A COMPARATIVE STUDY OF CONSUMPTION RATES AND PREFERENCE FOR SOME SPECIES OF AQUATIC PLANTS BY Tilapia rendalli.

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ABSTRACT

The daily consumption rates and preference of juvenile <u>Tilapia</u> <u>rendalli</u> for some macrophytes, <u>Ceratophyllum</u> <u>demersum</u>, <u>Lagarosiphon major</u>, <u>Najas pectinata</u> and <u>Valisheria aethiopica</u> were determined. Fish were offered single macrophyte diets to determine daily consumption and a mixture of the four macrophytes in equal quantities to to determine selection. Consumption rates were 821.50mg, 829.05mg, 940.00mg and 2 293.53mg per fish per day respectively. The difference in consumption rates were significant. Preference was shown for <u>V. aethiopica</u>, whilst <u>C. dermersum</u> was least selected. Fish fed on single species lost weight whereas those fed on a variety of macrophytes gained in weight.



2. INTRODUCTION

Extensive work related to feeding has been done on the herbivorous fish, <u>Tilapia rendalli</u> (Munro 1965, Junor 1969, Wager and Rowe-Rowe 1972, Batchelor 1978, Caulton 1976, 1976a, 1977, 1977a, 1978, Caulton and Bursel 1977). Daily consumption rates were worked out for laboratory fish fed on <u>Ceratophyllum demersum</u> (Caulton 1976) and also for fish feeding on <u>Panicum repens</u> in the wild (Caulton 1977). Rate of consumption and preference for different macrophytes have not been compared.

Junor (1969) reported on the detrimental effects of T. rendalli destructive feeding on aquatic plants. This demonstrated potential effectiveness of the fish in controlling aquatic weeds. However, in some situations where <u>I.</u> rendalli is present the fish is unable to control submerged weed, for example in Lake Robertson (Chimbuya, personal communication). Food selection studies could explain such a situation. Knowledge on selection and consumption rates is important since these parameters determine effectiveness of control and stocking densities required for effective control. Some preliminary work of this nature was carried out on a grass carp Ctenopharyngodon idella) and some grass carp hybrid (Stoff & Orr 1970, Edwards 1974, Mitchell 1977, 1980, Mitzner 1978, Colle <u>et</u> al 1978, Flowler and Robson 1978, Cassani 1981, Shireman et al 1978, 1983, Young et al 1983, Harberg and Moddle 1985).

The process that relate to food consumption, appetite, food kind, amount consumed and frequency of consumption, digestibility and rate of movement through gut, absorption of nutrient and conversion efficiency determine growth and production of any fish population. Estimates on these consumption rates can be related to food abundance in the wild. Caulton (1977) made a quantitative assessment of daily ingestion of <u>Panicum repens</u> by <u>T. rendalli</u> and used the ingestion rates to calculate biomass of fish that can be supported during annual flooding of grass on the shore of Lake Kariba.

In fish that have a varied diet like <u>T. rendalli</u> there are basically two factors that determine quantities of individual food items in the diet. These are abundance of food item and selection of some items by the fish (Mwebaza-Ndawula 1984, Horn <u>et al</u> 1982, Prejs and Prejs 1987). <u>T. rendalli</u> can change from being completely herbivorous to partially carnivorous depending on availability of macrophytes (Junor 1969). Plants are selected according to different palatability and nutritive qualities (Westoby 1974, 1977). There is, for example, a tendency for many temperate and tropical fishes not to eat calcareous rhodophytes (coralline algae) and phacophytes (Horn <u>et al</u> 1982). They also found that coralline algae tend to have a high proportion of structural material and relatively low carolic value and hence may be relatively unpalatable and low in the nutritive value. Phacophytes may be avoided for their tough thallus and poly-phenolic compounds (Horn <u>et al</u> 1982).

Energy and caloric value and protein content are regarded as the most important components in determining nutritive value (Westoby 1974, Mattson 1980). Plants with either or both high energy and protein content are favoured. Nitrogen requirement of animals in generaly higher than that of plants because animals use proteins for structural building blocks whereas plants use carbohydrates. Moreover, animals use nitrogen less efficiently because a significant fraction of their daily waste consists of various N-compounds whereas plants ecxrete little nitrogen. Because animals contain 8-14% dry weight more nitrogen than plants (Mattson 1980) it is essential that a herbivore consumes plants with the highest concentration possible.

