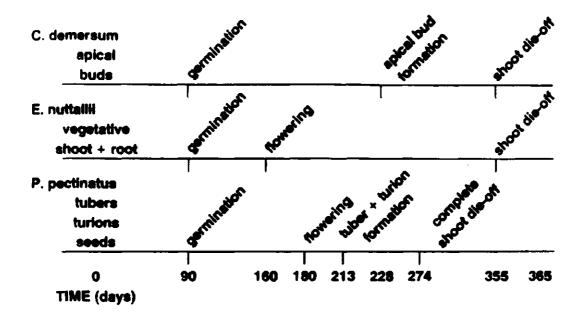


Fig.1. Life cycles of C. demersum, E. nuttallii and P. pectinatus.

The life cycles of the species are of great importance to the understanding of plant growth (Fig. 1). <u>C. demersum perennates</u> by dormant apices. These apices germinate in mid-spring (day 90). The formation of apical buds takes place at the end of the summer (day 228) and dormancy initiates in early autumn. The life cycle ends in the beginning of winter (day 355) with shoot die-off and liberation of dormant apices from the parent plant. In temperate regions flowering does not occur and the species propagates vegetatively.

The life cycle of <u>E. nuttallii</u> starts with new shoot formation by the perennating plants in early spring (day 90). Flowering occurs in the beginning of summer (day 160). The biomass increases during the summer and early autumn. The life cycle ends with shoot die-off in the beginning of winter (day 355).

<u>P. pectinatus</u> plants germinate in early spring (day 90) from tubers, turions or seeds. Flowering takes place in mid-summer (day 180). Formation of storage organs (tubers and turions) occurs in the beginning of autumn (day 213) and the life cycle ends with a complete shoot die-off in mid-autumn (day 274).

## Experimental design and sampling of plant material

For the regrowth experiment, the plants are grown in plastic pots placed on the bottom of ponds of 0.75 m depth at the CABO area, Wageningen. Per species plants are grown in 2 ponds. In each pond 4 blocks of 12 pots are distinguished. Each pot contains one plant sample unit, composed by 12 plants (Appendix I).

Plant samples were harvested at the beginning of May (dayno. 122), at the end of July (dayno. 206) and at the beginning of September (dayno. 248). One half of the samples consisted of merely aboveground parts, as the present data concern the first year of a two-years regrowth experiment of which the plants are completely harvested in the second year. The rest consisted of whole plants and was divided into aboveground and underground parts.

## Measurement of light extinction in the experimental ponds

The light extinction in the ponds was measured biweekly using a relative irradiation meter equipped with Si-cells (manufactured by the Technical Physical Service of Agriculture, Wageningen). Values were read at the water surface and with intervals of 0.1 m down the water column. The vertical extinction coefficient of the ponds was calculated according to Wetzel (1975).

## Plant analyses

#### Biomass

The plant samples were freeze-dried and the dry weight of each sample was measured. Subsequently, the ash content was determined of each sample using quantities of 5-15 mg milled sample. These preweighed samples were placed in an oven for 1 hour at 600 °C. After reweighing, the ash free dry weight of plant samples was calculated.

#### Reserves

The contents of water soluble sugars and starch were measured in 50 % of the samples (two pots of each block in both ponds). The determinations were performed in the chemical laboratory of the Center for Agrobiological Research.

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The analysis was based upon the reducing power of non structural carbohydrates in their ene-diol state. Glucose, mannose, fructose and fructosans were extracted from the plant material in a 40 % ethanol solution, hydrolized into weak acid ( $H_2SO_4$ ) solution and determined as Cu<sup>2+</sup>-complex (Schaffer and Somogyi, 1933 ; AOAC, 1975). Starch was extracted from the pellet of the sugar extract using water, boiling, autoclavation (130 °C, 1 hour, 1.8 atm), hydrolysis by -amylo- glucosidase, clearing using a Carrez solution (Schormüller, 1967) and subsequent determination of the sugars as described.

