

Infection rates of a silver eel population *Anguilla anguilla* (L.) of the river Yser basin (Flanders) with *Anguillicola crassus* Kuwahara, Niimi and Itagaki and effects of the parasite on the muscle composition and energy content of migrating male silver eel

C. Belpaire¹, A. Jansen², B. Denayer¹, D. De Charleroy¹ and F. Ollevier²

1 Institute for Forestry and Game Management, Ministry of the Flemish Community, A. Duboislaan 14, B-1560 Groenendaal, Belgium.

2 Laboratory for Ecology and Aquaculture, Zoological Institute, Naamsestraat 59, B-3000 Leuven, Belgium.

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Abstract. This paper describes the infection of migrating silver eel from the Blankaart Nature Reserve (River Yser basin) during autumn 1991 with *Anguillicola crassus*. Possible effects of parasitic infection on body composition of male silver eel are analysed by measuring lipid, protein, ash and water content and energy value of muscle tissue from heavily, and slightly or non-infected eel. Apart from differences in ash content, no evidence is found for significant variations in muscle composition between both groups. There is no significant difference in energy value between the low and the highly infected group.

During the past five years the Working Party on Eel (EIFAC 1989, 1991, 1993) gave evidence that European eel stocks were declining. Serious concern should be given to the status of European *Anguilla* stocks and to environmental and human factors which could be responsible for this decline.

In 1989 the EIFAC Working Group on Eel warned for possible effects of the parasite on the silver eel migration and consequently on spawning success of the eel. As was suggested by several workers (Boëtius 1989, Ghittino et al. 1989, Fontaine et al. 1990, Sprengel and Luchtenberg 1991) infection with *Anguillicola crassus* might have a possible effect on the normal function of the swimbladder and thus inhibit normal swimming and migrating activities of eel. Consequently EIFAC (1991) recommended that research about the potential effects of *Anguillicola crassus* on silver eel should be encouraged.

This study aims to measure potential effects of *Anguillicola* on the body composition of silver eel.

MATERIALS AND METHODS

Study area

As for various reasons the river Yser was of particular interest for eel, this river catchment was selected as a model for a general study on different aspects of ecology and migration of eel in Belgium (Denayer and Belpaire 1992 and 1993). In this river catchment the Blankaart Nature Reserve was chosen as study area since it is a small eutrophic lake (30 ha) with a dense eel population. Migrating silver eel are relatively easy to sample, as they leave the reserve through the Stenensluisvaart, an affluence of the river Yser (Figure 1). Moreover, infection of yellow eel with *Anguillicola crassus* was known and reported previously (Belpaire et al. 1989).

The silver eel population leaving the Blankaart Nature Reserve: prevalence and intensity of infection with *Anguillicola crassus*

The silver eel used for these experiments were leaving the Blankaart Nature Reserve and collected on the Stenensluisvaart by fyke netting during the nights of 4 to 6 of November 1991. Characteristics of this silver eel run have been described previously by Denayer and Belpaire (1992) and by Jansen (1992). The length frequency distribution of these silver eel is given in Figure 2. 72% of the eels were male (total length between 35 and 45 cm), 28% were female (45 - 90 cm). An uniform batch of 110 eels of the male group was selected for these experiments. Total length and weight of these eels was measured. Length frequency distribution of this batch is given in Figure 3. Mean length was 40.0 ± 1.8 cm (min 36.0 cm, max 44.0 cm), mean weight was $111.7 \text{ g} \pm 11.1 \text{ g}$ (min 88.7 g, max 137.1 g). Parasitological examination of the eels consisted in dissecting and examining the swimbladder. No other tissues were analysed. Anguillicolid nematodes were identified and for each stage (except L₂) the number of individuals was counted. Adult helminths were sexed. Occurrence or absence of L₂ stages was checked. Prevalence of infection was calculated as the percentage of the total batch which was infected either by larval stages, by adult or by preadult worms. Intensity of infection was determined as the mean number of parasites (all stages) per infected eel.

Body content analysis of male silver eel with a low vs. a high infection level of *Anguillicola crassus*

As not all of the 110 eels could be analysed on the body content and energy value it was necessary to select two batches, a low and a high infected group (for selection procedure see below).

Body content analyses were carried out on muscle tissue, removed and homogenised from each individual.

Dry weight was determined from 2 g samples after placing during 16 hrs in an oven at 100°C and cooling in a desiccator.

Lipids were measured after acid hydrolysis of the sample followed by the petroleum/ether extraction (soxtec).

The samples were analysed on their protein content by using the Kjehldahl method.

Ash was determined by treating the samples 2 hrs in the muffle oven at 300°C followed by 14 hrs at 550°C.

Water, lipid, protein and ash content are expressed in % body weight.

The energy value of the samples was measured by using a bomb calorimeter. The caloric value is expressed in cal per g body weight.

RESULTS

Prevalence and intensity of infection with *Anguillicola crassus* for the male silver eel leaving the Blankaart Nature Reserve

The swimbladder analyses of the 110 male silver eel revealed that 106 eels were infected by *Anguillicola crassus* (adult, preadult or larval stages). The prevalence reached 96.4%.

78 eels (70.9 %) had adults in their swimbladder, 41 (37.3 %) preadults, 94 (85.5 %) L₄ larval stages and 81 (73.6 %) L₃ larvae. The number of adults in one eel varied between 0 and 27 specimens. Frequency distribution of the adult worms is represented in Figure 4. For L₃, L₄ and preadult stages the numbers ranged from 0 to 96, 0 to 60 and 0 to 5 individuals respectively. The overall intensity of infection was 16.1 parasites per infected eel.

In 30 (27.3 %) cases the presence of L₂ stages was noticed. 29 swimbladders (26.4 %) were fibrotic.

One eel was extremely infected by 96 L₃, 60 L₄, 3 preadults and 27 adults.

