

# VARIATIONS IN LIVER AND BODY CONDITION DURING GONAD DEVELOPMENT OF ATLANTIC HALIBUT, *HIPPOGLOSSUS HIPPOGLOSSUS* (L.)

By

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## ABSTRACT

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Data were collected from Atlantic halibut (*Hippoglossus hippoglossus*) caught in gill nets and on long lines in northern Norway between September and March during the years 1981-1986. The liver is significantly depleted during the spawning season, thus indicating that it is an important energy source for the halibut in this period. The carcass seems less affected by the energy expenditures involved in the seasonal accumulation of reproductive tissues and in spawning, particularly in females where no significant sacrifice of body weight was observed.

## INTRODUCTION

In recent years there has been a growing interest in marine fish species in sea ranching and aquaculture. Owing to its high market price the Atlantic halibut has particularly received the attention of present-day aquaculturists, and rearing and farming experiments with the species are in progress at several Norwegian institutions.

The halibut is a long-lived species which is believed to spawn seasonally for a number of consecutive years (see JAKUPSSTOVVU and HAUG 1988). In northern Norway the spawning of Atlantic halibut takes place at various localities within the fjords and on the edge of the coastal banks (HJØRT 1905, DEVOLD 1938). Spawning, which usually takes place at depths of 300-700 m and at water temperatures of 5-7°C, lasts from December to March, with peak activity occurring between late January and early February (KJØRSVIK *et al.* 1987).

For a number of flatfish species it is known that energy reserves, which are generally deposited in liver and carcass, are considerably depleted during

the course of gonad development and spawning (LOVE 1970, DAWSON and GRIMM 1980, JOBLING 1980, ROFF 1982). Whether this is also true for the halibut is not yet known. With reference to the emerging interest in halibut aquaculture, it is obvious that this question needs to be resolved. It has relevance to the rearing of broodstocks of adult fish aimed at producing eggs and milt, and to the production of halibut food fish. This may well proceed beyond the stage of sexual maturity, since the halibut is a multi-year spawner showing rapid growth rates in body tissues even after sexual maturity (JAKUPSTOVU and HAUG 1988).

The aim of this paper, therefore, is to study seasonal variation in the size of gonads, liver, and body tissues of wild Atlantic halibut, preparing for spawning in autumn and accomplishing the spawning in winter.

#### MATERIALS AND METHODS

Halibut stocks in Norwegian waters have been quite heavily depleted in recent years (HAUG 1984, HAUG and TJEMSLAND 1986). In order to provide enough data for this study, therefore, material had to be collected over several years and from several localities. The fish were collected during 1981–1986 by gill netting and longlining at six sites in northern Norway (Fig. 1). Samples were collected from September to March from commercial catches (Vestfjord, Røstbanken, off Vesterålen), and research cruises (Andfjord, Malangen, Sørøysund). All fish were sexed, and total fish lengths (TL) were measured to the nearest centimeter. Eviscerated weights without removing head and gills ( $W$ ) were recorded to the nearest 0.05 kg, while gonad weights (GW) and liver weights (LW) were recorded to the nearest 0.001 kg. All data from the different sites and years were pooled.

Gonad maturity was determined according to gross criteria using the scale given by KJØRSVIK *et al.* (1987). Thus stage 5 was maturing fish, stage 6 fish immediately before commencing of spawning, stages 7 fish with running gonads, and stage 8 fish with spent gonads. Classification of male gonads generally followed a simplified scale in that stage 5 and 6 were lumped together as "maturing fish". This was mainly due to difficulties in separating between these two stages.

In order to examine the gonad weight-to-total weight relationship, an equation of the type  $GW = \alpha + \beta \cdot W$  was determined by linear regression.

The relative gonad weight, or the gonosomatic index, GI, was then examined using the ratio:

$$GI = 100 \cdot GW/W$$

The variation in GI with maturity stage and time of the year was studied by analysis of variance (ANOVA).

