FATTY ACID COMPOSITION OF HERRING (*CLUPEA HARENGUS* L.) LARVAE OFF WESTERN NORWAY

ARILD FOLKVORD, INGER MARGRETHE HAALAND, OTTO GRAHL-NIELSEN & ARNE JOHANNESSEN

SARSIA

FOLKVORD, ARILD, INGER MARGRETHE HAALAND, OTTO GRAHL-NIELSEN & ARNE JOHANNESSEN 1996 02 27. Fatty acid composition of herring (*Clupea harengus* L.) larvae off western Norway. – *Sarsia* 80:299-305. Bergen. ISSN 0036-4827.

Herring larvae (*Clupea harengus* L.) were collected off western Norway in April 1991, 1992, and 1993. The larvae were length measured, staged and analysed individually for fatty acid content. Both absolute and relative amounts of individual fatty acids were used as input in principal component analyses (PCA), and the results revealed significant size and stage specific trends in composition. These differences were attributed to the same fatty acids all three years. Qualitative differences in the fatty acid composition of larvae with above average and below average total fatty acid content within respective developmental stages indicate possible maternal effects and effects of previous feeding history. The efforts on using fatty acid profiles as nutritional indices of the individual herring larvae will therefore be continued.

Arild Folkvord and Arne Johannessen, Department of Fisheries and Marine Biology, University of Bergen, Bergen High-Technology Center, N-5020 Bergen, Norway. E-mail: arild.folkvord@ifm.uib.no – Inger Margrethe Haaland and Otto Grahl-Nielsen, Department of Chemistry, University of Bergen, Allégt. 4, N-5020 Bergen, Norway.

INTRODUCTION

Growth of fish larvae affects survival both directly and indirectly. If growth is very low or negative over a certain time period, the larvae will die of starvation. In addition growth determines the time the larvae will stay in any given size interval and thus be susceptible to various predators (ANDERSON 1988). The development of methods to assess growth and nutritional state of fish larvae in the field is therefore of great importance in recruitment studies, and several methodologies have been proposed (e.g. CLEMMESEN 1989; THEILACKER 1986; SUTHERS & al. 1992). The use of fatty acid composition as an index of nutritional state in herring, *Clupea harengus* L., larvae is motivated by: a) absolute and relative amounts of fatty acids are expected to contain information about past growth history (FRASER & al. 1987; VAN DER MEEREN 1993), b) it is easily accessible (analytic methodology well described), c) analysis can be undertaken on individual larvae (sensitive method), and d) the data are suitable for powerful multivariate statistical analysis (GRAHL-NIELSEN & BARNUNG 1985; BARNUNG & GRAHL-NIELSEN 1987; HOVE & GRAHL-NIELSEN 1991).

The requirement of essential fatty acids for successful development of marine fish larvae is widely recognized (WATANABE 1993). Larvae of many fish species like turbot, cod, herring and mahi-mahi (e.g. WITT & al. 1984; FRASER & al. 1987, 1988; OLSEN & al. 1991; KRAUL & al. 1992) have been studied, all showing the importance of the n3 polyunsaturated fatty acids. In this study the emphasis has been put at analysing individual larvae found in the field. The need to analyse larvae on an individual basis is essential when the goal is to use the fatty acid composition as an index of nutritional state. By using larvae found in the field we also expect to obtain a measure of the variability in larval fatty acid composition under natural conditions. We sampled herring larvae in 1991, 1992, and 1993 to see if the emerging pattern in composition during the early larval stages was consistent between years.

MATERIAL AND METHODS

The herring larvae were collected off south-western Norway during three separate cruises in mid-April 1991-1993 onboard RV *Håkon Mosby*. In 1991 and 1992 the larvae were sampled north of 60° N in April 7-13 and April 5-13 respectively, and in 1993 the larvae were sampled south of 60° N in April 15-22 (Fig. 1). Vertical hauls with an 80 cm diameter net with 375 µm mesh (T-80) were taken from 150 m depth (or from 10 m above the bottom) to the surface. The larvae were immediately sorted out from the sample and counted onboard. Standard lengths of larvae used for fatty acid analysis were measured prior to preservation (see below) to the nearest 0.1 mm under a dissecting microscope (1992 and 1993) or to the nearest mm below using mm paper (1991). The larvae were simultaneously staged according to DOYLE (1977) with the inclusion of an extra stage, 1d, representing larvae with no visible yolk remains or dorsal fin