Body content and energy values of male silver eel with a low vs. a high infection level of *Anguillicola crassus*

Initially the aim was to compare the body composition of highly infected silver eel with eels which were not infected at all. However as it was impossible to get enough uninfected eels of an uniform batch, due to the high *Anguillicola crassus* prevalence of this population, we had to include some individuals which had a low level of infection. All pathological effects (histopathology, effects on blood composition, effects on swimming behaviour,...) seem to be related to the number of nematodes present and thus eel harbouring a small number of *A. crassus* are apparently not seriously affected by the presence of the parasite (Thomas 1993).

Consequently the experimental groups were composed as follows: a low infected batch ('L') of 17 silver eel and a high infected group ('H') of 20 eels. Group L consisted of 4 eels free of *Anguillicola*, 14 eels with less than 10 larvae and 3 eels with 10 to 76 larvae. No eels with preadults or adults or with fibrotic swimbladder were included in this group. Individuals of the H condition were selected on basis of Figure 4: at least 11 adults were present in each swimbladder (between 11 and 27). Besides, they have a larger total number of larvae ($L_3 + L_4 +$ preadults) in their swimbladder (Table 1).

The weight/length relationship of the eels is illustrated in Figure 5. There is no evidence for any effect of *Anguillicola crassus* infection on the condition of both groups.

The results of the chemical analyses of each eel are represented in Table 1. Table 2 gives an overview of the mean values for both groups.

Figures 6 to 10 illustrate muscle composition as a function of body length for both conditions. As can be seen in Table 3 no statistically significant differences occur between both groups for water, lipid and protein content and for energy value. Only the ash content of muscle tissues from the highly infected group was significantly higher than for the L group ($p = 0.0056$).

DISCUSSION

High infestation rates of silver eel leaving the Blankaart Nature reserve are not surprising: a 1988 survey revealed for this yellow eel population a prevalence of 90.9 % (Belpaire et al. 1989). Compared to these results prevalences still increased (up to 96.4 % for male silver eel). These results are similar to *Anguillicola crassus* infections of other eel populations in Belgium: e.g. Thomas (1993) found in 1990 in the Albertcanal a population of eels infected with a prevalence of 90.2 % and an infection intensity of 17 nematodes per infected eel.

The eels which were fyked for our experiments were just starting their seaward migration (in these polders migration is initiated by activation of drainage pumps (Denayer and Belpaire 1993)). There was no evidence that the start of this migration of infected eels was influenced by the presence of parasites.

Recent publications confirm that a considerable effort has been done to evaluate the impact of *Anguillicola crassus* on eel. Histopathological work was carried out by several authors (e.g. van Banning and Haenen 1990, Molnar et al. 1993). The potential effects of *Anguillicola* on haematological parameters of the eel have been described (Boon et al. 1989 and 1990 a,b). Increased mortality in both cultured (Liewes and Schaminee-Main 1987, Mellergaard 1988) and wild eel (Hartmann 1987, Székely et al. 1991, Molnar et al. 1991, Molnar et al. 1993) were reported.

Following various reports conditions of infected and non-infected eels are not significantly different (Koops and Hartmann 1989, Möller et al. 1991). Our weight/length relationships endorse this statement.

In some cases however, divergent results and contradictory reports on the effect of *Anguillicola crassus* on the eel have been presented, especially about their effect on silver eels and their migration. Heavy *Anguillicola* infestations could influence the hydrostatical role of the swimbladder by the vertical movements of the eel during its seawards migration, but could also have an impact on condition and the amount of fat and energy reserves migrating silver eel have and consequently on spawning success. Silver eel which are migrating to the sea exhibit considerable performances of horizontal as well as vertical movement, as was shown by recent tracking studies (Tesch 1989 and 1993, Tesch et al. 1991). Following Tesch (1993) an intact swimbladder seems to be indispensable for the oceanic migration of the eel. Fontaine et al. (1990) however reported that infestation of silver eel with *A. crassus* does not cause drastic consequences for the capacity to acclimate in marine water at high hydrostatic pressure (60 atm).

The experiments of Sprengel and Lüchtenberg (1991) demonstrate the effects of *A. crassus* infection on swimming performance of eel. In eels between 17 and 45 cm length maximum swimming speed was reduced by 2.9 % for weakly infected eels (1 parasite) and up to 18.6 % for eels with more than 10 nematodes in the swimbladder (compared to the maximum swimming speed of uninfected eels) (Fig. 12). Unfortunately, the authors do not mention the stage (yellow eel or mature silver eel) of the eels used in their experiments. However these experiments give evidence that this parasite hampers swimming performances of the host. In contrast Möller et al. (1991) could not detect negative effects of *A. crassus* infection on body condition and liver somatic index.

The results of body content analyses (Fig. 11) are comparable to the findings of Boëtius and Boëtius (1985) who found for male silver eel similar values: water 52.9 %, lipids 28.3 %, protein 14.0 % and ash 2.0 %. Only the ash content was considerably higher in the Danish eel (2.0 % vs. 0.97 % and 1.10 % for our L and H condition). This is probably due to the fact that in the latter experiments whole eel homogenates were used (including bony structures such as skull, vertebrae and spines).

The absence of any significant difference in water, lipid and protein content between the low and the high infected group of male silver eel is an indication that there is no evidence for a clear or drastic impact of the parasite on its hosts body composition at the start of the seaward migration. However one has to bear in mind that only muscle tissues of male silver eel were studied in these experiments. (Analyses of whole body homogenates of (larger) female silver eel could give different results).

No explanation was found for the significant lower ash content in low infected eels.

Several authors emphasize the importance for silver eel to accumulate a sufficient energy reserve to accomplish a successful migration and breeding as migrating silver eel do not feed (Boëtius and Boëtius 1980). In our experiments the energy values in muscle tissues of the low infected group were not significantly different from those of the high infected eel. No evidence was found for a negative impact of *Anguillicola crassus* infection on energy reserve of male silver eel. These data however do not exclude the possibility that an abnormal functioning swimbladder increases the energy demand necessary to fulfill a successful spawning migration.

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Figure 1 : Location of the Blankaart Nature Reserve in the Yser basin and sampling locality (arrow) of the silver eel from the Blankaart Nature Reserve at the Stenensluisvaart on 4 to 6 November 1991.