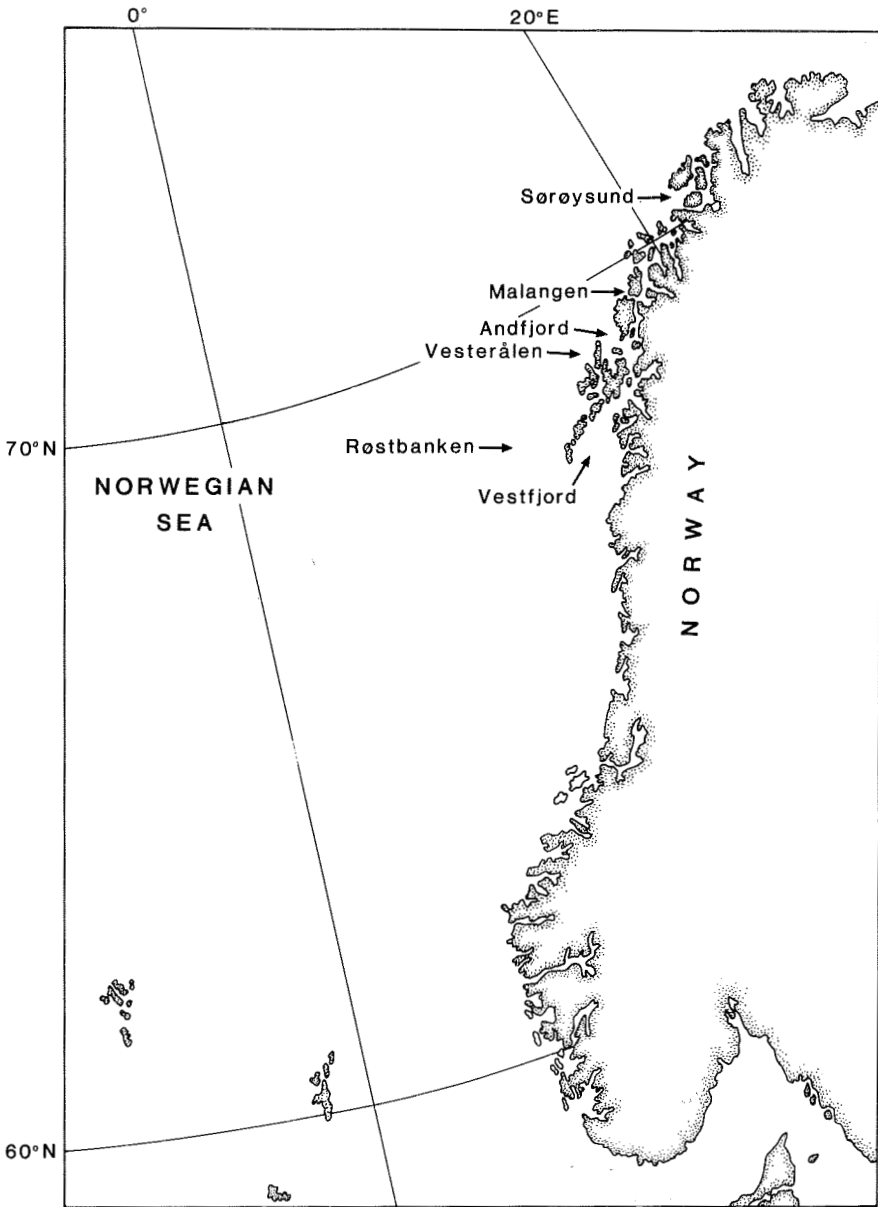


Fig. 1. The sampling sites in northern Norway.

In examining the relative liver and carcass weights, cubic growth, or isometry, of the tissues cannot be assumed *a priori*. The effect of fish lengths, which varied substantially in the present material, were, therefore, eliminated by using relative indices based on empirical length-weight relationships (LE

CREN 1951). Although several regression types can be fitted to length-weight relationships, the best fits are most often provided by a power-regression  $W_t = \alpha \cdot TL^\beta$ , which in its linearized form can be expressed as  $\ln W_t = \ln \alpha + \beta \cdot \ln TL$ , where  $W_t$  is the weight of the tissue, and  $\alpha$  and  $\beta$  are the regression constant and regression coefficient, respectively (RICKER 1975).

In this paper, therefore, we chose to fit power functions to the liver weight-length and eviscerated weight-length relationships in order to estimate the precise  $\beta$ -values. All  $\beta$ -values are tested for homogeneity among fish in various maturity stages using analysis of covariance (ANCOVA). Provided homogeneity was present, pooling of the data was performed in order to calculate common  $\beta$ -values, irrespective of maturity stages, to be used in liver and condition indices. These indices, showing the relative sizes of liver and carcass, were defined as follows:

Liver index:

$$LI = 1000 \cdot LW/TL^\beta$$

Condition factor (indicating relative carcass size):

$$K = 100 \cdot W/TL^\beta.$$

Potential variation with maturity stage or time of the year in LI and K were analyzed by means of ANOVA.