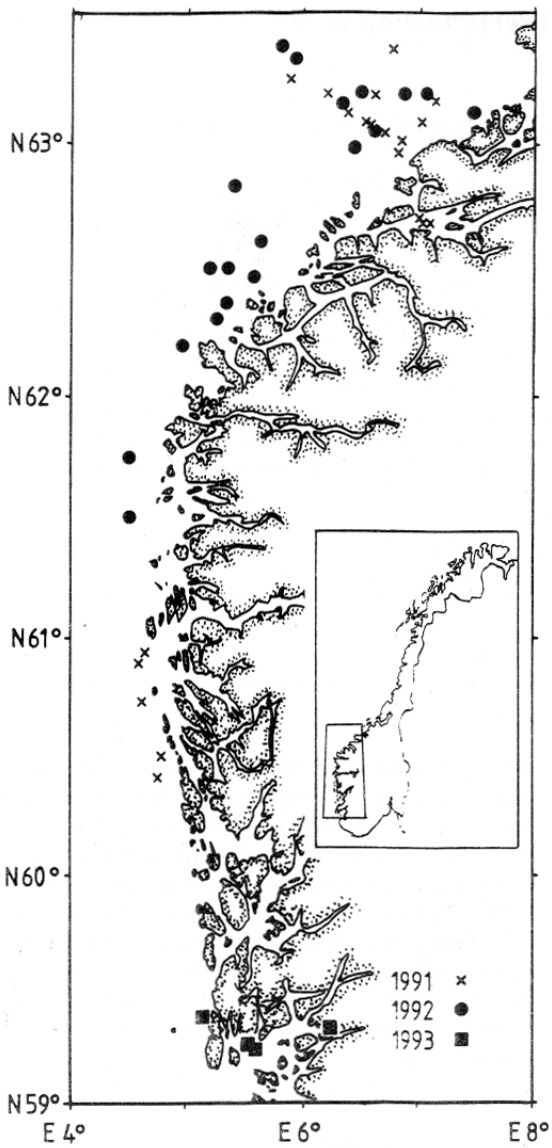

Fig. 1. Location of stations where larval samples for fatty acid analysis were taken in 1991-1993.

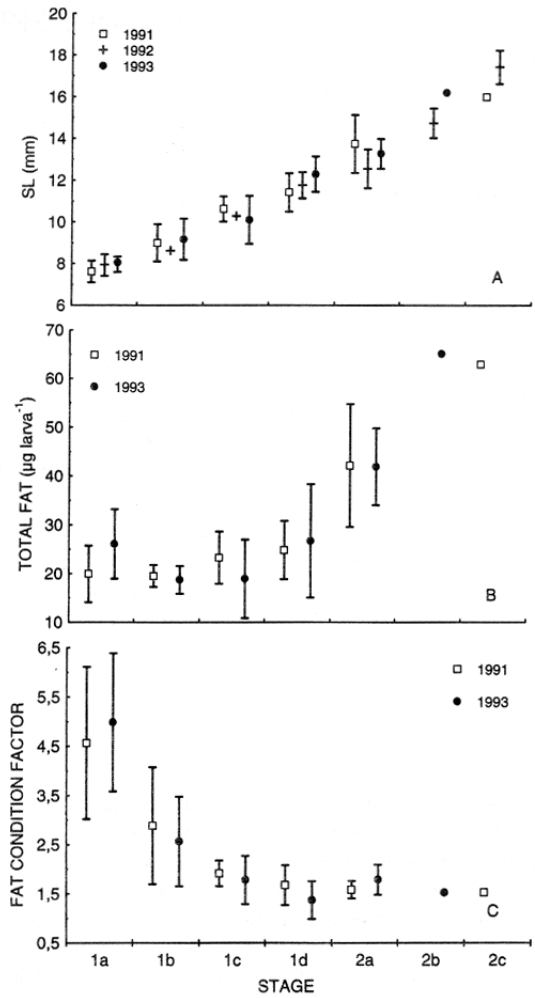

Fig. 2. Stage specific: A) standard length (mm) of herring larvae, B) total fatty acid content (µg larva^{-1}) of herring larvae, C) fat condition factor (100*total fatty acid/SL^3) of herring larvae. Whiskers indicate standard deviation, stages according to DOYLE (1977) and ØIESTAD (1983).

structures (ØIESTAD 1983). A maximum of 20 larvae were used per station to enable rapid preservation after sampling.

After staging and measuring, external water on the larvae was removed by letting them dry on a Teflon plate before they were transferred to thick-walled glass-tubes, one larva per tube. Anhydrous methanol, 300 µl, containing 2N HCl, was added to the tube. For the 1991 samples, the methanol solution contained the saturated fatty acid, 21 : 0, as internal standard, 0.6 µg per larva. In 1992 only qualitative analyses were performed, no internal standard was used. For the 1993 samples the internal standard, 21 : 0, in chloroform solution, was added to the tubes first, 1.53 µg per tube. The chloroform was evaporated before the larva was transferred to the glass-tube. After the HCl/MeOH was added, the glass-tubes were securely tightened with Teflon-lined screw-caps. The tubes were stored on board approx. one week at -18° C before analysis on land. The tubes were then placed in an oven at 100° C for 15 hours for complete methanolysis. The HCl/methanol solution was then evaporated down to approx. 200 µl under a stream of nitrogen before an equivalent amount of water was added. The fatty acid methylesters were extracted from the methanol/water phase with 2 x 450 µl hexane by mixing the phases on a whirl-mixer followed by centrifugation to separate the phases.