Figure 2 : Length frequency distribution for migrating silver eel leaving the Blankaart Nature Reserve and caught at the Stenensluisvaart (Yser basin) during 3 consecutive nights on 4 to 6 November 1991 (after Denayer and Belpaire 1993).

Figure 3 : Length frequency distribution of 110 migrating male silver eels used for analysis of infection levels with *Anguillicola crassus*.

Figure 4 : Frequency distribution of adult *Anguillicola crassus* in the swimbladder of a sample of 110 male silver eels from the Blankaart Nature Reserve (N=110).

Figure 5 : Body weight versus total length of male silver eel low and high infected with *Anguillicola crassus*. Silver eel run of 4 to 6 November 1991 at the Blankaart Nature Reserve. Low infection: N=17, high infection: N=20.

Figure 6 : Water content (% of body weight) of male silver eel low and high infected with *Anguillicola crassus*. Silver eel run of 4 to 6 November 1991 at the Blankaart Nature Reserve. Low infection: N=17, high infection: N=20.

Figure 7 : Lipid content (% of body weight) of male silver eel low and high infected with *Anguillicola crassus*. Silver eel run of 4 to 6 November 1991 at the Blankaart Nature Reserve. Low infection: N=17, high infection: N=20.

Figure 8 : Protein content (% of body weight) of male silver eel low and high infected with *Anguillicola crassus*. Silver eel run of 4 to 6 November 1991 at the Blankaart Nature Reserve. Low infection: N=17, high infection: N=20.

Figure 9 : Ash content (% of body weight) of male silver eel low and high infected with *Anguillicola crassus*. Silver eel run of 4 to 6 November 1991 at the Blankaart Nature Reserve. Low infection: N=17, high infection: N=20.

Figure 10 : Energy value in the body of male silver eel low and high infected with *Anguillicola crassus*. Silver eel run of 4 to 6 November 1991 at the Blankaart Nature Reserve. Low infection: N=17, high infection: N=20.

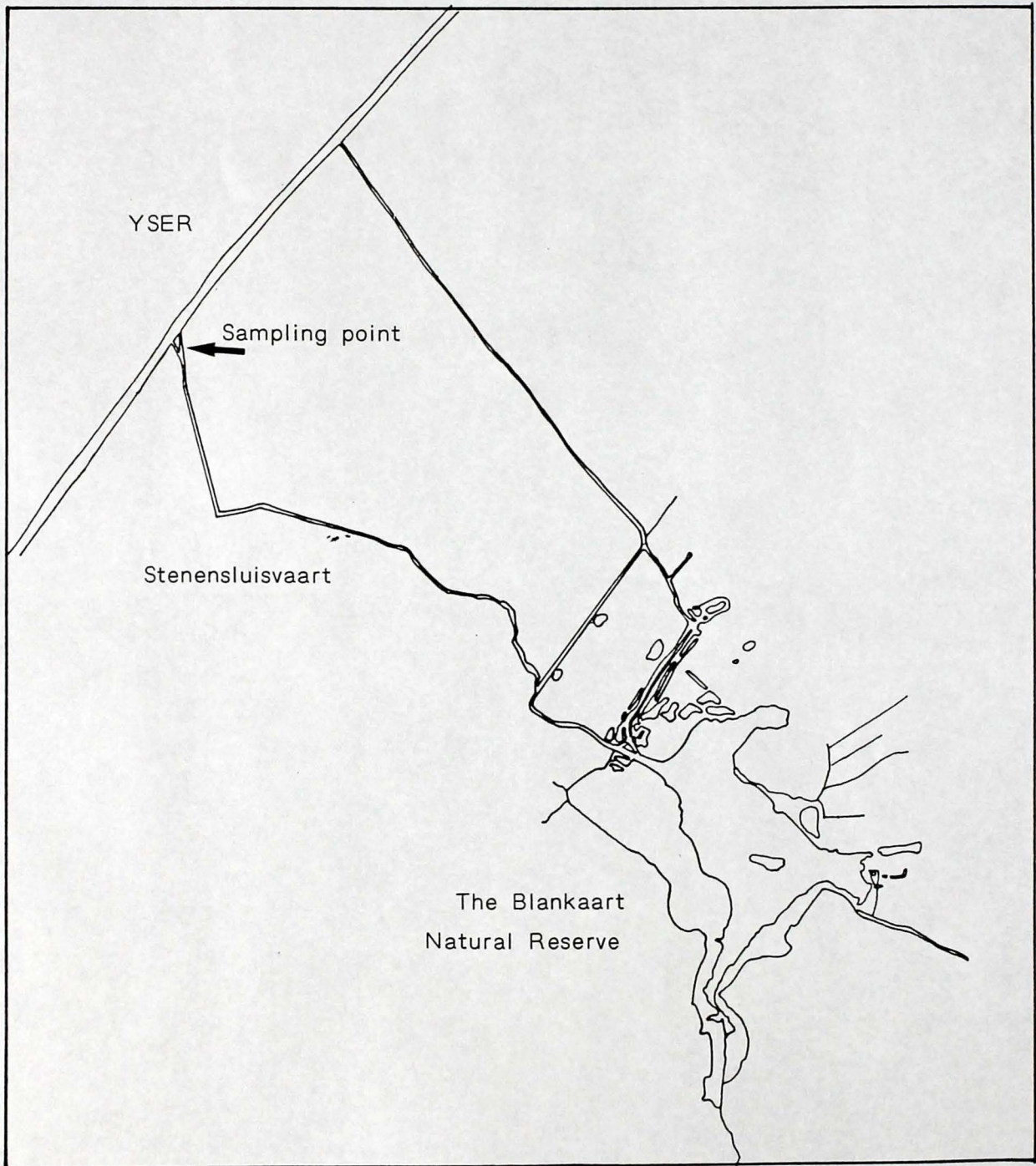
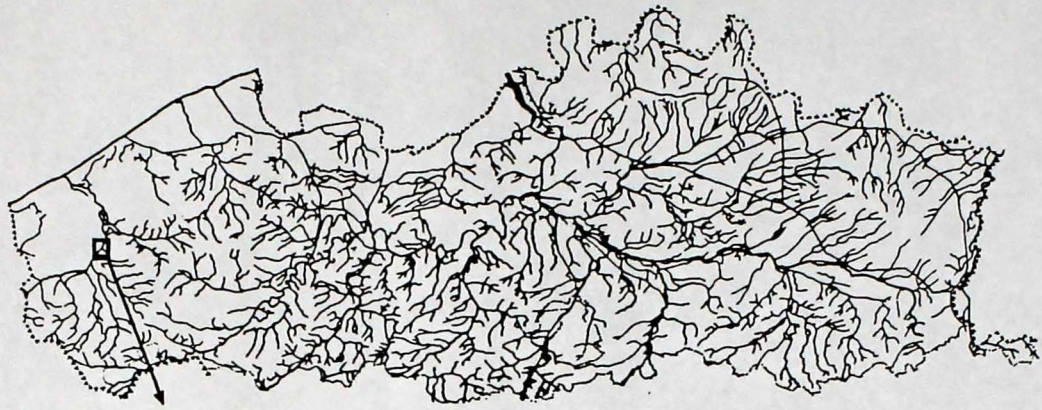
Figure 11 : Body composition of male silver eel (mean values) of an uninfected group (from homogenates of whole eel, after Boëtius and Boëtius 1985) and of the low and high infected groups (from muscle tissue homogenates).