Statistics were provided from the BMDP (DIXON 1981) programs P1R (multiple linear regression), P1V (one-way analysis of variance and covariance), and P3D (comparison of two groups with t-tests) run on a VAX computer.

## RESULTS

### GONAD SIZE

Regression analyses of the gonad weight-eviscerated weight relationships in males and females of various maturity stages and of the whole data set for each sex, were significant ( $p < 0.05$ ) in all cases (Table 1). ANCOVA indicated, however, that the regression coefficients ( $\beta$ ) of the different maturity stage groups were not homogeneous either in females ( $F_{3,66} = 9.283$ ;  $p < 0.001$ ) or in males ( $F_{2,372} = 7.899$ ;  $p < 0.001$ ). The highest  $\beta$ -values of females were recorded in fish with maturing gonads (stage 6), while in males, fish with running gonads (stage 7) had the highest  $\beta$ -value (Table 1). In both sexes, the lowest  $\beta$ -values were recorded in fish with spent gonads.

The highest mean values of the gonosomatic index, GI, were observed in females in maturity stage 6; thereafter a decrease in mean gonad index with increasing maturity stage was observed (Fig. 2). In males the highest GI-values were recorded in fish in maturity stage 5 + 6 (Fig. 2). Analyses of variance revealed significant heterogeneity among stages both in females ( $F_{3,70} = 24.377$ ;  $p < 0.001$ ) and in males ( $F_{2,375} = 39.646$ ;  $p < 0.001$ ).

Table 1. Relationship for fish in various maturity stages of gonad weight (GW) to eviscerated weight (W) described by linear regression equations, and of liver and carcass weights (LW and W) to total fish length (TL) described by power regression equations. N is the number of fish examined,  $\alpha$  and  $\ln\alpha$  are the regression constants,  $\beta$  is the regression coefficient, and  $r^2$  is the coefficient of correlation. In the regressions,  $H_0: B = 0$ ; thus, rejection ( $p < 0.05$ ) means that the variation in weight can be explained by regression. \* = rejection at  $0.01 < p < 0.05$ ; \*\* = rejection at  $0.001 < p < 0.01$ ; \*\*\* = rejection at  $p < 0.001$ ; ns =  $H_0$  accepted (i.e.  $p > 0.05$ ).

GONAD MATURITY STAGE	GW = $\alpha \cdot \beta \cdot W$					$\ln LW = \ln\alpha + \beta \cdot \ln TL$					$\ln W + \ln\alpha + \beta \cdot \ln TL$				
	N	$\beta$	$\alpha$	$r^2$	p	N	$\beta$	$\ln\alpha$	$r^2$	p	N	$\beta$	$\ln\alpha$	$r^2$	p
FEMALES															
5	37	0.0751	1944.61	0.29	***	63	3.2011	-3.98	0.76	***	39	3.0909	-2.16	0.97	***
6	12	0.2689	-5079.47	0.89	***	13	3.7161	-5.31	0.86	***	12	3.3013	-2.63	0.97	***
7	10	0.0635	2584.71	0.55	*	13	3.6884	-5.33	0.86	***	14	3.3926	-2.85	0.98	***
8	15	0.0217	215.46	0.37	*	21	3.7879	-5.60	0.74	***	20	3.0361	-2.05	0.94	***
Combined	74	0.0772	1228.33	0.28	***	110	2.8419	-3.30	0.49	***	85	3.1293	-2.25	0.97	***
MALES:															
5+6	38	0.0580	148.23	0.15	*	42	3.6207	-5.07	0.67	***	38	3.0808	-2.14	0.96	***
7	304	0.0647	-144.76	0.65	***	305	2.9277	-3.87	0.78	***	308	3.1203	-2.26	0.98	***
8	36	0.0070	21.38	0.61	***	36	3.2185	-4.59	0.79	***	36	3.1227	-2.28	0.99	***
Combined	378	0.0644	-150.93	0.51	***	383	3.2209	-4.45	0.77	***	382	3.1493	-2.32	0.97	***