One microliter of the hexane solution was gas chromatographed on a 30 m x 0.32 mm fused silica column, from J &W Scientific, with 50 % cyanopropylmethyl- 50 %-methyl-phenyl polysiloxane, of 0.25 µm thickness, as stationary phase, and helium as mobile phase. The components eluting from the column were detected by a flame ionization detector, and the detector output was coupled to a VG Multichrome lab-data system for storage and treatment of the chromatograms. A total of 17 fatty acids were identified by comparison with a standard mixture (GLC reference standard no. 68B from Nu-Chek-Prep, Inc., Elysian, Minn., USA). The amounts of the 17 fatty acids were determined by integration of the areas of their peaks. For the 1991 and 1993 samples the absolute amounts of the fatty acids per larva were determined by comparison with the internal standard. The relative amounts were expressed as percentages of the sum.

A fat condition factor (FCF) was calculated as : $FCF=TF \cdot 100/SL^3$ where TF is total fatty acid content in µg $larva^{-1}$ and SL is standard length in mm. Stage and year specific univariate tests were calculated with 2-way ANOVA and the response variables were occasionally log transformed to correct for heteroscedasticity and non-normality of errors. Tukey HSD tests were used for post-hoc comparisons. A total of 217 larvae were preserved in 1991-1993. Six of the larvae were not staged and 9 larvae were excluded from further analysis for analytical reasons leaving 86, 71, and 45 larvae left for analysis from 1991, 1992 and 1993 respectively (Table 1).

For multivariate statistics, the amounts of the fatty acids were logarithmically transformed to level out the large differences in numerical values between the least and the most abundant fatty acids. With each sample defined by 17 log-transformed variables (fatty acids), we used principal component analysis to extract the two coordinates (principal components, PC's) that described the direction of the largest and second largest variance among the samples. In this manner the relationship among the samples, can be displayed in two dimensions without considerable loss of the total original variance and with conservation of the systematic variance among the samples.

RESULTS

Average standard lengths of larvae increased linearly from stage 1a to 2c, and were similar between years (Fig. 2A). Considerable overlap in length distributions were found

Table 1. Fatty acid composition (in % ± SD) in stages 1a to 2c from 1991 to 1993. Total fat content in µg $larva^{-1}$, n is number of larvae analyzed. Larvae from stages 1a to 1c contain visible yolk remains (Doyle 1977).

	Stage 1a			Stage 1b			Stage 1c		
Fatty acids	1991	1992	1993	1991	1992	1993	1991	1992	1993
14:0	3.1±0.4	4.2±0.9	4.4±0.7	3.0±0.7	3.3	3.7±0.3	2.5±0.2	3.0	3.5±1.4
16:0	26.5±2.0	26.6±2.9	27.4±4.8	26.1±1.4	25.6	29.6±3.0	26.1±1.2	26.2	29.8±6.0
16:1 (n-7)	2.9±0.2	3.4±0.5	3.5±0.5	3.0±0.5	2.9	2.9±0.4	2.5±0.2	3.1	2.4±0.3
16:1 (n-5)	0.6±0.1	0.6±0.1	0.5±0.1	0.6±0.1	0.5	0.4±0.1	0.5±0.1	0.5	0.4±0.1
18:0	4.7±0.5	4.1±0.7	4.5±1.5	4.6±0.5	3.9	6.1±1.3	5.0±0.4	4.6	7.2±2.1
18:1 (n-9)	8.9±3.4	9.9±0.9	10.1±0.6	9.5±1.4	8.9	9.3±0.9	9.1±1.1	8.8	8.3±1.0
18:1 (n-7)	3.6±0.7	3.4±0.2	3.5±0.3	3.6±0.3	3.4	3.4±0.4	3.6±0.3	3.8	3.2±0.5
18:1 (n-5)	0.8±0.2	0.8±0.1	0.6±0.0	0.8±0.1	0.7	0.6±0.1	0.7±0.2	0.7	0.6±0.1
18:2 (n-6)	1.1±0.1	1.2±0.1	1.1±0.2	1.1±0.1	1.1	0.9±0.1	1.1±0.4	1.2	1.1±0.2
18:3 (n-3)	0.9±0.2	1.1±0.2	0.8±0.2	0.9±0.2	0.8	0.6±0.2	0.8±0.1	0.8	0.6±0.2
18:4 (n-3)	0.7±0.2	1.0±0.1	0.6±0.2	0.7±0.2	0.7	0.6±0.1	0.9±0.4	1.1	0.9±0.5
20:1 (n-9)	1.7±0.3	1.6±0.1	1.5±0.3	1.6±0.4	1.2	1.1±0.4	1.3±0.2	1.0	0.8±0.3
20:4 (n-6)	0.7±0.1	0.7±0.1	0.8±0.1	0.8±0.3	0.9	0.9±0.1	1.2±1.2	0.9	1.0±0.2
20:5 (n-3)	11.1±0.5	11.4±1.1	12.5±1.9	10.5±1.4	12.7	11.4±1.3	10.6±0.7	12.9	11.3±2.7
22:5 (n-3)	0.9±0.2	0.3±0.2	1.0±0.3	0.8±0.2	0.7	0.9±0.3	0.8±0.2	0.2	0.8±0.2
22:6 (n-3)	29.9±1.2	28.7±2.9	26.1±3.5	31.5±2.5	31.6	26.7±3.4	32.0±1.4	30.1	26.9±4.8
24:1 (n-9)	2.1±1.6	1.2±0.2	1.2±0.4	1.0±0.2	0.9	1.0±0.3	1.3±0.5	1.3	1.3±0.6
Saturates	34.3±2.4	34.9±4.2	36.3±6.3	33.6±1.7	32.8	39.4±3.7	33.7±1.5	33.8	40.5±8.8
Monoenes	20.6±3.0	20.8±0.7	20.8±1.3	20.0±2.2	18.5	18.6±1.7	19.0±1.3	19.2	16.9±2.1
Polyunsat.	45.2±2.0	44.4±3.8	42.9±5.6	46.3±2.8	48.5	41.9±4.5	47.3±1.8	47.2	42.6±7.8
(n-3)	43.4	42.5	40.9	44.4	46.5	40.1	45.0	45.1	40.6
(n-6)	1.8	1.9	1.9	1.9	2.0	1.8	2.3	2.1	2.0
(n-3) : (n-6)	24.3	22.3	21.3	23.6	23.3	22.7	19.7	21.5	20.1
Total (µg)	19.9±5.8		26.1±7.1	19.5±2.3		18.7±2.9	23.3±5.4		19.0±8.1
n	8	6	11	6	1	7	19	1	14