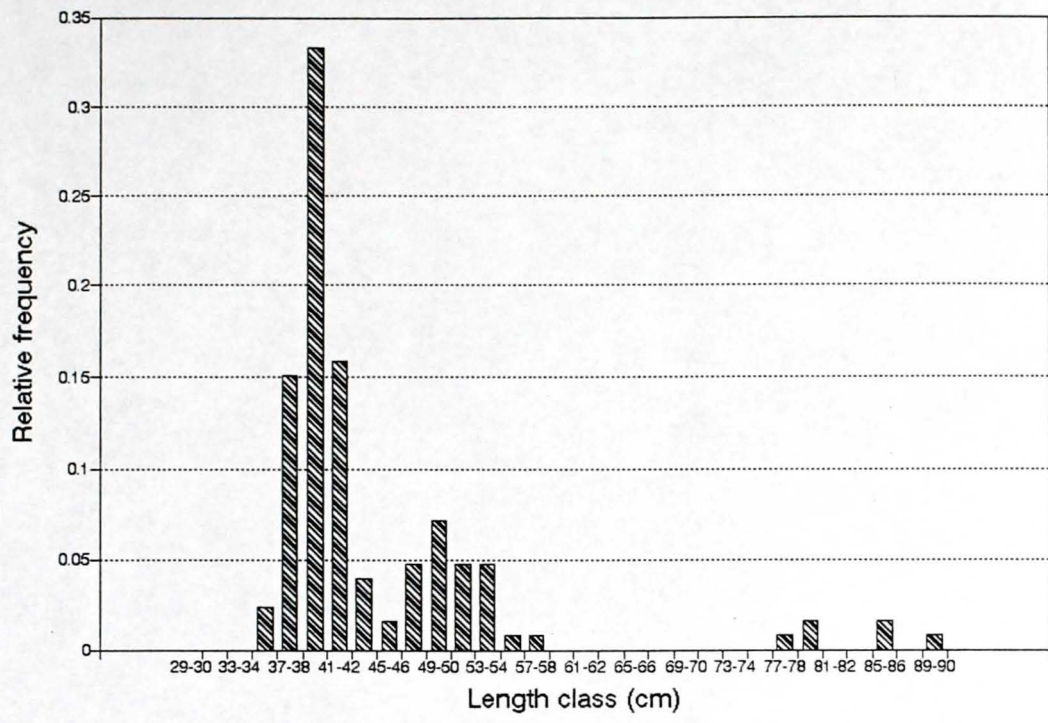
Figure 12 : Reduction of swimming performances of eel measuring 17 to 45 cm (back-calculated to 32 cm standardised length) as a function of the intensity of infection with *Anguillicola crassus* (after Sprengel and Lüchtenberg 1991)

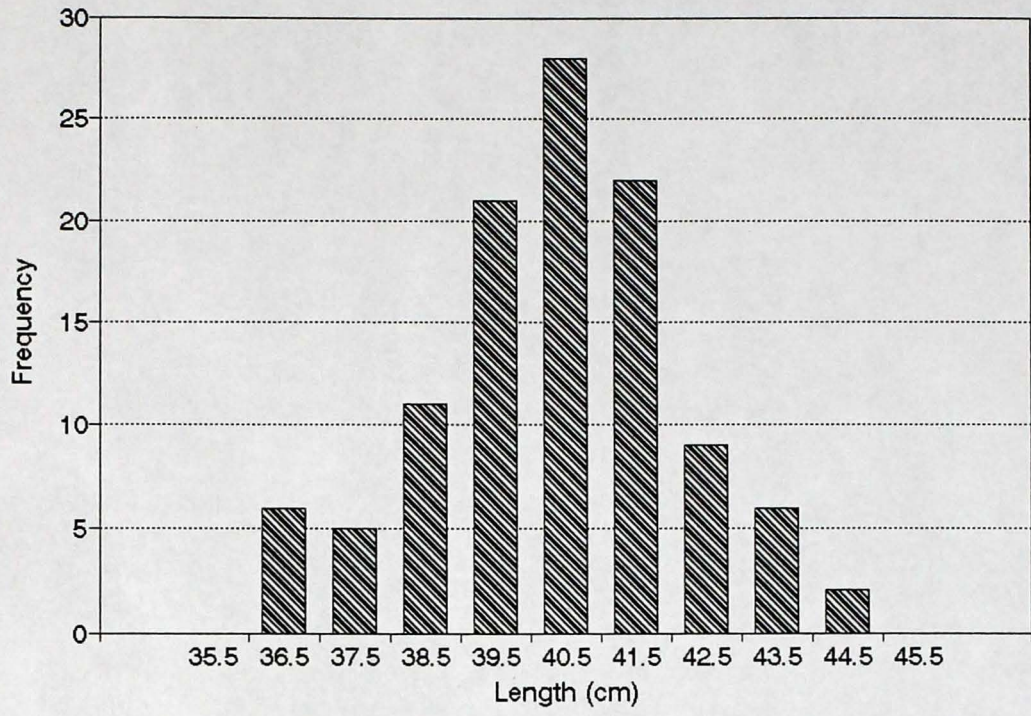
Table 1 : Weight, length, *Anguillicola* infection, body composition and caloric value of each silver eel from L and H condition.