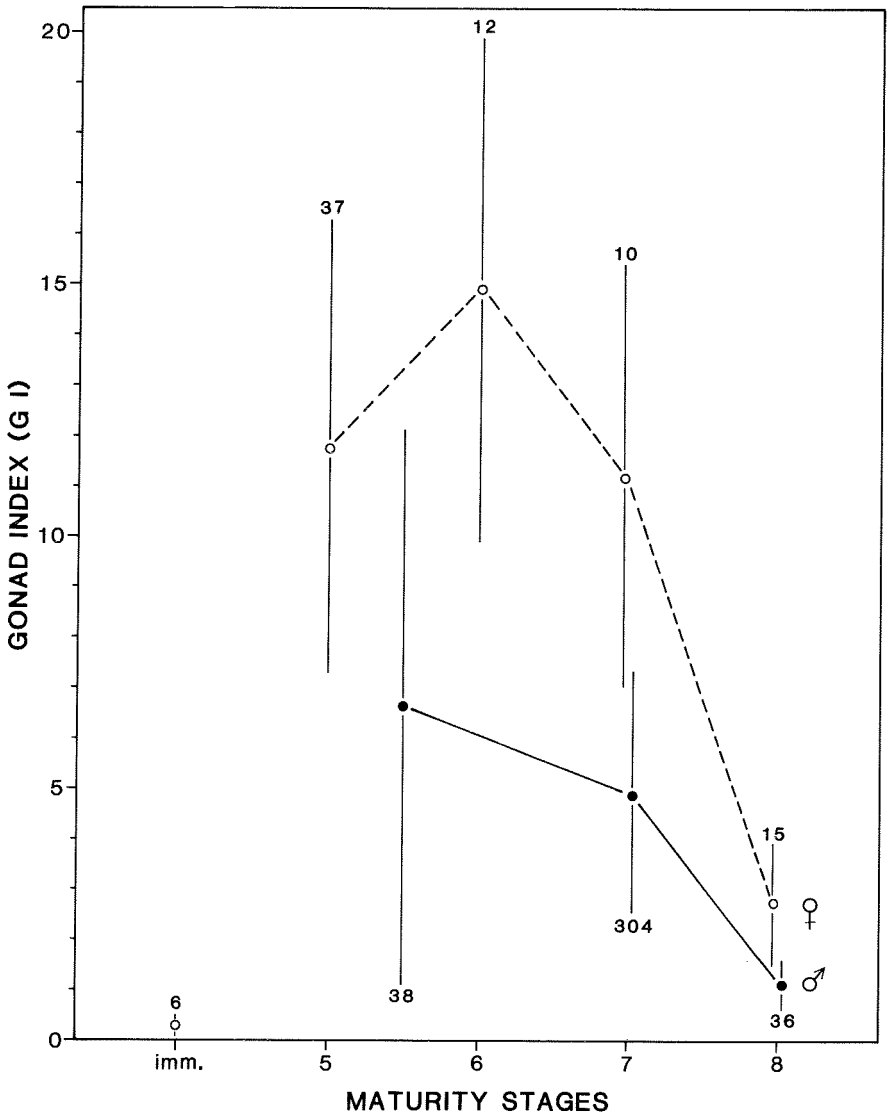


Fig. 2. Mean gonad indices (GI) per maturity stage of halibut females (stipled line). The mean values of males are also given (solid lines). Standard deviations are indicated by the bars and the numbers of examined fish are given.

In both sexes the mean GI values increased during autumn, reaching the highest values in November (males) and December (females). Thereafter, the average gonad sizes decreased (Fig. 3). ANOVA revealed that the observed variation in the monthly means of GI was significant both in males ( $F_{6,371} = 40.919$ ,  $p < 0.001$ ) and in females ( $F_{6,67} = 2.317$ ,  $p = 0.043$ ).