between consecutive sub-stages (e.g. 1b and 1c), whereas virtually no overlap in length was found between stages which were three sub-stages apart (e.g. 1a and 1d). At the same time, no consistent differences in total fatty acid content were found between stages 1a and 1d ($p > 0.05$, 2-way ANOVA, Fig. 2B). Larvae from stage 2a, on the other hand, had substantially higher fatty acid content than all the other stage 1 larvae, except for the 1993 stage 1d larvae ($p < 0.05$, 2-way ANOVA). The fat condition factor indicated that the relative fatty acid content was highest among the early larvae and declined towards the end of the yolk sac stage ($p < 0.05$, 2-way ANOVA, Fig. 2C). No further decline was observed beyond stage 1d.

The most abundant fatty acid in most years and stages was 22:6n3 (docosahexaenoic acid, DHA) followed by 16:0 and 20:5n3 (eicosapentaenoic acid, EPA) (Table 1). In all years there was a declining trend in the proportion of monoenes during the yolk sac stages, and in 1991 and 1993 this was mirrored by an increase in proportion of polyunsaturated fatty acids (Table 1). The ratio of n3 : n6 fatty acids was somewhat higher in stage 2 (feeding stages) than in stage 1 (yolk sac stages). The n3 : n6 ratio was also consistently higher in 1991 than in 1992, which again was higher than in 1993 (Table 1).

The larvae with above average total fatty acid content within each stage and year, also had a higher proportion of the 20:5n3 fatty acid than the larvae with below average content ($p < 0.05$, 2-way ANOVA, Fig. 3A). The larvae at stage 2a also had a higher proportion of the same acid than the other stage 1 larvae ($p < 0.05$, 2-way ANOVA, Fig. 3A). The 22:6n3 fatty acid was also relatively more abundant in stage 1 larvae with above average total fat content, but in contrast to the 20:5n3 acid, the proportion dropped somewhat among the 2a larvae (Fig. 3B). The larvae with above average total fatty acid content also had a higher relative abundance of fatty acids than the below average larvae as determined by the fat condition factor ($p < 0.05$, 2-way ANOVA, Fig. 3C). This effect was especially apparent in the earliest yolk sac stages where the relative fatty acid content was highest ($p < 0.05$, 2-way ANOVA, Fig. 3C).

Table 1. Cont.