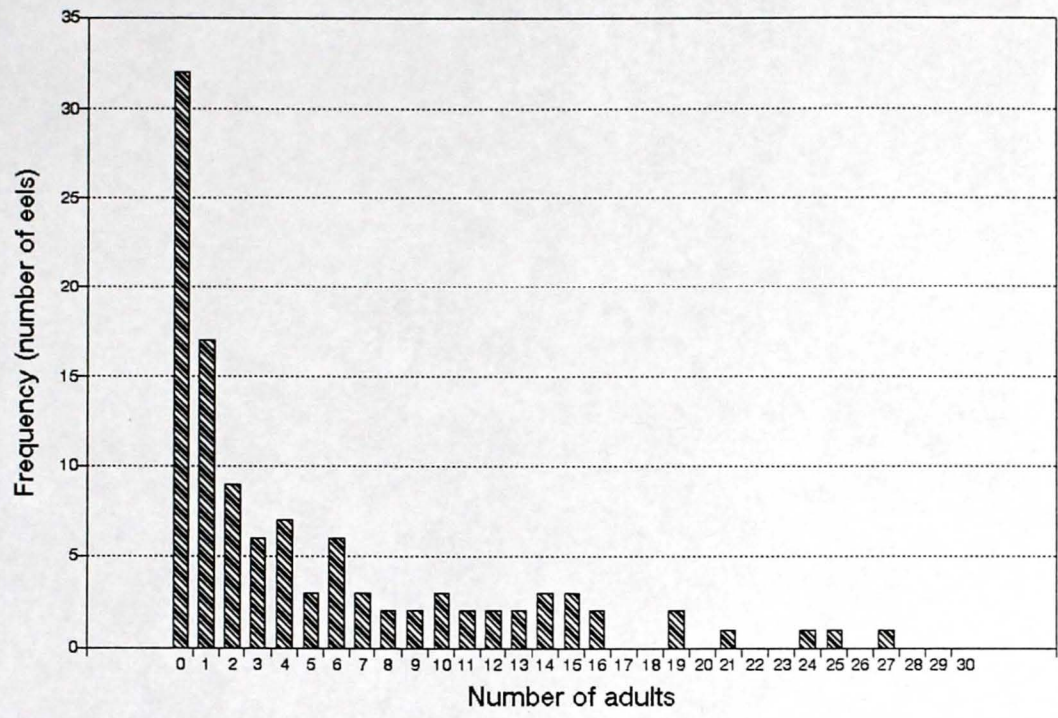
Table 2 : Body composition and caloric value of L and H condition. Mean values, standard deviation, minimum and maximum values.

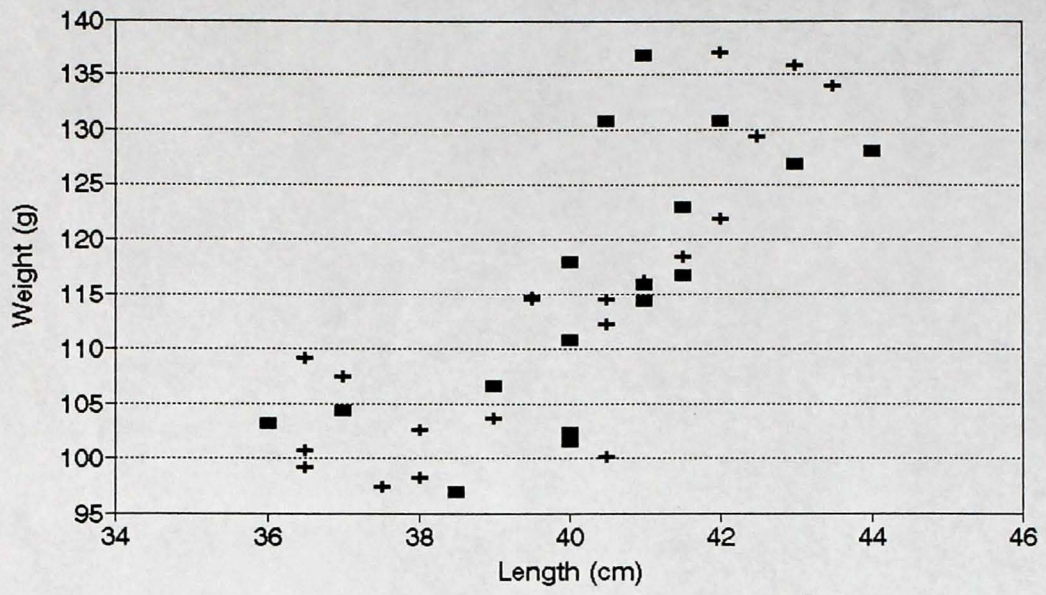
Table 3 : Statistical analysis (comparison of means) between the mean values of both groups for each component. Means of groups with same letter are not significant different.