Plants vary in thier nutritive status and this would determine how much of that plant is consumed as much as which ones are selected as discussed above. When fed on a variety of diets the rate of feeding is such as to maintain a relatively constant energy intake (Jobling 1980, 1987). Consumption rate in this case is determined by the rate of gastric emptying which tend to be faster when dilute meals are fed (Jobling 1980, 1987, Flowerdew & Groove 1979, Groove at al 1978). There is therefore some compensation by increasing consumption rate when nutritional quality of food is low. However limits are set to this form of compensation by the physiological maximum for digestion. Maels diluted with kaolite were evacuated more rapidly (Flowerdew & Groove 1979, Groove <u>et al</u> 1979) causing a feeling of hunger enabling the fish to consume more.

The objectives of the present study were to compare consumption rates and to determine selectivity of <u>T</u>. <u>rendalli</u> fed on four different macrophytes, <u>Ceratophyllum</u> <u>demersum</u>, <u>Lagarosiphon major</u>, <u>Valisneria aethiopica</u> and <u>Najas pectinata</u>. Selection under experimental aquaria condition was compared with selection in the wild deduced from stomach content analysis. Nitrogen content, total organic matter and water content of plants was measured to determine if there was any relationship between plant selection, daily consumption rates and these parameters. Weight changes and condition of the fish for the different trials were also compared.

3 MATERIALS AND METHODS

3.1 Maintenance of experimantal tanks

Feeding trials were carried out in four metal and one glass aquarium/fish tanks of volumes 1.19m and 0.35m respectively. The water level was kept 18cm below the brim in all tanks to prevent fish jumping out. Metal tanks were painted to prevent rusting. All tanks were housed under a corrugated iron roofed shed such that there was no direct sunlight on the water.

The four metal tanks were connected with water pipes. In tank four there was an inlet equiped with a floating ball valve. The water would flow through the system and exit in tank 1 (Fig. 1). In addition each tank had a separate water outlet.

The bottom of the metal tanks were covered with sand. Each tank had a glass window 49×84 cm for veiwing fish. In order to simplify cleaning there was no sand in the glass aquarium (tank 5). However, the glass aquarium was fitted with an aerator, type RENA 301.

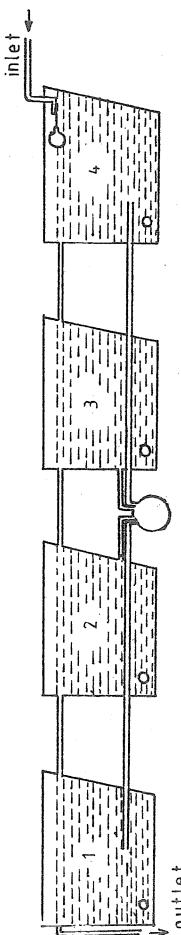
Initially water was slowly flushed out using the through flow system for the metal tanks (Fig. 1). When this method proved unsatisfactory the water was left standing and only changed when oxygen level dropped to between 10 and 15% saturation. Tanks were then drained to one- fifth of initial volume and refilled with tap water. The temperature, dissolved oxygen and pH were monitored throughout the experiment period. A YS1 model 58 dissolved oxygen meter was used to measure both dissolved oxygen and temperature and for pH a Hach digital pH was used. Readings were taken each day between 1300 and 1330 hours.

3.2 Stocking of Aquaria

Tanks were stocked with fish from the wild and a Prawn Farm in Kariba. Beach seining was employed to capture fish from the wild. Ideally fish were to be collected from the wild, but to facilitate rapid stocking fish were obtained from the All the fish from the farm however had fungal Prawn Farm. infection so were treated with 1 ppm malachite green for an hour before stocking. (Recommended repeated treatment was unnecessary since fish showed signs of improvement). After this treatment fish were individually weighed and the standard length measured. Twenty-six fish were introduced into each of the four metal tanks. The size range in the tanks was from 7.7 to 10.5 cm. Tank 5 was stocked with a size range of 5.6 to 11.7 cm. The fish were acclimatized for twenty days, during which period they were fed with the macrophytes to be used in the trials.



Diagram shoving vater-flow system on fish tanks 1 to 4 FFG 1.



outlet

o Individual outlets

During the acclimatization period an attempt to use the through flow resulted in 100% mortality in tank four. The tank was restocked with fish from the wild. An attempt to flush water from tank 5 resulted in 90% mortality and the tank was restocked with fish from the wild. There were not enough large fish to go into the later stocking hence fish with a larger size range were used. Tanks 4 and 5 ended up with fish 5.6 to 10.6 cm and 5.0 to 11.7 cm respectively.