	Stage 1d			Stage 2a			Stage 2b		Stage 2c	
Fatty acids	1991	1992	1993	1991	1992	1993	1992	1993	1991	1992
14:0	2.4±0.3	3.2±0.5	3.4±0.4	2.4±0.4	3.2±0.4	2.4±0.1	3.3±0.4	2.4	2.1	3.5±0.6
16:0	25.9±2.4	29.0±4.2	28.9±5.8	24.2±1.1	26.5±1.5	24.0±1.5	27.5±3.8	24.2	24.3	28.3±4.5
16:1 (n-7)	2.5±0.2	3.2±0.5	2.9±0.4	2.7±0.2	3.9±0.5	2.4±0.2	4.6±0.7	2.5	2.4	4.8±0.5
16:1 (n-5)	0.5±0.1	0.5±0.1	0.4±0.1	0.4±0.1	0.4±0.1	0.4±0.0	0.4±0.1	0.3	0.4	0.4±0.1
18:0	5.2±0.6	5.1±1.1	5.7±1.4	4.6±0.4	4.6±0.6	4.9±0.5	4.1±0.8	4.3	4.3	3.9±0.5
18:1 (n-9)	8.5±1.1	7.4±0.8	7.0±0.6	7.0±1.1	7.0±0.8	7.0±0.6	7.7±1.2	6.8	7.7	7.4±0.8
18:1 (n-7)	3.5±0.4	3.8±0.5	3.3±0.3	3.3±0.6	4.2±0.7	3.2±0.3	5.2±0.8	3.2	3.8	5.3±0.3
18:1 (n-5)	0.6±0.4	0.6±0.1	0.7±0.1	0.8±0.1	0.5±0.1	0.8±0.1	0.5±0.1	0.9	1.0	0.4±0.0
18:2 (n-6)	1.0±0.1	1.1±0.1	1.2±0.2	1.0±0.1	1.1±0.1	1.3±0.1	1.0±0.1	1.3	1.0	0.9±0.1
18:3 (n-3)	0.8±0.2	0.7±0.2	0.9±0.3	1.0±0.2	0.6±0.1	1.2±0.2	0.5±0.2	1.4	1.1	0.6±0.1
18:4 (n-3)	1.2±0.5	1.0±0.5	1.4±0.5	1.7±0.6	1.3±0.4	1.7±0.5	1.4±0.6	2.3	1.9	1.6±0.4
20:1 (n-9)	1.1±0.3	0.8±0.2	0.5±0.2	0.8±0.2	0.7±0.3	0.5±0.0	0.8±0.3	0.4	0.6	0.8±0.2
20:4 (n-6)	1.0±0.6	0.8±0.2	0.9±0.2	0.7±0.1	0.7±0.1	0.8±0.1	0.6±0.1	0.7	0.5	0.6±0.2
20:5 (n-3)	11.1±1.3	12.4±2.1	14.1±3.2	13.7±1.5	15.5±1.8	16.4±1.0	16.9±2.4	17.6	13.4	16.5±2.2
22:5 (n-3)	0.6±0.1	0.5±0.4	0.8±0.2	0.6±0.1	0.4±0.2	0.8±0.1	0.5±0.3	0.8	0.6	0.7±0.3
22:6 (n-3)	32.5±2.5	28.7±3.9	26.3±3.3	32.7±1.3	28.2±2.2	30.4±1.5	24.0±3.0	28.1	33.0	23.0±4.3
24:1 (n-9)	1.4±0.9	1.1±0.2	1.6±0.3	2.5±1.0	1.2±0.3	1.9±0.6	1.3±0.2	2.7	1.8	1.3±0.4
Saturates	33.5±3.0	37.4±5.5	38.0±7.4	31.1±1.2	34.2±1.9	31.3±1.6	34.9±4.6	30.9	30.7	35.7±5.5
Monoenes	18.2±1.5	17.4±1.3	16.5±1.1	17.5±2.1	18.1±1.3	16.2±1.4	20.3±2.0	16.8	17.7	20.4±13
Polyunsat.	48.3±3.6	45.2±5.4	45.6±7.2	51.4±2.5	47.7±2.3	52.6±2.5	44.8±4.6	52.2	51.5	43.8±6.4
(n-3)	46.3	43.3	43.5	49.7	45.9	50.6	43.3	50.2	50.0	42.4
(n-6)	2.0	1.9	2.1	1.7	1.8	2.0	1.5	2.0	1.5	1.5
(n-3) : (n-6)	23.0	22.9	20.5	29.3	25.8	24.7	28.3	25.1	33.3	29.1
Total (µg)	24.9±6.0		26.7±12	42.2±13		41.9±7.9		65.1	62.9	
n	44	15	5	8	25	7	16	1	1	7

Despite the large individual spread among the larvae, similarities in relative proportions of the various fatty acids in the larvae from the same stage from the three years are demonstrated (Fig. 4A). The fatty acid content changes gradually from stage 1a via 1d to stage 2a, i.e. the larvae with empty yolk sacs which have started to consume food have a distinctly different pattern of fatty acids than the newly hatched larvae with yolk sacs. The position of the fatty acids in the plot indicate how they change with the stages. The acids 18:4n3 and 20:5n3, positioned on the right side of the plot, at the same side as the stage 2a larvae, have the highest relative increase from stage 1a to 2a. But since the directions from the origin in the plot towards the two acids are not the same, they are poorly correlated among the individual larvae. On the other hand, the acids positioned to the left in the biplot, 20:1n9 in particular, are decreasing in amount from stage 1a to stage 2a (Fig. 4A).