## Statistics

An analysis of variance was carried out on the data on a) aboveground biomass, b) total biomass and c) contents of reserves in aboveground plant parts. No ANOVA was performed on the data on reserves of total plants because the number of samples was insufficient. Testing considered differences among the two ponds of each species and differences among the data sets of each harvest. The ANOVA package of GENSTAT 5 was used for the statistical analyses on a VAX mainframe. An example of a program run for the analyses is shown in Appendix II.

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### RESULTS

#### Light extinction in the experimental ponds

There were large differences between all ponds in the patterns and values of light extinction coefficients plotted against time (Fig. 2).

In the <u>C. demersum</u> ponds pond 1 was initially more turbid than pond 2, but became more clear from mid-spring onwards. In autumn the turbidity of both ponds was similar.

In the ponds of <u>E. nuttallii</u> an extremely high turbidity occurred in winter in pond 1 which was not observed in pond 2. This difference in turbidity between both <u>E. nuttallii</u> ponds continued until the beginning of summer and both ponds stabilized subsequently at a fairly low ( $\boldsymbol{\varepsilon} = 1.5 \text{ m}^{-1}$ ) light extinction coefficient.

In the ponds of <u>P. pectinatus</u> turbidities were similar until the beginning of summer, except for a peak in pond 2 in spring (day 90). The turbidities in both ponds increased to high values ( $\mathcal{E}$  =3-4 m<sup>-1</sup>) in summer and decreased subsequently in pond 1, but remained high in pond 2.

#### Plant analyses

#### Biomass

The seasonal changes in biomass of the three species are shown in Fig. 3 and Fig. 4. The patterns of total biomass and aboveground biomass, respectively, were similar for all species, in that the highest amount of biomass occurred at the end of the observation period. Interpreting the curves, attention should be paid to the fact, that the standard deviations are not shown into the figures. However, they are indicated in the table of Appendix III. The biomass of <u>C. demersum</u> and <u>E. nuttallii</u> showed a large variance among the ponds, especially at the last harvest. At the summer harvest, the biomass of <u>C. demersum</u> tended to be higher than that of <u>E. nuttallii</u>. <u>P. pectinatus</u> had the lowest biomass throughout the observation period.

The biomass formation of the same plant species varied strongly between the two ponds. In case of <u>C. demersum</u>, the biomass in pond 1 showed a remarkable maximum in mid-summer, while in pond 2 it increased during the whole period. The biomass of <u>E. nuttallii</u> showed similar patterns in both ponds, but in pond 2 it was higher than in pond 1. The biomass of <u>P. pectinatus</u> increased generally in both ponds. The total biomass in pond 1 seemed to decrease in autumn, but this decrease was actually due to the large variation in autumn data.

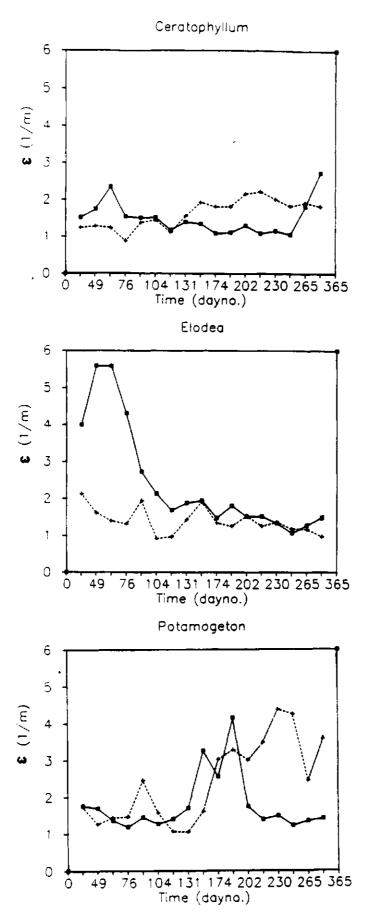


Fig.2. Vertical extinction coefficients on experimental ponds. Pond 1 \_\_\_\_\_\_, pond 2 \_\_\_\_\_.