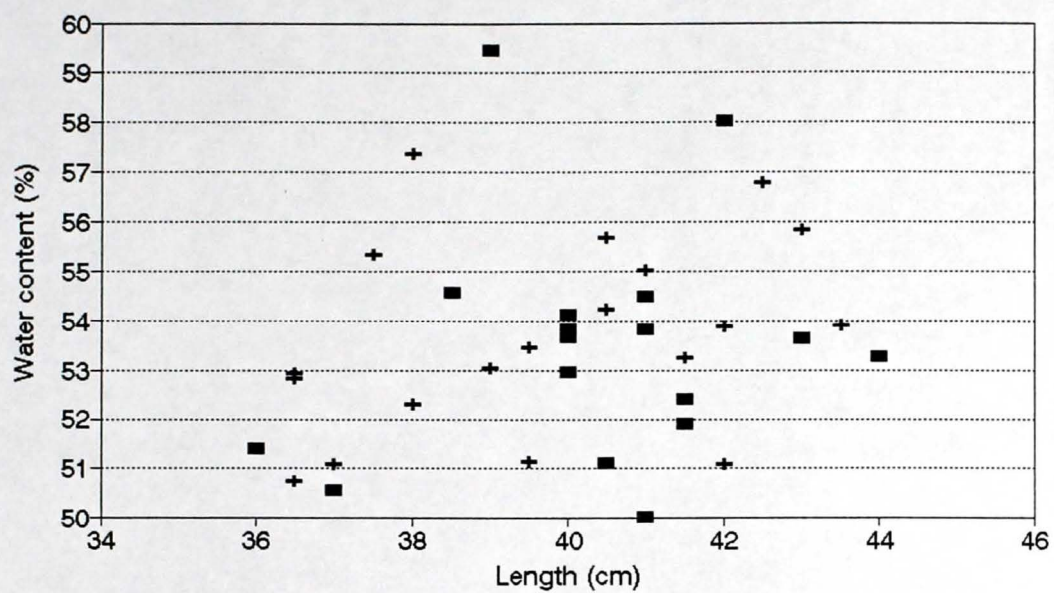




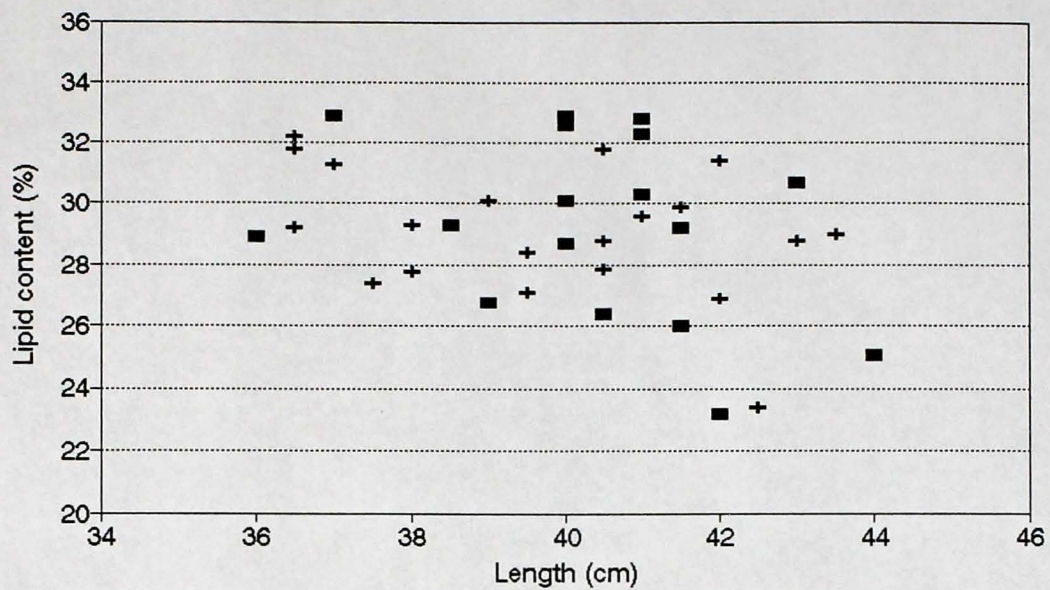




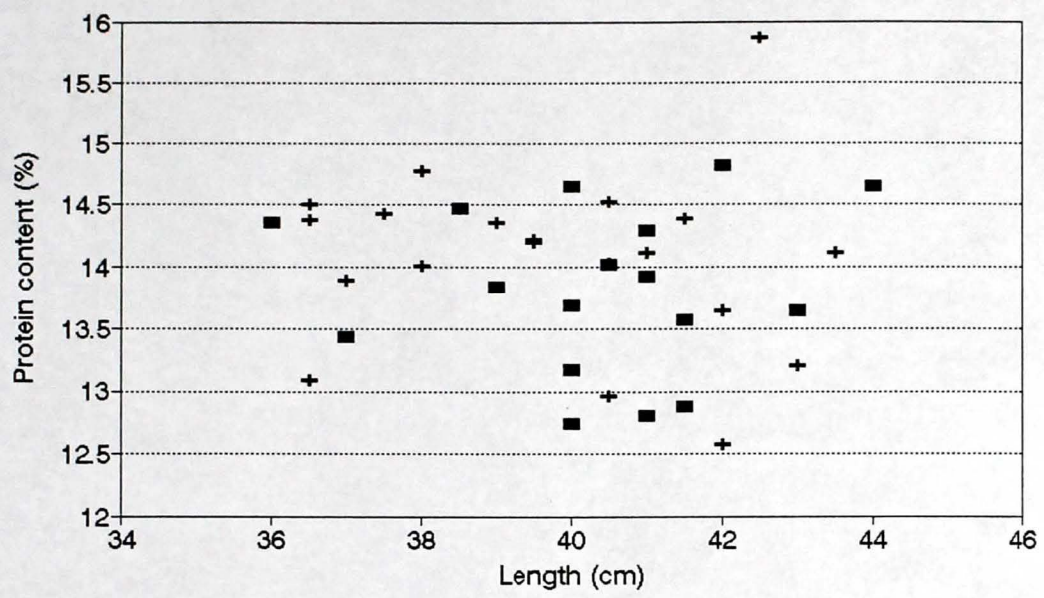
■ Low infection + High infection



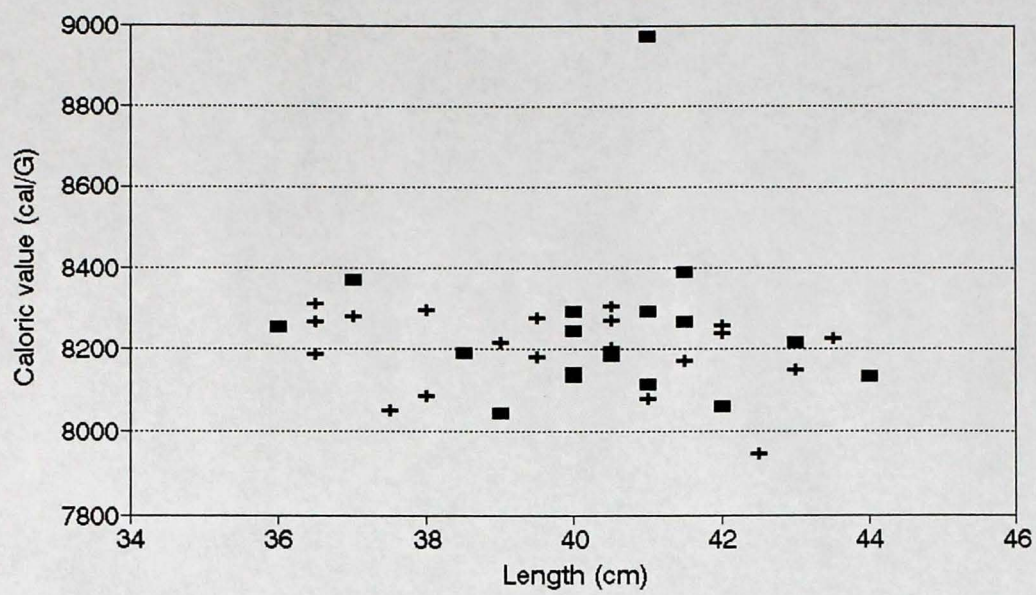
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