

# Atlantic Halibut, *Hippoglossus hippoglossus*

# 7

**Barbara Grisdale-Helland<sup>1</sup> and Ståle J. Helland<sup>1,2</sup>**

<sup>1</sup>AKVAFORSK (Institute of Aquaculture Research AS), N-6600

Sunnalsøra, Norway; <sup>2</sup>Department of Animal Science, Agricultural University of Norway, N-1432 Ås-NLH, Norway

## Introduction

---

In 1999, about 400 metric tons (t) of farmed Atlantic halibut with a value of 36 million Norwegian crowns (NOK) were processed in Norway, up from 275 t in 1998 and 113 t in 1997. Halibut larvae have a long yolk-sac stage requiring carefully controlled temperature, salinity, light and microbial conditions and a low disturbance level (Mangor-Jensen *et al.*, 1998). Feeding of larvae involves the use of live organisms, followed by gradual replacement with dry diets. Juveniles and grow-out fish are fed dry pellets in tanks on land or in sea pens. The fish are normally processed at a weight of about 5 kg after a production time of about 37–44 months, depending on the techniques used (Engelsen, 1998). Females may reach this weight 6 months before males (Engelsen, 1998). Currently, larvae are produced from brood stock originally captured in the wild and from first-generation domesticated fish.

## Nutrient Requirements

---

### *Protein and amino acids*

---

During the yolk-sac stage and the initial period of exogenous feeding, free and then later protein-derived amino acids are the major sources of energy for Atlantic halibut larvae (Rønnestad, 1993; Rønnestad and Naas, 1993; Finn *et al.*, 1995). The major amino acids in the yolk proteins are alanine and leucine, whereas aspartic acid and glutamic acid predominate in the proteins of the larval body and in juveniles (Finn *et al.*, 1995; Kim and Lall, 2000).

The optimal protein level in diets for Atlantic halibut appears to be dependent on the size of the fish. Hjertnes and Opstvedt (1989) reported a linear increase in the growth rate of halibut grown from 7 to about 180 g, in response to increasing the dietary protein level from 41% to 62% on a dry matter (DM) basis and

decreasing the carbohydrate level from 27% to 3%. Similarly, we found a linear response in the growth of halibut from 60 to 125 g when the dietary protein level was increased from 41% to 56% and the carbohydrate level was decreased from 24% to 7% (Helland and Grisdale-Helland, unpublished). In contrast, Hjertnes *et al.* (1993) found no effect when the dietary protein level was increased from 40% to 55%, concomitant with a decrease in dietary carbohydrate from 24% to 8%, on halibut grown from 34 to about 300 g. However, the authors reported a significant increase in growth when the dietary protein level was further increased to 60% and the carbohydrate level was reduced to 3%. Reducing the protein level of this diet from 60% to 55% and increasing dietary fat from 26% to 30% resulted in significantly lower growth. Halibut grown from about 150 g to 550 g benefited from an increase in the dietary protein level from 41% to 51% (dietary carbohydrate reduced from 27% to 15%), but higher dietary protein levels, obtained through reductions in either carbohydrate or fat, did not result in increased growth of this size of fish (Aksnes *et al.*, 1996; Helland and Grisdale-Helland, 1998). In the weight range of 0.6–1.5 kg, the growth of halibut was not improved by increasing the dietary protein level from 37% to 50% (DM basis) with a reduction in dietary fat content (Berge and Storebakken, 1991) or from 48% to 54% (replacing either dietary fat or carbohydrate) (Grisdale-Helland and Helland, 1998).

The digestibility by halibut of protein in dry diets based on low-temperature-produced fish-meal is in the range of 82–86% (Grisdale-Helland and Helland, 1998; Berge *et al.*, 1999) and that in moist diets has been found to be 75–88% (Berge and Storebakken, 1991; Berge *et al.*, 1991; Haugen, 1999). Protein digestibility, as measured in mink, has been reported to influence the amount of feed consumed by halibut to reach satiation (Aksnes and Mundheim, 1997).

Few alternative protein sources to fish-meal have been tested in halibut diets. The replacement of fish-meal with soybean-protein concentrate (SPC) (44% of total dietary protein) and a methionine supplement in the diet of 600–900 g halibut had no effect on nitrogen digestibility, but resulted in slightly lower feed efficiency, probably due to the fibre content in SPC (Berge *et al.*, 1999). The fish compensated for the lower overall nutritive value of the SPC diet by increasing feed intake and there was no effect on growth. Replacement of fish-meal with full-fat soybean meal (30% of dietary protein) had no effect on feed intake, growth or feed utilization (B. Grisdale-Helland, S.J. Helland, G. Baeverfjord and G.M. Berge, unpublished data). Wheat gluten is also an acceptable replacement for fish-meal in diets for Atlantic halibut, but may require supplementation of lysine when levels as high as 30% of diet are used (Helland *et al.*, 2000). A lysine requirement of 19–21 g kg<sup>-1</sup> weight gain (2.9–3.5% of dietary DM) for 60–125 g halibut fed a 60% protein diet is suggested from these results. This is in the upper range of what has been reported for other species (NRC, 1993).