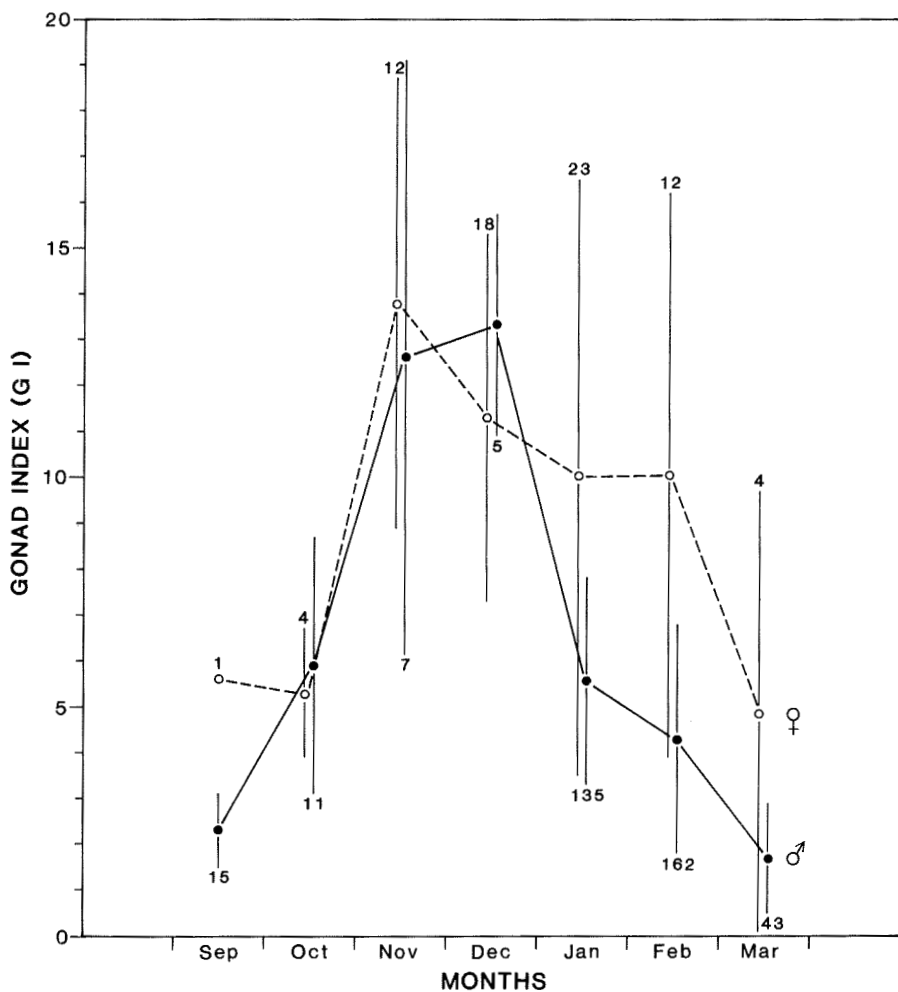


Fig. 3. Mean monthly gonad indices (GI) for halibut males (solid line) and females (stipled line). Standard deviations are indicated by the bars and the numbers of examined fish are given.

#### LIVER CONDITION

Power curve regressions fitted to the liver weight-total length data were highly significant ( $p < 0.001$ ) in both males and females in all maturity stages (Table 1). ANCOVA showed that the regression slopes ( $\beta$ ) from the different gonadal maturity stages were homogeneous in females ( $F_{3,102} = 0.678$ ;  $p = 0.568$ ) as well as in males ( $F_{2,377} = 1.867$ ;  $p = 0.156$ ), thus permitting the use of combined  $\beta$ -values (2.8419 and 3.2209 for females and males, respectively, Table 1) when calculating the liver index in each of the sexes. Both the combined regressions were highly significant ( $p < 0.001$ ). The liver indices thus became:

Females:

$$LI = 1000 \cdot LW/TL^{2.8419}$$

Males:

$$LI = 1000 \cdot LW/TL^{3.2209}$$

A general decrease in mean LI was observed with increasing maturity stage in adult fish of both sexes (Fig. 4). ANOVA revealed that the observed intermaturity stage heterogeneity of LI was highly significant both in males ( $F_{2,380} = 24.057$ ,  $p < 0.001$ ) and in females ( $F_{3,106} = 33.719$ ,  $p < 0.001$ ). In immature females the mean LI was similar to those observed in maturity stage 6–8 females, but significantly lower ( $p < 0.001$ ) than those observed in stage 5 females.