3.3 Feed and feeding schedule

Feeding trials were carried out using four types of macrophytes, <u>Ceratophyllum demersum</u> (hornwort, coontail), <u>Najas pectinata</u> (saw weed), <u>Lagarosiphon major</u> and <u>Valisneria aethiopica</u> (eel grass, wild celery). To compare the daily consumption rates, the fish were fed only one type of macrophyte in tanks 1 to 4. Treatments were allocated at random such that tanks 1, 2, 3 and 4 had <u>L. demersum</u>, <u>L. major</u>, <u>N. pectinata</u> and <u>V. aethiopica</u> respectively. In tank five all four macrophytes in approximately the same weight were presented to determine selection of macrophytes by the fish.

Macrophytes for feeding fish were collected as required from the lake and stored in 5.4m concrete ponds. Initially all plants were collected by SCUBA divers, but later except for <u>V. aethiopica</u> a boat anchor and hooks were used to pull out plants. Only the stem with leaves were collected except for <u>V. aethiopica</u> where roots were necessary in improving keeping quality.

For the first fifteen days food was weighed and presented to the fish from 0730 hours and removed and weighed again at 1800 hours. The length of the feeding period was approximately equals to the length of time the fish feeds in the wild (Caulton 1976). On the later half material was left the previous day was removed and weight at 0700 hours and new materials added.

The plant material was washed thoroughly under running water to remove any attached material, particularly snails. Macrophytes with a visible layer of epiphytes were not used. After this cleaning, <u>V. aethiopica</u> was clipped to remove excess roots, (some roots and stem were neccessary for holding plants onto the feeding trays). As much water as possible was removed by blotting for <u>V. aethiopica</u> and tossing in a feeding try until water stopped dripping from the other plants. Material was then weighed to the nearest gramme using a Mettler PC 8 000 digital balance. The materials were then secured onto the feeding trays which ensured that macrophytes remained submerged and in upright position. The trays also simplified retrieval of remaining feed. In the daily comsumption trials fish were presented with excess feed material. For the selection trial between 25 and 30g of each plant were used.

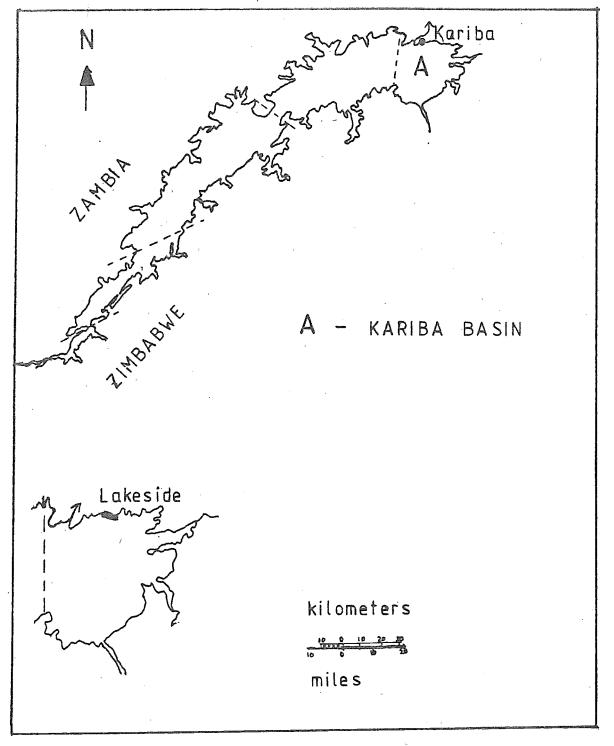
3.4 Stomach content analysis

Stomach content of fish caught all over Kariba basin (Fig.2) were used. Fish were either caught from Lakeside, (Fig.2 shaded part), using gill nets or in other parts by spear fisherman. The stomachs were preserved in 10% formalin for later analysis. All fish were analysed within four weeks of capture. For analysis the stomach contents were diluted with water. A dissecting microscope was used when necessary otherwise materials were identified by unaided visual inspection. All plant species and food items were recorded. From these the percentage occurrence of each food was calculated as follows:

% Occurrence = <u>Number</u> of stomachs containing food item x 100 Total number of stomachs analysed

- 3.5 <u>Condition factor</u> Relative condition factor was calculated using the equation $Kn = \frac{1}{4L^2}$ where a is a constant and b is a power in the length-weigth relationship; W = aL⁶ as given by Le Cren (1951).
- 3.6 Determination of chemical composition of macophytes The water content and organic matter for the different macrophytes were worked out after drying plants in a 100°C oven to constant weight and incernarating at between 450°C and 500°C to constant weight. Protein content was worked out from Kjeidahl nitrogen (Appendix 1) using the factor of 5.1 as given by Grondzinski <u>et al</u> (1975) for leaf protein. All assay were performed in triplicate.