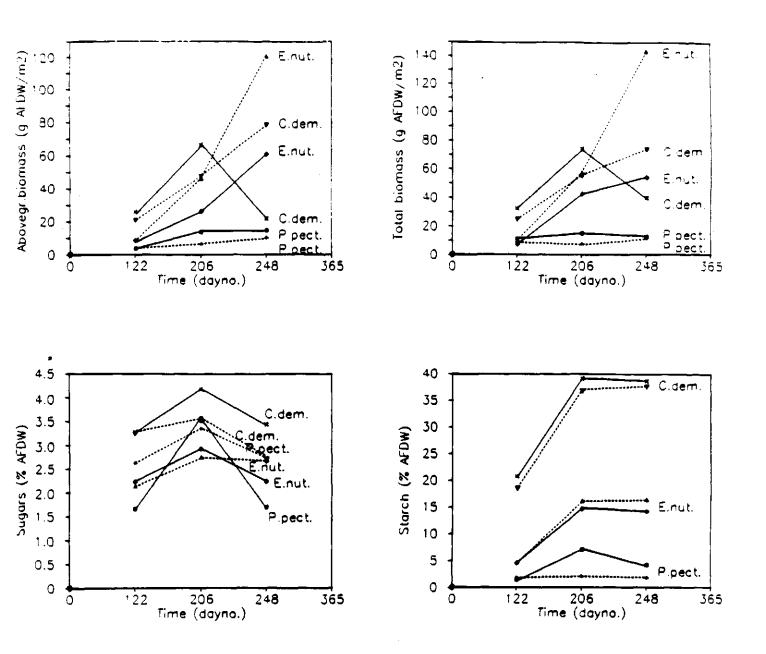


Fig.3. Measured data on seasonal changes in biomass and reserves; comparison of C. demersum, E. nuttallii and P. pectinatus. Pond 1 \_\_\_\_\_, pond 2 \_\_\_\_\_.

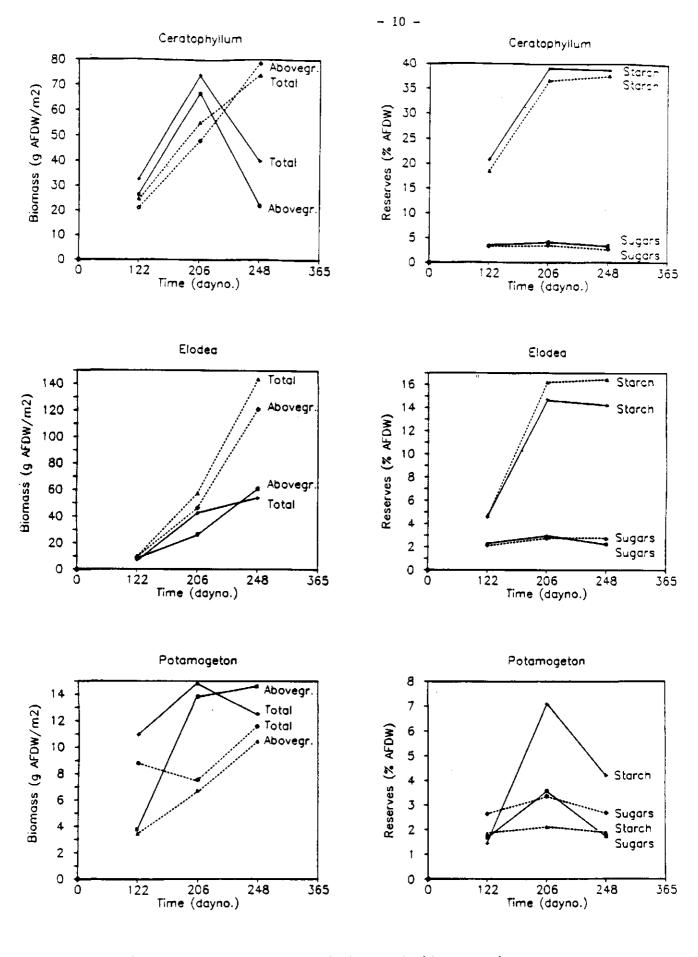


Fig.4. Measured data on seasonal changes in biomass and reserves; comparison of ponds. Pond 1 \_\_\_\_\_\_, pond 2 \_\_\_\_\_.

#### Reserves

The seasonal changes in carbohydrate reserves of the three species are shown in Fig. 3 and Fig. 4.