ANOVA shows that the monthly mean values of liver indices (Fig. 5) vary significantly during the period of investigation in both males ( $F_{6,376} = 9.684$ ,  $p < 0.001$ ) and females ( $F_{6,103} = 12.585$ ,  $p < 0.001$ ). In the males, the mean LI decreased clearly during the whole period from September to March inclusive. In females, relative liver weight did not start to decrease until December. After January, a slight increase in the mean LI of females again occurred.

#### *BODY CONDITION*

Power curve regressions fitted to the eviscerated weight-total fish length data of females and males in various stages of gonadal maturity were highly significant ( $p < 0.001$ ) in all cases (Table 1). ANCOVA indicated that the regression coefficients of the different maturity stages were homogeneous in females ( $F_{3,77} = 1.382$ ;  $p = 0.255$ ) as well as in males ( $F_{2,376} = 0.067$ ;  $p = 0.935$ ). This permitted the use of combined  $\beta$ -values (3.1293 for females, 3.1493 for males, Table 1) when calculating the condition factor in each of the sexes. The condition factors (K) thus became:

Females:

$$K = 100 \cdot W/TL^{3.1293}$$

Males:

$$K = 100 \cdot W/TL^{3.1493}$$

As revealed by ANOVA, mean values of the condition factor calculated for sexually mature fish in various maturity stages (Fig. 4) vary significantly in males ( $F_{2,379} = 14.391$ ,  $p < 0.001$ ), but not in females ( $F_{3,81} = 1.965$ ,  $p = 0.126$ ). In males, a general decrease in mean K with increasing maturity stage was observed. No significant differences ( $p < 0.05$ ) were observed between the mean K-value for immature females and those observed in mature females.

In males, the mean monthly K-values were quite stable in autumn (September–November), followed by a decrease to a lower level maintained from