FIG. 2 MAP OF LAKE KARIBA SHOWING KARIBA BASIN AND LAKESIDE WHERE FISH FOR STOMACH CONTENT ANALYSIS WERE CAUGHT.



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4. RESULTS

4.1 Water quality

There was no correlation between daily ingestion of macrophytes and the fluctuation in oxygen, temperature and pH (Table 1). Appendix 2 shows the mean oxygen, temperature and pH in the tanks during the experiment.

4.2 <u>Comparison of daily consumption</u>

The quantity of the food taken increased throughout the experiment period (Fig. 3-6). There was significant difference (p < 0.001, ANOVA) between the quantities of each plant consumed. Fish fed on <u>V. aethiopica</u> consumed more than the other fish (Fig. 7). The material was eaten at the following mean rates; 2 293.53mg, 829.05mg, 821.25mg and 940mg per fish per day for <u>V. aethiopica</u>, <u>L. major</u>, <u>C. demersum</u> and <u>N. pectinata</u> respectively.

4.3 Macrophyte selection

Results of the selection trial are given in Fig. 8. There was a significant difference (p < 0.001, ANOVA) between the mean weight of each macrophyte consumed. Fish preferred <u>V</u>. aethicpica, <u>N</u>, pectinata, <u>L</u>. major and <u>C</u>. demensum in that order.

4.4 Stomach content analysis

Percentage occurrence of food items is given in Fig. 9. Considering stomachs containing aquatic plants only the plants used in the present study occurred in the following proportions:

<u>v.</u>	aethiopica	35.9%	L. major	38.5%
Ν.	<u>pectinata</u>	25.6%	<u>C. demesum</u>	0.0%

There was significant difference (p < 0.001, chi-square) between what is consumed in the wild and what was selected in the selection trial.

4.5 Chemical Composition .

The results of the chemical analysis are given in Table 2. The percentage organic matter , ash and N content of plants differed significantly (P < 0.01, ANOVA) and that of water differed at p < 0.05, ANOVA.

4.6 Growth and condition factor

Length and weight measurements were taken from fish and gave the regression formula $W = 2.826L^{2.570}$. The initial and final weight, length and condition are given in Table 3. All fish fed individual macrophytes lost both weight and condition. Only fish in tanks 1, 3 and 4 increased in length.

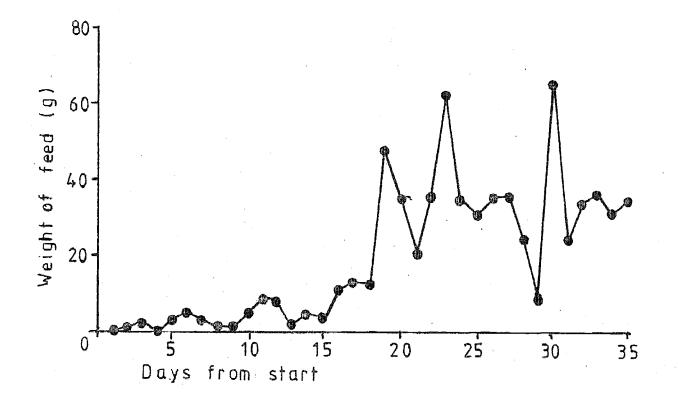
4.7 General Observations

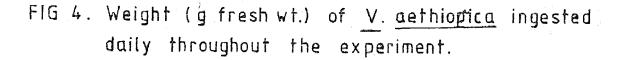
Some fish died soon after stocking so that and tank 1 to 5 had 24, 21, 22, 17 and 26 fish respectively. Fish did not feed during acclimatization period and feeding only commenced as soon as fish showed signs of feeding. More fish were observed feeding in the morning than other parts of the day (Appendix 3). Only leaves were taken from the plants presented to the fish, even when the stem is available except for <u>N. pectinata</u> where stem was occasionally included.