The general pattern of the sugar contents was similar in all three species: the content increased during spring and in the beginning of summer. In mid-summer a maximum occurred and during autumn the sugar reserves decreased.

<u>C. demersum</u> had generally higher sugar contents than both other species. The sugar values of <u>E. nuttallii</u> and <u>P. pectinatus</u> showed a large variation and, consequently, it was not possible to decide which species had the higher content. The sugar contents of <u>C. demersum</u> plants in pond 1 were higher than those in pond 2. Those of <u>E. nuttallii</u> plants were similar in both ponds until mid-summer but were higher in mid-autumn in pond 2 than in pond 1. <u>P. pectinatus</u> showed a large variation in sugar contents among both ponds. The contents of the plants in pond 1 had the largest seasonal changes, but those of pond 2 were higher in spring and autumn than those of pond 1.

The starch reserves varied strongly between the three species. <u>C. demersum</u> showed starch contents increasing from 20 % AFDW in spring to 40 % AFDW in mid-summer. The starch contents of plants in pond 1 were higher than those in pond 2. Yet, the plants of both ponds followed similar patterns. The starch contents of <u>E. nuttallii</u> changed from 5 % AFDW in spring to 15 % in mid-summer. The plants in pond 2 had the highest starch contents. In both, <u>C. demersum</u> and <u>E. nuttallii</u> the mid-summer and autumn values were similar. <u>P. pectinatus</u> showed a maximum starch content in summer of at the highest 8 % AFDW. The summer and autumn starch contents of pond 1 were at least twice as high as those of pond 2.

## Statistics

The results of the statistical analyses on the measured data are listed in Table 1.

No significant difference was found among the <u>C. demersum</u> ponds at the 1 % significance level, nor at the 5 % significance level. However, the starch data showed a small trend of diffence (Fprob=0.122). It was not possible to perform any analysis on data on aboveground biomass, because the plant samples were not always divided into aboveground parts and underground parts. The test on difference among the data set of the harvests confirmed a significant seasonal change in starch content on the 1 % significance level. At this level no difference was found for other parameters. All parameters showed a significant seasonal change, when the test was performed at the 5 % level.

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In case of <u>E. nuttallii</u>, the ponds did not differ at the 1 % significance level. A significant difference occurred among data on aboveground biomass, when the test was performed on the 5 % significance level. The other parameters showed a tendency to differ among ponds. Seasonal changes of all parameters were found to be significant at both significance levels.

There was no difference among the ponds of <u>P. pectinatus</u> at the 1 % significance level. A significant difference occurred among the data on aboveground - and those on total biomass, when the test was performed at the 5 % level. The reserve contents tended to differ among the ponds (sugar: Fprob=0.148; starch: Fprob=0.056). Significant seasonal changes were only seen for the data on aboveground biomass (5 % and 1 % levels).

Table 1. H<sub>0</sub>-hypotheses and Fprob. factors from analyses of variance. The values could not be calculated on the aboveground biomass of <u>C</u>. <u>demersum</u>, because the plant samples were not always divided into aboveground parts and underground parts (¥). hl, data from spring harvest; h2, summer harvest; h3, autumn harvest.

H <sub>0</sub> -hypkhesis	Abovegr.		Sugars	Starch
	biomass n = 48		n = 24	n = 24
	Fprob.	Fprob.	Fprob.	Fprob.
C. demersum				
pond 1 = pond 2	ł	0.738	0.996	0.122
hl = h2 ≖ h3	Ħ	0.017	0.012	0.002
E. nuttallii				
pond $1 = pond 2$	0.019	0.054	0.071	0.135
h1 = h2 = h3	<0.001	0.002	<0.001	< 0.001
P. pectinatus				
pond 1 = pond 2	0.014	0.024	0.148	0.056
h1 = h2 = h3	<0.001	0.411	0.083	0.087

## DISCUSSION

## Relations between growth and light

Seasonal biomass production of <u>C. demersum</u> was higher in pond 2 than in pond 1. The plants in pond 1 showed a remarkable decrease in biomass at the end of summer and early autumn. However, the difference among the ponds was not statistically significant. Comparing the light extinction coefficient in the two ponds pond 1 seemed to have a higher light availability than pond 2. This discrepancy between available light and biomass formation in both ponds might be explained by a large amount of filamentous algae observed on the plants in pond 1. Consequently, the plants of pond 1 in reality had less light available than those in pond 2 and were not able to produce enough energy to maintain their standing crop.