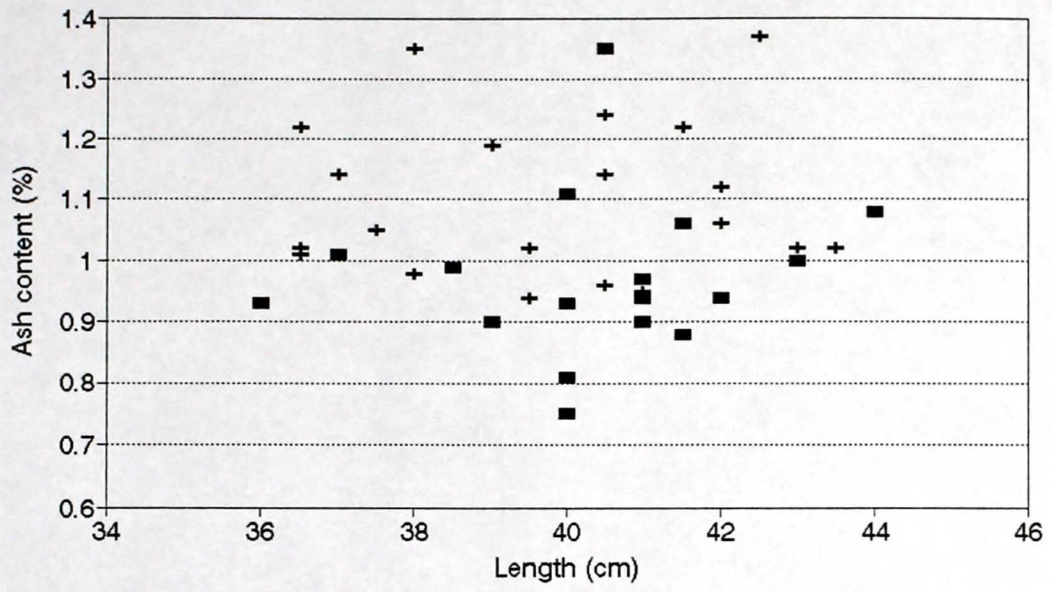
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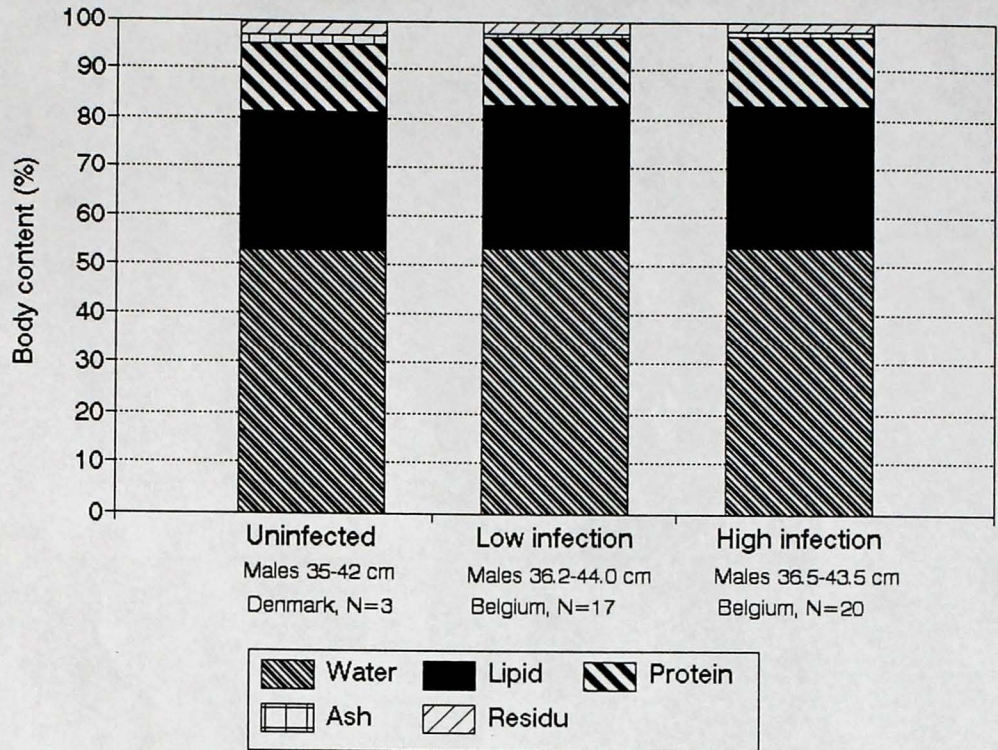
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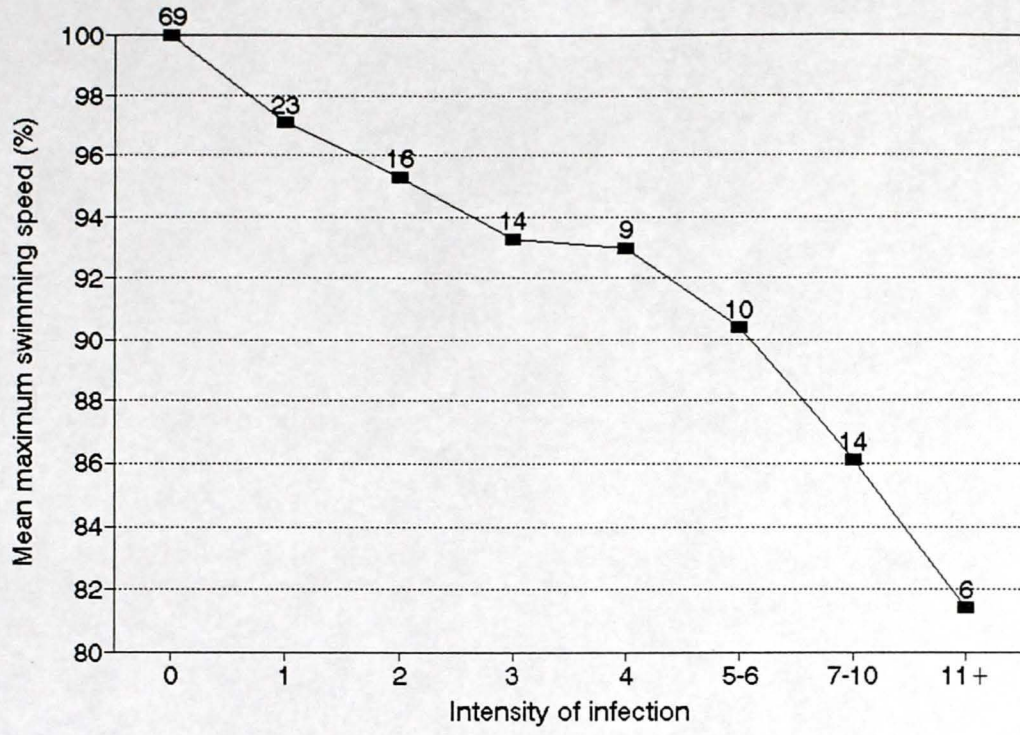