Table 1 : r - values for correlation between food taken (FT) and pH temperature (T) and Dissolved Oxygen (DO)

FT vs T	FT vs DO	FT vs pH	T vs DO
0.048	0.169	-0.299	0.784
0.048	0.049	0.317.	0.629
0.025	0.363	0.480	0.594
0.265	0.189	-0.087	0.684
-0.135	-0.148	0.636	-0.149
	0.048 0.048 0.025 0.265	0.0480.1690.0480.0490.0250.3630.2650.189	0.048 0.169 -0.299 0.048 0.049 0.317. 0.025 0.363 0.480 0.265 0.189 -0.087

FIG 3. Weight (g fresh wt.) of <u>C. demersum</u> ingested daily throughout the experiment.





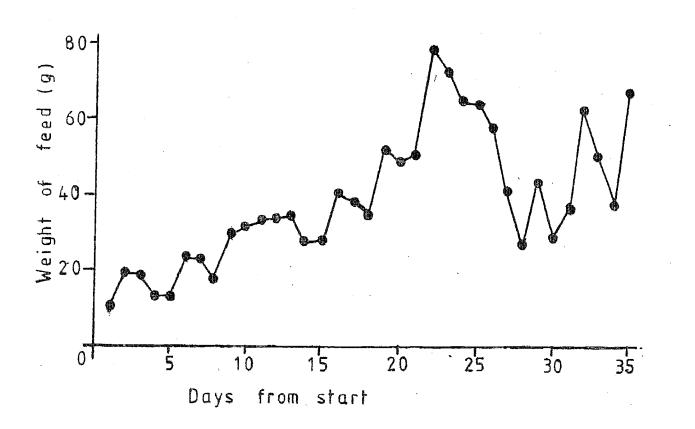
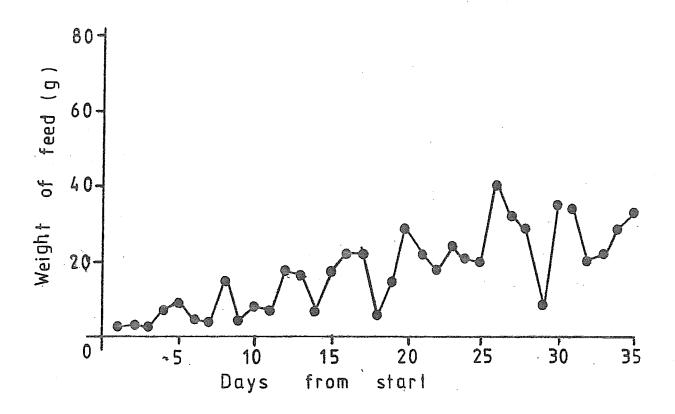
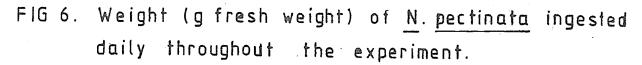


FIG 5. Weight (g fresh weight) of <u>L</u> major ingested daily throughout the experiment.





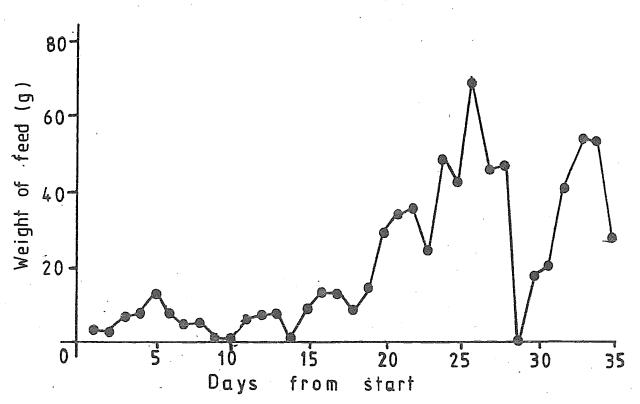
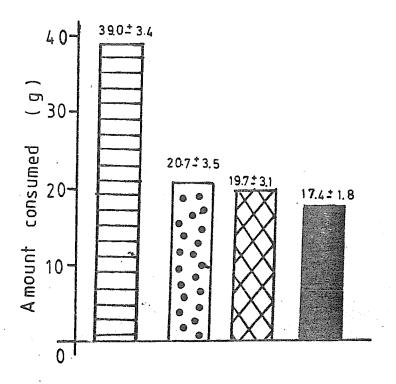


FIG 7. Mean weights (g fresh weight) of each macrophyte consumed per day in tanks 1 to 4.