The light conditions in the <u>E. nuttallii</u> ponds were quite different during the first half of the growth season (May to July) in that pond 1 usually had the highest light extinction coefficient. The biomass formation in pond 1 lagged behind that of pond 2 during the same period, assumingly because of lower light availability. The difference between the biomass formation in the ponds was even more manifest at the end of the observation period but this time it could not be explained by differences in the under water light climate.

In reality, the light extinction coefficients and, therefore, light availability was similar in both ponds during the second half of the growth season (July to October). Thus, shading by filamentous algae might be an explanation, but the plants in both ponds suffered from these and it was not possible to decide which pond had the most serious attack. A more likely explanation would be that the development of the plants in pond 1 started somewhat later, than in pond 2. Consequently, the development state (Best & Visser, 1987) of the pond 1 plants lagged behind that of the pond 2 plants, providing little opportunity to reach similar levels of produced biomass at the end of the growth season.

The biomass of <u>P. pectinatus</u> in both ponds differed at the 5 % significance level with pond 1 having the highest production. No filamentous algae were found in these ponds. Thus, the difference in growth pattern should be explained only by the differences in light extinction coefficients of the water column. The light extinction coefficient of pond 2 did exceed that of pond 1 only in spring (day 90) and from late summer onwards. Particularly spring is critical to the plants because at that time the growth season starts when the increasing water temperature initiates germination, stimulates respiration and the plants are developing shoot material to capture available light. The high water turbidity in pond 2 in spring decreased the light available to the plants at that critical moment and, therefore, probably caused the observed lag in biomass formation.

#### Relations between growth and reserves

Starch was generally the major carbon reserve in the three species and its content showed a large seasonal variation. The sugars remained at a quite low percentage of the ash free dry weight throughout the growth season. All species showed higher sugar values in summer than in spring and autumn, possibly due to the fact that the respiration, the main consumer of sugars, decreased in these periods due to lower temperatures.

The <u>C. demersum</u> and <u>E. nuttallii</u> plants used the major part of the energy fixed by photosynthesis during the first half of the growth season to build up their starch reserves. Later on, these reserves were maintained and the excess of energy was used primarily for biomass production. In case of <u>P. pectinatus</u>, this did not occur. In this species the starch content showed a similar trend as the sugar content with a maximum in mid-summer. However, the reserve values listed here were measured in the aboveground plant parts and knowing that <u>P. pectinatus</u> forms tubers too, it would be relevant to investigate the reserves in these organs. At present, there are not enough data on the underground plant parts to draw any conclusions, but the few available data showed a tendency to higher (about 8 % AFDW) starch contents than those of the aboveground parts. On the other hand, the growth season of <u>P. pectinatus</u> is shorter than that of both other species (Fig.1) and, consequently, the reason for the maximum starch content in mid-summer might indicate initiation of decay of the plants at that moment.

## ACKNOWLEDGEMENTS

F.H.H. Jacobs, W. van der Zweerde and J.M. Braber are acknowledged for their technical assistance. The employees of the chemical department provided help in the chemical analyses. Finally, I feel indebted to E.P.H. Best for coaching and for critical reading of the manuscript and linguistic corrections.

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APPENDIX I

OPZET EN STATISTISCHE VERANTWOORDING EFFECTEN AARD EN FREQUENTIE MECHANISCH ONDERHOUD OP DE HERGROEI VAN SUBMERSE WATERPLANTEN

1. <u>Doel</u>

- a. opname groeicurve jaar 1,
  opname groeicurve jaar 2,
  bepaling potentiële hoeveelheid gemaaide biomassa jaar 1
  bepaling potentiële hoeveelheid biomassa jaar 2
  dit alles aan de hand van oogstmomenten overeenkomend met de maaimomenten van het maai-experiment zelf
- b. maai-experiment: effect maaitijdstip op potentiële hergroei principe: opleggen van maaibeheer in jaar 1 (v, z, n, l.n.) en oogst op normale maaitijdstippen jaar 2 onderzochte factor: maaitijdstip