When absolute amounts of fatty acids in the larvae from 1991 and 1993 are considered, differences among the three stages are also obvious (Fig. 4B). However, the transition between the stages is different from the case with relative amounts in Fig. 4A. The difference between stage 1a and 1d is along the second principal component, which only accounts for 10 % of the total variance among the samples. The further change from stage 1d to stage 2a is along PC 1, which takes care of 75 % of the total variation. This means that the difference between the stages 1a and 1d is much smaller, and also in different fatty acids, than the difference between the stages 1d and 2a, i.e. when the larvae have started to eat. The fatty acids are all positioned on the right side of the origin in the biplot, indicating that they have all increased in the stage 2a larvae compared with the earlier stages.

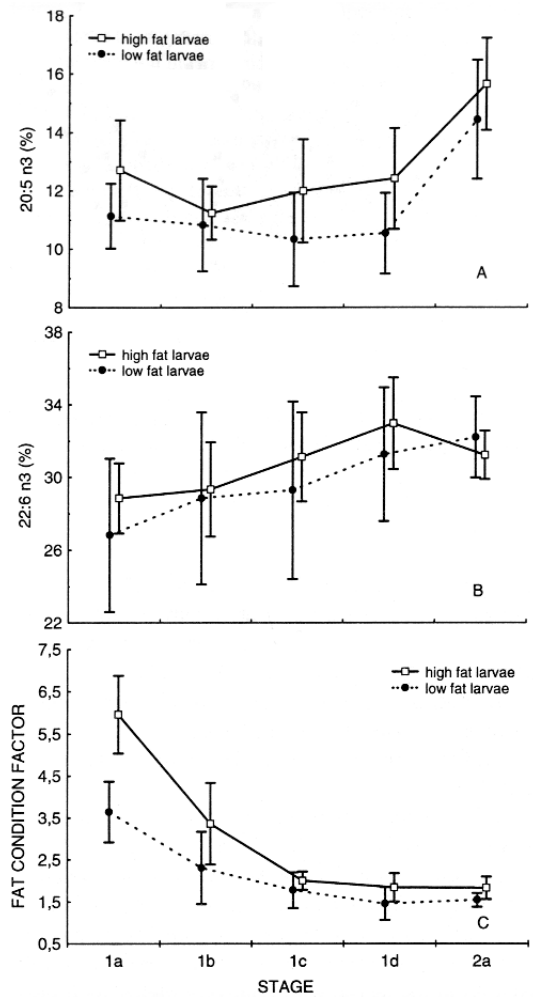

Fig. 3. Stage specific: A) proportion of 20:5n3 fatty acid, B) proportion of 22:6n3 fatty acid, and C) fat condition factor. Squares represent values for herring larvae with total fatty acid content above average within respective stages and years (1991 and 1993). Circles represent values for herring larvae with total fatty acid content below average in respective stages and years.

DISCUSSION

Several methods of measuring fatty acid composition have been proposed in connection with the development of nutritional indices in fish larvae. The triacyl glycerols (TAG) have been emphasized as a suitable indicator of nutritional state of fish larvae (Fraser & al. 1987; Olsen & al. 1991). The determination of TAG fatty acid composition is, however, rather elaborate (Fraser & al. 1989), and requires more effort than the analysis of total fatty acid compositions as employed in this study. The present method of methanolysis results in the loss of lipid class information, but on the other hand it permits the use of multivariate statistics on fatty acid data of individual larvae (Barnung & Grahl-Nielsen 1987; Hove & Grahl-Nielsen 1991). It is impossible to obtain a total picture of how the pattern of fatty acids changes with the development of the larvae by a univariate evaluation of a large table of numbers (Table 1). A biplot, resulting from a multivariate, principal component analysis, reveals a much more comprehensive view on the results (Fig. 4A,B).

The increase in total fatty acid content from 1d to 2a in our study indicates that the 2a larvae on average were in an active growth phase. There was a fair amount of individual variation in both the 1d and 2a stage, however. This could possibly to some extent be due to varying

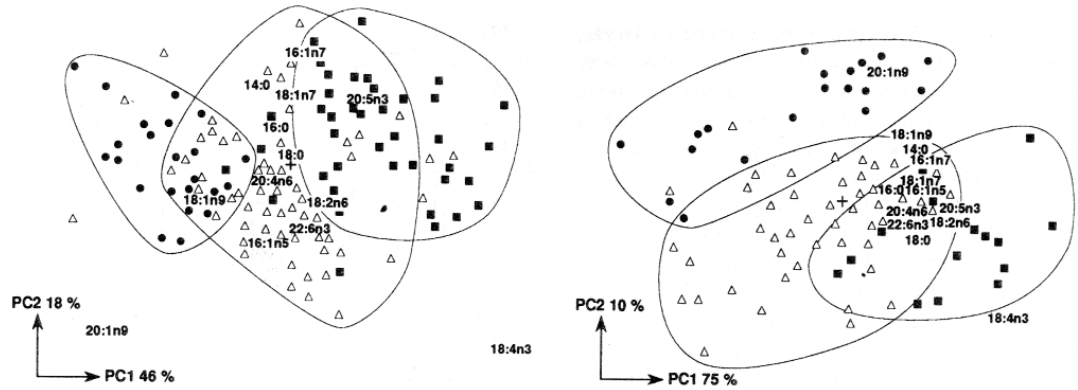

Fig. 4. PCA biplots of A) relative and B) absolute amounts of the various fatty acids in stages 1a (filled circles), 1d (triangles) and 2a (filled squares). Data for all available years are combined.