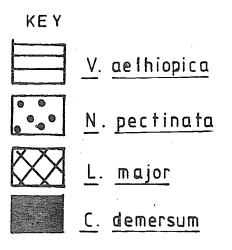


FIG 8. Proportions of macrophytes consumed in the selection trial.

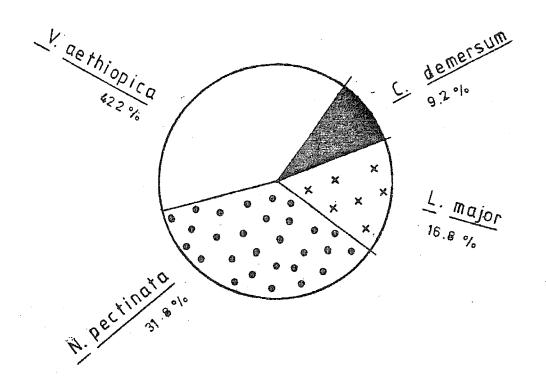
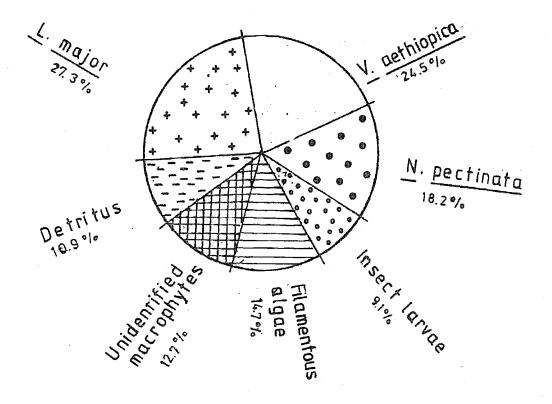


FIG. 9. % occurrence of food items in stomachs of <u>T. rendalli</u> collected from the wild.



TYPE OF PLANT		% WATER	% ORGANIC MATTER	% ASH	% PROTEIN (dry wt.)
L. major	leaves whole	91,69 91.84	7,39 7.42	0.92 0.74	8.63 10.56
<u>C. demersum</u>	leaves whole	95.34 93.15	4.10 6.23	0.56	12.99 8.61
V. aethiopica	leaves	93.73	5.45	0.82	15.12
<u>N. pectinata</u>	leaves whole	94.75 94.49	4.49 5.15	0.76 0.36	12.53 9.79

Table 2 : Percentage water, organic matter, ash and protein content of macrophytes used in the study.

Table 3 : Initial and final mean standard length, weight and relative condition of fish used in the 5 trials

TANK NUMBER	1	2	3	4	5
Initial weight (g)	26.61	25.65	29.35	16.91	10.98
Final weight (g)	22.27	22.30	24.70	16.18	11.01
% Change	-16.31	-13.06	-15.84	-0.59	0.27
Initial length (mm)	85.0	84.0	86.7	72.2	60.9
Final length (mm)	85.2	84.0	88.1	74.9	60.9
Length change (mm)	0.2	0.0	1.4	2.7	0.0
Initial condition	4.85	4.97	5.20	4.83	4.77
Final condition	4.19	4.45	4.25	4.33	4.99
Change in condition	-0.66	-0.52	-0.95	-0.50	0.22
Number of fish '	24	21	22	17	26

5. DISCUSSION

5.1 Daily consumption rates

The amount of food consumed per day in all treatments increased throughout the experimental period (Fig. 3-6). This trend is likely due to fish becoming progressively used to the aquaria. It was observed that fish did not feed during acclamatization period even though food was available at all times. The experiment was started as soon as feeding became evident (leaves nibbled off the plants) and before fish had established a stable food intake. Acclamatization period should have been longer perhaps two months (Caulton 1976).

Besides the general increase of food consumption throughout the experiment there were fluctuations in the amount each day in the same treatment (Fig. 3-6). Since the type of food was the same each day such changes could be due to environmental factors. Temperature and oxygen levels influence the amount of food consumed by fish (Caulton 1976. Prejs 1984, Hermann et al 1967). Caulton (1976) showed that the amount of <u>C. demersun</u> comsumed by <u>T. rendalli</u> increased curvilinearly, with increased temperature up to 30°C. Marked changes in oxygen levels caused a decrease in food consumed and growth with marked changes in gross food conversion efficiency occuring at 4mg/l oxygen in coho salmon (Hermann et al 1967). The correlation between pH, oxygen and temperature with daily consumption rate was however low (Table 1). In the present study these factors had no obvious effect on daily comsumption maybe because they were overshadowed by other water quality parameters that were not measured. However, on day 29 (Fig. 3-6) when the temperature fell from about 25°C to 20°C overnight there was a sharp drop in the amount of food taken.