## 2. Opzet

Lokatie: 1 (CABO-terrein) Aantal vijvers: 6; per soort 2; elke vijver te beschouwen als een afzonderlijk experiment. Per vijver: 4 blokken Per blok: 12 bakjes (3 rijen van 4, per blok volledig geloot; voor loting dus 24 reeksen van 12 bakjes opgegeven; hele proef uitgevoerd in viervoud: elk blok in dezelfde vijver is een herhaling) Totaal aantal bakjes per vijver: 48 Totaal aantal bakjes per soort: 96

## 3. Verklaring codering bakjes

J101	Jaarl	Oogstmoment	1
J102	Jaarl	Oogstmoment	2
J103	Jaarl	Oogstmoment	3
J104	Jaarl	Oogstmoment	4

J201	Jaar2	00	gstmoment	1	
J202	Jaar2	005	gstmoment	2	
J203	Jaar2	008	gstmoment	3	
J204	Jaar2	008	gstmoment	4	
H01	Hergroe	ei	Oogstmome	ent	1
H02	Hergroe	ei	Oogstmome	ent	2
H03	Hergroe	ei	Oogstmome	ent	3
н04	Hergroe	ei	Oogstmome	ent	4

*	Jaar	1	1988
	Jaar	2	1989
	Oogsti	momentl	begin mei
	Oogsti	moment2	eind juli
	Oogst	moment3	begin september
	Oogsti	moment4	eind november

# 4. Groepering bakjes in blokken, vanaf punt x

Pot.1			Pot.2		
н 04	J102	J201	J201	J202	н 04
H 01	J204	H 03	J101	H 01	Н 02
J104	J202	J103	Н 03	J102	J203
H 02	J203	J201	J204	J104	J103
J103	H 01	J102	Н 02	H 04	н ОЗ
J202	J101	Н 04	J201	J102	J204
H 02	J203	J201	H 01	J101	J202
J204	H 03	J104	J104	J203	J103
J201	J202	H 03	J202	H 04	J203
J204	J102	J101	J103	H 02	H 03
J104	Н 02	н 01	J101	H 01	J104
J103	J203	н 04	J204	J201	J102
H 04	J203	J102	н 03	J103	Н 01
J103	J104	H 01	J104	НО	J201
J101	H 02	Н 03	J101	J102	J203
J201	J204	J202	J202	H 02	J204
	x			x	