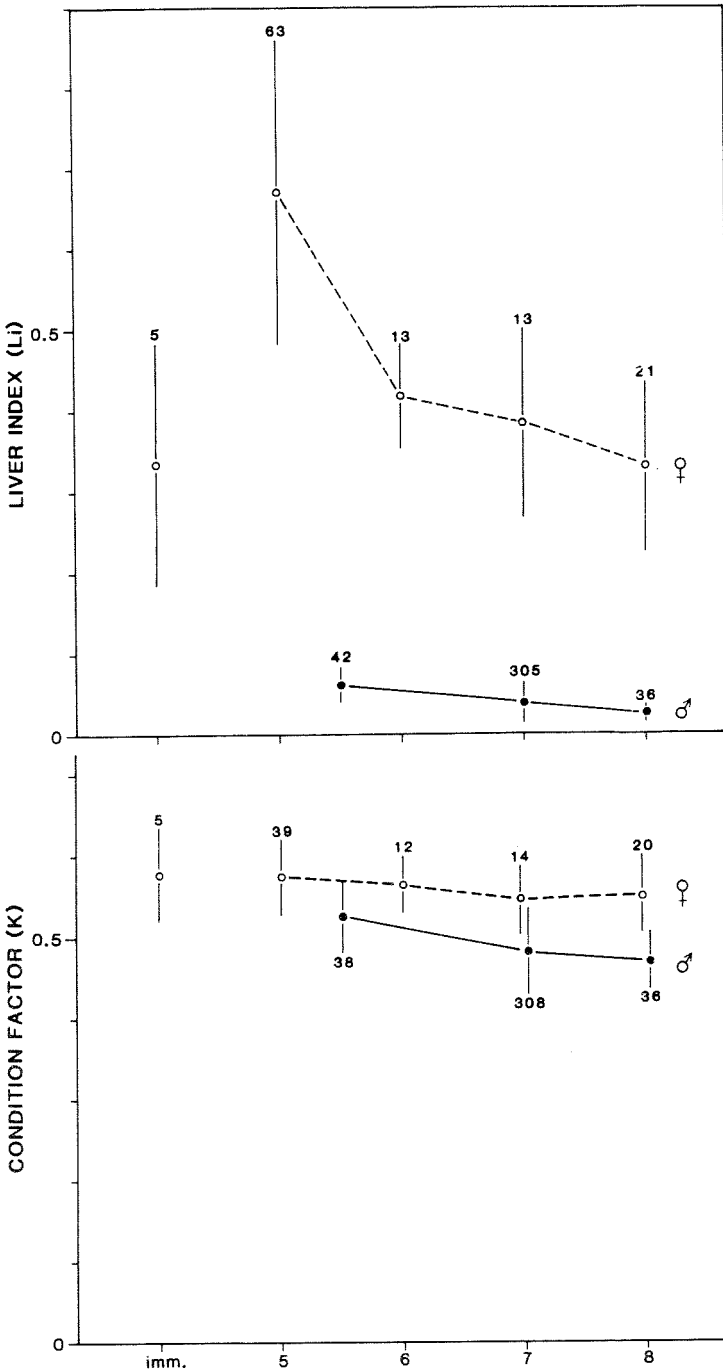


Fig. 4. Mean liver indices (LI, above) and condition factor (K, below) per maturity stage of halibut males (solid lines) and females (stipled lines). Standard deviations (bars) and numbers of examined fish are given.

December through March (Fig. 5). ANOVA revealed that this observed heterogeneity among monthly mean values of the condition factor in males was highly significant ( $F_{6,375} = 5.795$ ,  $p < 0.001$ ). The female monthly mean values of K appeared to vary in the same manner as for the males (Fig. 5), but an analysis of variance revealed no significant variation between months in the females ( $F_{6,78} = 1.749$ ,  $p = 0.121$ ).

#### DISCUSSION

The observed significant heterogeneity among slopes ( $\beta$ -values) of regressions of gonad weights against eviscerated weights indicates that this relationship varies with the stage of maturity both in female and male halibut, i.e., fish have gonads of different relative weight proportions depending on their state of maturity. In the females, fish in maturity stage 6, namely the last phase of vitellogenesis before the oocytes start to absorb fluid and are released from the ovary follicles, had the steepest slope (Table 1). This indicates that the larger females in this stage generate larger ovaries in proportion to eviscerated body size than do the smaller females, as compared with other maturity stages. According to DE VLAMING *et al.* (1982), who critically reviewed the use of the gonosomatic index in studies of breeding in fishes, such heterogeneity among females in different maturity stages is commonly observed also in other fish species. These authors discussed the biological significance of such size-related heterogeneity, and especially pointed out that body size might have a greater impact on relative ovary weight when females had "ripe" ovaries than when ovaries were inactive. This is highly consistent with our present observations. There seems to be less variation of  $\beta$ -values with maturity stage in males than in females, although the pooling of stages 5 and 6 in the males complicates more precise intersexual comparison.

At the individual level, the mean GI's reached their highest values in fish in maturity stage 6 (females) or 5 + 6 (males) (Fig. 2), which is quite natural since this is the latest stage of gonad maturity before the spawning starts and drains the gonads of gametes and weight. At the population level, the continuous decreases in mean GI from November–December to March (Fig. 3) is consistent with the conclusion of KJØRSVIK *et al.* (1987) that this is the spawning season of the species. The increase in mean GI values from September to November–December emphasizes that this is a period of intense accumulation of reproductive tissues by the species. The observed heterogeneities of regression slopes mean that dividing gonad weight by eviscerated weight may have a different result depending on the phase of the gonad cycle. Thus, the GI may provide a misleading indication of gonadal activity, and the validity of comparing mean values among maturity stages and months is, therefore, debatable.