Mean daily food consumption in the experiment were less than those expected. Caulton (1976), worked out linear regression equations for expected daily consumption of <u>C</u>. <u>demersum</u> by <u>T</u>. <u>rendalli</u> at varying tenperatures. Acording to the regression equations fish were expected to eat between 1.63 and 3.14g <u>C</u>. <u>demersum</u>, 1.56 and 2.10g <u>L</u>. <u>major</u>, 1.82 and 3.50g <u>N</u>. <u>pectinata</u>. 0.94 and 2.13g <u>V</u>. <u>aethiopica</u> and 0.52 and 1.48g from the selection trial per fish per day. Only <u>V</u>. <u>aethiopica</u> was consumed at a rate above expected. In the selection trial the rate was close to that expected.

The over all low consumption rate might have resulted from fish eating very little at the beginning of the experiment. This could explain why fish lost both weight and condition (Table 3).

Macrophytes were consumed at significantly different rates (p < 0.001, ANOVA) (Fig. 7). Nutritive quality affects rate

of food ingestion through its effect on gastric evacuation rates (Jobling 1980, 1987, Flowerdew and Groove 1979, Groove <u>et al</u> 1978). Fish fed on meals with varying levels of nutrients adjusted their feeding rates (Flowerdew and Groove 1979).

Energy and protein content are important measures of nutritive quality. There is disagreement as to which of the two is important in determining rate of food intake. Cho & Kausik (1985) and Lee and Putnam (1973) consider that energy is more important whilst Page & Andrews (1973) and Shireman et al (1978) showed that both protein and energy play the regulatory role. Protein content in this study does not appear to affect consumption rate; on the contrary <u>V. aethiopica</u> which had the highest protein level had the highest consumption rate (Table 2, Fig. 7). Perhaps energy is the regulating factor. There is a need to determine the energy content of the plants used in the present study to verify this supposition.

5:2 <u>Selection of macrophytes</u>

Fish showed higher preference for V. aethiopica (Fig. 8) which had a significantly high protein content (Table 2). This agrees with the expected diet selection strategy whereby a general herbivore food item selection is geared to maximizing intake of nutrients, particularly nitrogen and energy (Westoby 1974, Mattson 1980). The order of preference for the other macrophytes does not agree with that of protein levels. Usually selection is due to an interaction of factors although some factors like nutrient; content may be dominant. Factors like palatability which /includes characteristics like fibre content affect selection (Westoby 1977, Prejs 1984). Plants with high structural material and polyphenolic compounds are selected against (Horn et al 1982). Horn et al showed that morphological features such as high surface to weight ratio, life span were important in diet selection. Tan et al (1970) indicated that superiority of <u>Hydrilla</u> verticillata over other plants tested as diet was due to its soft nature (low fibre content) and high ash content. A comprehensive chemical analysis of the plants used in the study as well as an assessment of leaf toughness, spatial availability of leaves on plants and other parameters measured by Prejs (1984) need be carried out in order to explain some aspects of selection in the present study.

<u>C.</u> demensum which was the least preferred of the four macrophytes was not observed in the stomachs analysed. This indicates further negative selection by <u>T. rendalli</u>. It appears the plant is being avoided by the fish. The proportion in which the other plants <u>V. aethiopica</u>, <u>N. pectinata</u> and <u>L. major</u> were consumed in the wild differ significantly (P < 0.001, chi-square) from those obtained in the selection trial (Fig. 8 & 9). Selection in the wild is

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governed not only by nutrition level but also by abundance (Horn et al 1982, Mwebaza-Ndawula 1984). For <u>T. rendalli</u> in the wild diet is strongly related to abundance (Caulton 1977). In the Kariba basin <u>L.major</u>, <u>N. pectinata</u> and <u>C.</u> <u>demersum</u> are dominant macrophytes (Machena 1987). It seems therefore that there was selection of <u>V. aethiopica</u> since its abundance in the wild was low. In the experiment plants were presented to the fish in equal propotions which does not ocuur in the wild. Selection in the wild is expected to be a compromise between preference and selection so as to balance the cost of ingestion. (including cost of search) of a particular food kind and the benefit derived from it (Prejs 1984).