degree of stomach filling, but herring larvae are known to evacuate the gut content during capture, and no gut contents were found in the larval material investigated from 1993 (Johannessen & al. 1995). The relative proportion between the main constituents found in our herring larvae also shows the same overall pattern reported for herring larvae in other studies (e.g. Fraser & al. 1987).

The qualitative differences in fatty acid composition between relatively 'fatty' wild larvae and less 'fatty' wild larvae within respective stages are essential in relation to the overall objective of establishing a fatty acid based nutritional index. In this study we found higher proportions of long chained highly unsaturated fatty acids (HUFA) in the larvae with relatively higher total fatty acid content. The 18:4n3 and 20:5n3 (EPA) fatty acids have also been found in relatively higher proportions in well nourished larvae than in undernourished larvae grown in large enclosures (Fraser & al. 1987). Without intake of HUFA, the relative amounts of for example 22:6n3 (DHA) will decline (Navarro & al. 1993). This fatty acid has been shown to play an important role in the development of neural and visual tissue, and reductions in DHA and EPA in starving herring larvae have been accompanied by behavioural changes (Navarro & Sargent 1992). The qualitative differences in fatty acid composition found in this study may therefore well represent a survival advantage of the larvae with higher fat content.

The data also seem to indicate a qualitative difference between offspring prior to feeding. Yolk sac larvae with relatively high fat content also tended to have higher proportions of HUFA. This could be due to maternal effects, where eggs from certain females also have a more optimal fatty acid composition. Such differences have been suggested to occur in cod (Ulvund & Grahl-Nielsen 1988) and several other species (Rainuzzo & al. 1992). The conservation of high levels of some of these fatty acids in lipids during embryogenesis and early larval development is suggested to be a reflection of their importance (Tocher & al. 1985). Due to the common strong correlation between egg size and larval size (Miller & al. 1988), individuals from large eggs with more fat may maintain their qualitative advantage into the larval stage.

The change in fatty acid composition between yolk sac larvae and feeding larvae may also be due to dietary input either directly or indirectly. In some species the stomach contents of the larvae will influence the fatty acid profile if the whole larva is used in the analysis (Suthers & al. 1992). In the case of herring larvae used in this study, this is not very likely since no prey remains were found in other examined herring larvae from the study area as mentioned above. It can not, however, be ruled out that the increase in 18:4n3 and 20:5n3 fatty acids found in stage 2 larvae, can have a plankton origin as suggested by Gatten & al. (1983) and Fraser & al. (1989).

We observed differences in fatty acid composition between herring larvae of varying condition, but more information is needed on the underlying dynamics of lipid synthesis and metabolism before fatty acid composition can be used routinely as a nutritional index. Laboratory experiments with larvae grown under controlled conditions should be undertaken to clarify how the fatty acid composition changes as a function of starvation and dietary fatty acid input. This will enable us to make inferences about the past feeding history and the future survival potential of larvae sampled in the field.

ACKNOWLEDGEMENTS

This study was funded by the Norwegian Research Council (Programme MARE NOR), project No. I.501.024.

REFERENCES

Anderson, J.T. 1988. A review of size dependent survival during pre-recruit stages of fishes in relation to recruitment. – *Journal of Northwest Atlantic Fisheries Science* 8:55-66.

Barnung, T. & O. Grahl-Nielsen 1987. The fatty acid profile in cod (*Gadus morhua* L.) eggs and larvae. Developmental variations and responses to oil pollution. – *Sarsia* 72:415-417.

Clemmesen, C.M. 1989. RNA/DNA ratios of laboratory-reared and wild herring larvae determined with a highly sensitive fluorescence method. – *Journal of Fish Biology* 35:331-333.

Doyle, M.J. 1977. A morphological staging system for the larval development of the herring, *Clupea harengus* L. – *Journal of the Marine Biological Association UK* 57:859-867.

Fraser, A.J., J.C. Gamble & J.R. Sargent 1988. Changes in lipid content, lipid class composition and fatty acid composition of developing eggs and unfed larvae of cod (*Gadus morhua*). – *Marine Biology* 99:307-313.

Fraser, A.J., J.R. Sargent, J.C. Gamble & P. MacLachlan 1987. Lipid class and fatty acid composition as indicators of the nutritional condition of larval Atlantic herring. – *American Fisheries Society Symposia* 2:129-143.