**I-**2

E1.1			E1.2		
J103	J204	J202	J101	J103	н 04
J101	н 04	H 01	H 02	J104	J201
н 03	J201	J203	J102	J203	н 03
J102	J104	Н 02	J204	J202	н 01
J204	J102	н 01	J203	J204	J103
J201	J103	J104	J202	J201	J101
H 04	J101	H 03	н 01	J102	J104
H 02	J202	J203	H 02	н 04	H 03
H 02	Н 04	J204	H 04	J202	H 01
J203	J104	J101	J201	J204	н 03
J103	J201	J202	J203	J103	J101
H 01	J102	Н 03	J102	Н 02	J104
J103	J203	J204	H 02	J203	J201
H 01	J104	J102	J101	J204	н 03
H 02	H 03	H 04	J102	J104	н 01
J202	J201	J101	J202	J103	н 04
	37				
	x			x	
<u>Cer.l</u>			Cer.2		
		J204			J101
	J101			J202	
H 04 J203	J101	J202	H 03	J202 J201	Н 02
H 04 J203 J201	J101 H 01 H 02	J202	H 03 J203 J102	J202 J201 J103	H 02 J204
H 04 J203 J201 J104	J101 H 01 H 02 H 03	J202 J103 J102	H 03 J203 J102 H 01	J202 J201 J103 H 04	H 02 J204 J104
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H 04 J203 J201 J104 J202 J101 H 02	J101 H 01 H 02 H 03 J104 H 03 J201	J202 J103 J102 J203 J103	H 03 J203 J102 H 01 J203 J104 H 02	J202 J201 J103 H 04 J202 J101	H 02 J204 J104 H 03 J103 H 04
H 04 J203 J201 J104 J202 J101 H 02 J204	J101 H 01 H 02 H 03 J104 H 03 J201 J102	J202 J103 J102 J203 J103 H 04 H 01	H 03 J203 J102 H 01 J203 J104 H 02 J102	J202 J201 J103 H 04 J202 J101 H 01 J201	H 02 J204 J104 H 03 J103 H 04 J204
H 04 J203 J201 J104 J202 J101 H 02 J204 J104	J101 H 01 H 02 H 03 J104 H 03 J201 J102 J201	J202 J103 J102 J203 J103 H 04 H 01 J102	H 03 J203 J102 H 01 J203 J104 H 02 J102 H 04	J202 J201 J103 H 04 J202 J101 H 01 J201 H 02	H 02 J204 J104 H 03 J103 H 04 J204
H 04 J203 J201 J104 J202 J101 H 02 J204 J104 J202	J101 H 01 H 02 H 03 J104 H 03 J201 J102 J201 H 03	J202 J103 J102 J203 J103 H 04 H 01 J102 J103	H 03 J203 J102 H 01 J203 J104 H 02 J102 H 04 J204	J202 J201 J103 H 04 J202 J101 H 01 J201 H 02 J101	H 02 J204 J104 H 03 J103 H 04 J204 J202 J104
H 04 J203 J201 J104 J202 J101 H 02 J204 J104 J202 J101	J101 H 01 H 02 H 03 J104 H 03 J201 J102 J201 H 03 H 02	J202 J103 J102 J203 J103 H 04 H 01 J102 J103 H 01	H 03 J203 J102 H 01 J203 J104 H 02 J102 H 04 J204 J201	J202 J201 J103 H 04 J202 J101 H 01 J201 H 02 J101 J103	H 02 J204 J104 H 03 J103 H 04 J204 J202 J104 H 03
H 04 J203 J201 J104 J202 J101 H 02 J204 J104 J202 J101	J101 H 01 H 02 H 03 J104 H 03 J201 J102 J201 H 03 H 02	J202 J103 J102 J203 J103 H 04 H 01 J102 J103 H 01	H 03 J203 J102 H 01 J203 J104 H 02 J102 H 04 J204	J202 J201 J103 H 04 J202 J101 H 01 J201 H 02 J101 J103	H 02 J204 J104 H 03 J103 H 04 J204 J202 J104 H 03
H 04 J203 J201 J104 J202 J101 H 02 J204 J104 J202 J101 J203	J101 H 01 H 02 H 03 J104 H 03 J201 J102 J201 H 03 H 02 H 04	J202 J103 J102 J203 J103 H 04 H 01 J102 J103 H 01 J204	H 03 J203 J102 H 01 J203 J104 H 02 J102 H 04 J204 J201 H 01	J202 J201 J103 H 04 J202 J101 H 01 J201 H 02 J101 J103 J203	H 02 J204 J104 H 03 J103 H 04 J204 J204 J202 J104 H 03 J102
H 04 J203 J201 J104 J202 J101 H 02 J204 J104 J202 J101 J203 H 01	J101 H 01 H 02 H 03 J104 H 03 J201 J102 J201 H 03 H 02 H 04 J102	J202 J103 J102 J203 J103 H 04 H 01 J102 J103 H 01 J204 H 03	H 03 J203 J102 H 01 J203 J104 H 02 J102 H 04 J204 J201 H 01 H 03	J202 J201 J103 H 04 J202 J101 H 01 J201 H 02 J101 J103 J203 J201	H 02 J204 J104 H 03 J103 H 04 J204 J204 J202 J104 H 03 J102 J104
H 04 J203 J201 J104 J202 J101 H 02 J204 J104 J202 J101 J203 H 01 J101	J101 H 01 H 02 H 03 J104 H 03 J201 J102 J201 H 03 H 02 H 04 J102 H 04	J202 J103 J102 J203 J103 H 04 H 01 J102 J103 H 01 J204 H 03 J204	H 03 J203 J102 H 01 J203 J104 H 02 J102 H 04 J204 J201 H 01 H 03 J204	J202 J201 J103 H 04 J202 J101 H 01 J201 H 02 J101 J103 J203 J201 H 02	H 02 J204 J104 H 03 J103 H 04 J204 J204 J202 J104 H 03 J102 J104 H 01
H 04 J203 J201 J104 J202 J101 H 02 J204 J104 J202 J101 J203 H 01 J101 J202	J101 H 01 H 02 H 03 J104 H 03 J201 J102 J201 H 03 H 02 H 04 J102 H 04 J102	J202 J103 J102 J203 J103 H 04 H 01 J102 J103 H 01 J204 H 03 J204 J203	H 03 J203 J102 H 01 J203 J104 H 02 J102 H 04 J204 J201 H 01 H 03	J202 J201 J103 H 04 J202 J101 H 01 J201 H 02 J101 J103 J203 J201 H 02 H 04	H 02 J204 J104 H 03 J103 H 04 J204 J204 J204 H 03 J102 J104 H 01 J101