■ Low infection + High infection



■ Low infection + High infection





Group	N larvae			N adults	Weight g	Length cm	Fat %	Protein %	Water %	Energy Cal/g	Ash %
	L3	L4	PA								
L	0	0	0	0	118.0	40.0	32.6	13.18	53.84	8291	0.75
L	0	0	0	0	102.4	40.0	30.1	12.74	53.68	8142	0.81
L	0	0	0	0	110.8	40.0	32.9	14.65	52.98	8135	0.93
L	0	0	0	0	116.0	41.0	32.8	12.81	50.02	8292	0.97
L	0	1	0	0	101.6	40.0	28.7	13.69	54.13	8245	1.11
L	1	0	0	0	136.8	41.0	30.3	14.29	53.82	8116	0.94
L	2	0	0	0	103.3	36.0	28.9	14.36	51.43	8254	0.93
L	1	2	0	0	114.4	41.0	32.3	13.93	54.50	8974	0.90
L	3	0	0	0	130.8	42.0	23.2	14.83	58.01	8060	0.94
L	1	3	0	0	123.0	41.5	29.2	13.57	51.89	8266	1.06
L	0	5	0	0	130.8	40.5	26.4	14.02	51.14	8184	1.35
L	1	4	0	0	126.9	43.0	30.7	13.64	53.67	8217	1.00
L	1	5	0	0	116.8	41.5	26.0	12.88	52.42	8389	0.88
L	0	9	0	0	106.7	39.0	26.8	13.84	59.45	8045	0.90
L	13	5	0	0	104.5	37.0	32.9	13.44	50.57	8372	1.01
L	14	16	0	0	96.9	38.5	29.3	14.48	54.57	8191	0.99
L	34	42	0	0	128.0	44.0	25.1	14.65	53.28	8134	1.08
H	96	60	3	27	134.0	43.5	29.0	14.11	53.91	8225	1.02
H	2	13	1	25	107.6	37.0	31.3	13.89	51.09	8279	1.14
H	2	5	0	24	98.2	38.0	27.8	14.79	57.37	8084	0.98
H	1	1	0	21	109.1	36.5	31.8	13.09	52.94	8311	1.22
H	30	25	0	19	112.2	40.5	31.8	14.03	51.14	8303	0.96
H	0	3	0	19	100.2	40.5	27.9	12.97	55.69	8270	1.14
H	12	11	0	16	114.7	39.5	28.4	14.19	51.16	8278	1.02
H	11	8	2	16	121.8	42.0	26.9	12.58	53.88	8258	1.06
H	13	16	1	15	102.6	38.0	29.3	14.00	52.32	8295	1.35
H	0	3	2	15	114.5	39.5	27.1	14.22	53.47	8181	0.94
H	3	8	2	15	114.5	40.5	28.8	14.53	54.22	8203	1.24
H	0	5	1	14	103.7	39.0	30.1	14.36	53.03	8216	1.19
H	0	4	0	14	118.5	41.5	29.9	14.39	53.25	8172	1.22
H	9	13	1	14	129.4	42.5	23.4	15.88	56.80	7948	1.37
H	40	14	2	13	99.2	36.5	29.2	14.38	52.84	8186	1.02
H	8	9	4	13	135.8	43.0	28.8	13.21	55.85	8150	1.02
H	1	4	0	12	137.1	42.0	31.4	13.65	51.08	8236	1.12
H	1	4	0	12	100.7	36.5	32.2	14.50	50.76	8268	1.01
H	0	3	1	11	97.5	37.5	27.4	14.43	55.34	8049	1.05
H	1	4	0	11	116.2	41.0	29.6	14.11	55.01	8079	0.95

Group	Fat %	Protein %	Water %	Energy Cal/g	Ash %
L	29.3 A	13.82 A	53.49 A	8253 A	0.97 A
H	29.1 A	14.07 A	53.56 A	8200 A	1.10 B