Fraser, A.J., J.R. Sargent, J.C. Gamble & D.D. Seaton 1989. Formation and transfer of fatty acids in an enclosed marine food chain comprising phytoplankton, zooplankton and herring (*Clupea harengus* L.) larvae. – *Marine Chemistry* 27:1-18.

Gatten, R.R., J.R. Sargent & J.C. Gamble 1983. Diet-induced changes in fatty acid composition of herring larvae reared in enclosed ecosystems. – *Journal of the Marine Biological Association UK* 63:575-584.

Grahl-Nielsen, O. & T. Barnung 1985. Variations in the fatty acid profile of marine animals caused by environmental and developmental changes. – *Marine Environmental Research* 17:218-221.

Hove, H.T. & O. Grahl-Nielsen 1991. Fatty acid composition of start-feeding (*Salmo salar*) larvae. – *Aquaculture* 96:305-319.

Johannessen, A., G. Blom, A. Folkvord & H. Svendsen 1995. The effect of local wind on the distribution of Norwegian spring-spawning herring (*Clupea harengus* L.) larvae. – Pp. 365-384 in: Skjoldal, H.R., C.C.E. Hopkins, K.E. Erikstad, H.P. Leinaas (eds). *Ecology of Fjords and Coastal Waters. Proceedings of the Mare Nor Symposium on the Ecology of Fjords and Coastal.Waters, Tromsø, Norway, 5-9 December 1994*. Elsevier, Amsterdam.

Kraul, S., A. Nelson, K. Brittain, H. Ako & A. Ogasawara 1992. Evaluation of live feed for larval and postlarval Mahi mahi *Coryphaena hippurus*. – *Journal of the World Aquaculture Society* 23:299-306.

Meeren, T. van der 1993. Fatty acid composition of unfed cod larvae *Gadus morhua* L. and cod larvae feeding on natural plankton in large enclosures. – *Journal of the World Aquaculture Society* 24:167-185.

Miller, T.J., L.B. Crowder, J.A. Rice & E.A. Marschall 1988. Larval size and recruitment mechanisms in fishes: toward a conceptual framework. – *Canadian Journal of Fisheries and Aquatic Sciences* 45:1657-1670.

Navarro, J.C., R.S. Batty, M.V. Bell & J.R. Sargent 1993. Effects of two *Artemia* diets with different contents of polyunsaturated fatty acids on the lipid composition of larvae of Atlantic herring (*Clupea harengus*). – *Journal of Fish Biology* 43:503-515.

Navarro, J.C. & J.R. Sargent 1992. Behavioural differences in starving herring *Clupea harengus* L. larvae correlate with body levels of fatty acids. – *Journal of Fish Biology* 41:509-513.

Øiestad, V. 1983. *Growth and survival of herring larvae and fry (Clupea harengus L.) exposed to different feeding regimes in experimental ecosystems: outdoor basin and plastic bags*. Dr. philos. thesis, University of Bergen, Norway. 299 pp.

Olsen, R.E., R.J. Henderson & T. Pedersen 1991. The influence of dietary lipid classes on the fatty acid composition of small cod *Gadus morhua* L. juveniles reared in an enclosure in northern Norway. – *Journal of Experimental Marine Biology and Ecology* 148:59-76.

Rainuzzo, J.R., K.I. Reitan & Y. Olsen 1992. Comparative study on the fatty acid and lipid composition of four marine fish larvae. – *Comparative Biochemistry and Physiology B* 103:21-26.

Suthers, I.M., A. Fraser & K.T. Frank 1992. Comparison of lipid, otolith and morphometric condition indices of pelagic juvenile cod *Gadus morhua* from the Canadian Atlantic. – *Marine Ecology Progress Series* 84:31-40.

Theilacker, G.H. 1986. Starvation-induced mortality of young sea-caught jack mackerel, *Trachurus symmetricus*, determined with histological and morphological methods. – *Fishery Bulletin US* 86:1-17.

Tocher, D.R., J.R. Sargent, A.J. Fraser & J.C. Gamble 1985. Fatty acid composition of phospholipids and neutral lipids during embryonic and early larval development in Atlantic herring (*Clupea harengus*, L.). – *Lipids* 20:69-74.

Ulvund, K.A. & O. Grahl-Nielsen 1988. Fatty acid composition in eggs of Atlantic cod (*Gadus morhua*). – *Canadian Journal of Fisheries and Aquatic Sciences* 45:898-901.

Watanabe, T. 1993. Importance of docosahexanoic acid in marine larval fish. – *Journal of the World Aquaculture Society* 24:152-161.

Witt, U., G. Quantz & D. Kuhlmann 1984. Survival and growth of turbot larvae *Scophthalmus maximus* L. reared on different food organisms with special regard to long-chain polyunsaturated fatty acids. – *Aquaculture Engineering* 3:177-190.

Accepted 10 December 1995.