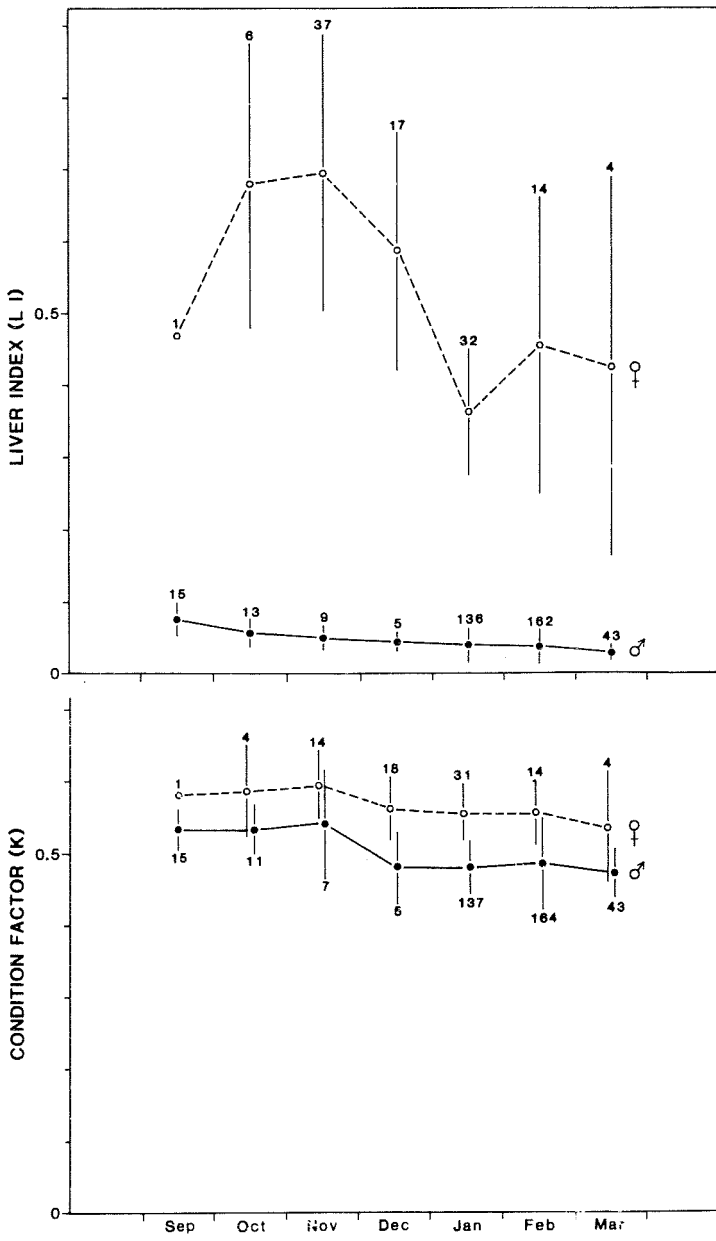


Fig. 5. Mean monthly liver indices (LI, above) and condition factor (K, below) for halibut males (solid lines) and females (stippled lines). Standard deviations (bars) and numbers of examined fish are given.

No significant variation with stage of maturity was observed in the  $\beta$ -values either in the regression of liver weight on total length or in the regression of eviscerated weight on total length. This applied to both sexes, and indicates that single regressions, based on pooled material irrespective of maturity stage, can be expected to adequately describe both the above-mentioned relationships for female as well as male halibut.

It is generally accepted that in flatfish, the main energy reserves are usually deposited in the liver and muscle tissues (LOVE 1970). Our results suggest, however, that in halibut these two different tissue types are differently affected by the costs of spawning. Obviously, the liver is significantly reduced as the maturation process proceeds (Fig. 4), leading to a significant decrease in mean relative liver weight (LI) throughout the spawning season (Fig. 5). Thus, it seems reasonable to conclude that the liver is an important energy source for female and male halibut during preparation for spawning. This is supported by studies of the lipids and fatty acid profiles of the species (HAUG *et al.* 1988). The carcass seems, however, much less affected by the energy expenditures involved in the seasonal accumulation of the reproductive tissues. Certainly, the males show a little, but still significant, decrease in relative body weight during gonad maturation (Fig. 4) and during the first part of the spawning season (Fig. 5). This was, however, not the case in females whose sacrifice of general body weight in order to build up ovaries seems minimal. The observed difference between females and males in mobilizing muscle tissue into reproductive tissue seems to support previous suggestions of sexual differences in physiology and growth/energy strategies of male and female halibut. These differences are also manifested in the female growth rate being far in excess of the males after the attainment of sexual maturity (MATHISEN and OLSEN 1968, JAKUPSSTOVU and HAUG 1988), and in the generally higher lipid levels in several tissues of females as compared with males during spawning (HAUG *et al.* 1988).

In general, the energy reserves of flatfish are considerably depleted during the course of gonad development or starvation which usually accompanies overwintering and spawning (DAWSON and GRIMM 1980, JOBLING 1980, ROFF 1982). With its low level of general body weight sacrifice during spawning, the Atlantic halibut follows this trend only in part. No doubt the food intake of the species is reduced during spawning (DEVOLD 1938, MCINTYRE 1953). We suggest, however, that feeding most probably does not cease completely, and that the general physical activity, particularly in females, is reduced to such an extent that a minimal loss of energy is ensured during spawning.

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