APPENDIX II

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Programme used for statistical analysis on total biomass data. The data in the
present version are on P. pectinatus.
JOB 'PROJECT 717'
UNIT [NVALUES=24]
VARIATE TOTBIOMASS
FACTOR [LEVELS=2] POND ; VALUES=!V(12(1,2))
FACTOR [LEVELS=8] REPL ; VALUES=!V((1...4)3,(5...8)3)
FACTOR [LEVELS=3] TREAT ; VALUES=!V(4(1,2,3),4(1,2,3))
READ TOTBIOMASS
29.27 32.05 32.04 36.07 95.48 80.60 48.78 71.49 29.43 51.66
50.22 27.03 24.26 25.25 26.05 22.36 45.91 58.38 51.98 64.78
36.47 108.12 103.16 47.50 :
PRINT POND, REPL, TREAT, TOTBIOMASS
MATRIX [ROWS=!T('JAAR 1', 'JAAR 2');COLUMNS=3] \
GROUP; !V(−5, 1, 4, ∖
           1,-3, 2)
BLOCKS REPL
TREATMENTS POND + REG(TREAT;2;GROUP)
ANOVA [CONTR=2; FPROB=YES] TOTBIOMASS
STOP
```

APPENDIX III

## Plant analyses

For above ground biomass n = 48 ; for other parameters n = 24 .

Time (dayno.)	-	Abovegr.biomass (g AFDW/m <sup>2</sup> )				Ū.		Starch (% AFDW)	
	mean	SD	mean	SD	mean	SD	mean	SD	
C. demers	<u>um</u>								
pond 1									
122	26.10	9.67	32.36	2.80	3.28	0.15	20.74	4.81	
206	66.77	15,05	74.09	19.56	4.18	0.48	29.16	3.66	
248	21.71	6.02	39.59	13.16	3.43	0.50	38.72	1.46	
pond 2									
122	20.78	4.50	24.48	1.59	3.24	0.14	18.48	0.56	
206	47.70	11.42	55.26	8.14	3.51	0.39	36.78	4.03	
248	78.45	31.26	73.81	37.08	2.70	0.24	37.64	8.22	
E. nuttal pond 1	<u>lii</u>								
122	8.5	4.29	7.10	3.86	2.23	0.35	4.57	1.66	
206	26.59	10.26	42.47	10.03	2.92	1.04	14.72	1.96	
248	60.78	28.19	54.02	34.52	2.23	0.70	14.22	2.69	
pond 2									
122	8.93	2.12	9.62	2.87	2.14	0.27	4.69	1.03	
206	46.40	11.18	57.34	16.10	2.75	0.11	16.21	1.88	
248	120.89	49,78	143.71	76.21	2.71	0.62	16.45	2.19	
P.pectina	tus								
pond l									
122	3.77	2.63	10.95	2.45	1.66	0.40	1.43	0.60	
206	13.91	4.10	14.96	3.21	3.56	1.93	7.09	4.26	
248	14.58	7.85	12.40	4.01	1.69	1.13	4.19	1.90	
pond 2									
122	3.52	2.76	8.81	1.83	2.63	1.10	1.83	0.22	
206	6.68	2.01	7.56	1.33	3.35	0.64	2.10	1.19	
248	10.40	1.84	11.60	2.77	2.65	2.86	1.83	0.67	