5.3 Effects of diet on growth

Fish in all treatments except the selection trial lost both weight and condition during the experiment. Loss of weight was due mainly to fish eating very little initialy. There however seems to be a relationship between weight losses and ash content (Table 2 & 3). High ash levels indicates levels of mineral nutrition (Tan 1970). V. aethiopica would be regarded as superior bacause of relatively high ash and protein content. Fish in the selection tank increased in weight. Maybe a variety of food is superior in quality compared to single item diets. Fish in the wild actually feed on a variety of species (Fig. 9). The animal component included in the diet improves the food quality and potential. for growth (Fischer 1973, Appler 1985). Change in length was small so the relationship to diet was not clear. Fish need to be monitored until weight starts to rise and length has changed significantly in order to reach meaningful conclusions on the effects on growth resulting from feeding on different macrophytes.

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25

Assay of the Organic Nitrogen Content of Tissues

Most of the nitrogen in plant tissues is in the form of proteins. The remainder is refered to as the non-protein nitrogen, or NPN, and includes amino acids, amides, purines, pyramidines, ammonium salts, nitrates and nitrites. A complete analysis of all the nitrogenous compounds in a sample of plant tissues would be prohibitively expensive and time assuming. Instead we can measure the nitrogen in a sample by the Kjeldahl method in which all amino nitrogen (-NH) is first converted to ammonium which is then titrated. This, method measures the protein nitrogen plus all the NPN except the organic nitrate and nitrite.

The Kjeldahl method has three stages:

- 1. Acid digestion of the sample and conversion of all amino nitrogen to ammonia.
- 2. Steam distillation of ammonia from the digest into a receiving flask
- 3. Volumetric titration of the ammonia and calculation of the nitrogen content of the sample.

Digestion

- 1. To each tube including blank, add 2 tablets of catalyst (Potassium and Copper) and 12ml of analytical grade sulphuric acid.
- 2. Digest samples at 420° C for 20-30 minutes by which time the sample should be clear.
- Remove tubes from digestion block and allow to cool for 5 minutes.
- 4. Dilute sample to approximately 100ml with distilled water.

Distillation

- 1. Transfer digestion tube to distillation apparatus.
- 2. Place in position the receiver flask containing approximately 20-30ml of 4% boric acid and ensure that the delivery tube is below the surface of the boric acid.
- 3. Dispense one "shot" of 40% NaOH into the digestion tube (sample will turn brown).
- 4. Set timer to 3 minutes and open steam valve.

- 5. After 3 minutes lower receiver flask so that the delivery tube is no longer immersed in the boric acid; and continue distillation for a further 20 seconds.
- 6. Close steam valve.
- 7. Remove receiver flask and add 0.5ml of indicator (bromocresol green + methyl red). Add magnetic stiring bar.

Titration

Titrate with 0.1M HCL to a colourless/grey end point. Calculate the weight of Nitrogen from the relationship:-

1 mole of HCL = 1 mole of Amonia N

e.g. The z ml of 0.1M HCL required to titrate a sample would contain

0.1 x z moles of HCL 1000 = 0.1 x z moles of Amonia N 1000

= 0.1 x z x 14.01g of Amonia N 1000

Trial	Temp. °C	Oxygen mg/1	pH ,
<u>C. demersum</u>	24.0 + 0.37	3.65 + 0.44	7.61 + 0.32
L. major	23.4 + 0.33	3.44 + 0.29	7.72 + 0.26
N. pectinata	23.6 + 0.32	2,68 + 0.36	7.67 + 0.33
V. aethiopica	23.9 + 0.35	4,45 + 0.35	7.69 + 0.33
Selection	25,0 + 0,36	3.62 + 0.55	7.60 + 0.27
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Mean temperature, oxygen and pH during the experiment period.

Mean number of fish observed feeding in a minute at 0900 hrs., 1400 hrs. and 1730 hrs.

Trial	0900 hrs	1400 hrs	1730 hrs
<u>C. demersum</u>	2.5	2.7	3.2
L. major	5.5	2.7	1.1
N. pectinata	4.9	1.3	0.9
V. aethiopica	6.6	2.5	1.0
Selection	9.1	5.3	5.9

Trial	0900 hrs	1400 hrs	1730 hrs
<u>C. demersum</u>	2.5	2.7	3.2
L. major	5.5	2.7	1.1
<u>N. pectinata</u>	4.9	1.3	0.9
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