Halibut in the 50–300 g weight range fed high-energy, fish-meal-based diets consume about 400 g protein and 4200 kcal gross energy (GE) kg<sup>-1</sup> gain (Table 7.1). Larger fish consume about the same amount of protein and 4700 kcal GE kg<sup>-1</sup> gain. The retention of protein in halibut fed high-energy diets containing 47–60% protein is generally high (Table 7.2). Isotope studies of

**Table 7.1.** Protein and energy consumed by Atlantic halibut per kg gain\*.

Initial weight (g)	Final weight (g)	Dietary protein content (g kg <sup>-1</sup> )	Dietary energy content (kcal kg <sup>-1</sup> )	Protein consumed <sup>†</sup>	Energy consumed <sup>‡</sup>	Reference
5	539	630	5115	536	4326	A
5	539	550	6095	436	4828	A
60	125	560	5688	383	3896	B
60	125	596	6071	359	3657	B
72	100	540	5497	501	5091	C <sup>§</sup>
140	260	511	5712	331	3705	D
170	315	484	5903	320	3920	E
630	864	472	6262	366	4852	F
750	860	509	5688	422	4708	G <sup>§</sup>
1020	1260	479	5832	410 (338)	4995 (4254)	H
2420	2700	535	5378	418	4183	I

\* Experiments in which feed intake was measured. If not specified, data were calculated from available information. When several diets were tested in the same experiment, that with the lowest protein level giving highest gain was chosen. When gross energy was not measured, it was calculated using the following factors (kcal kg<sup>-1</sup>): protein 5400, fat 9300, carbohydrate 4100.

<sup>†</sup> g kg<sup>-1</sup> gain (digestible protein in parentheses).

<sup>‡</sup> kcal gross energy kg<sup>-1</sup> gain (digestible energy in parentheses).

A, Aksnes *et al.*, 1996; B, S.J. Helland and B. Grisdale-Helland, unpublished; C, Nortvedt and Tuene, 1995; D, Helland and Grisdale-Helland, 1998; E, B. Grisdale-Helland, S.J. Helland, G. Baeverfjord and G.M. Berge, unpublished; F, Berge *et al.*, 1999; G, Haugen, 1999; H, Grisdale-Helland and Helland, 1998; I, Helland *et al.*, 1997.

<sup>§</sup> Data from four centrepont tanks.

protein synthesis rates also suggest that this species uses protein efficiently for growth (Fraser *et al.*, 1998).

## Energy

It is generally known that fish eat to meet their energy need and therefore, although bulk may restrain consumption, fish should be able to compensate for a lower energy-density diet with increased intake. Results from a study with 1 kg halibut fed diets differing in dietary levels of carbohydrate and protein show, however, that compensation for low digestible-energy (DE) content by increased intake does not always occur, even when bulk is apparently not a limiting factor (Grisdale-Helland and Helland, 1998).

The fasting metabolic rate for halibut has been estimated to be 1.4 (Jonassen *et al.*, 2000) to 2.7 g oxygen (O<sub>2</sub>) kg<sup>-0.78</sup> day<sup>-1</sup> (Davenport *et al.*, 1990) (calculated using the weight coefficient 0.78 determined for plaice (*Pleuronectes platessa*) and flounder (*Platichthys flesus*) by Fonds *et al.* (1992)). The routine metabolic

**Table 7.2.** Retention of consumed protein and energy in whole body of Atlantic halibut (%).

Initial weight (g)	Final weight (g)	Dietary protein content (g kg <sup>-1</sup> )	Dietary energy content (kcal kg <sup>-1</sup> )	Protein retained (%)	Energy retained (%)	Reference*
60	125	560	5688	43	47	B
60	125	596	6071	48	49	B
72	100	540	5497	33		C†
140	260	511	5712	46	58	D
170	315	484	5903	52	55	B
630	864	472	6262	47	57	F

\* See Table 7.1 for references.

† Data from four centrepont tanks.

rate has been estimated to be in the range 1.3–3.6 g O<sub>2</sub> kg<sup>-0.78</sup> day<sup>-1</sup> (Davenport *et al.*, 1990; Hallaråker *et al.*, 1995; Simensen, 1999; Jonassen *et al.*, 2000; B. Grisdale-Helland, S.J. Helland, G. Baevefjord and G.M. Berge, unpublished). The highest of these metabolic-rate estimates for both fed and fasted fish are from Davenport *et al.* (1990) and may be overestimates because the fish, which were collected in the wild, were noted to react poorly to handling.

### ***Lipids and fatty acids***

It is widely accepted that docosahexaenoic acid (DHA), 22:6n-3, and eicosapentaenoic acid (EPA), 20:5n-3, are essential fatty acids in diets for marine fish. Data from other marine fish species suggests that the requirement for n-3 highly unsaturated fatty acids (HUFA) is 1–2% of the diet for larvae and 0.5–1% for larger fish (reviewed by Sargent *et al.*, 1995; Harel and Place, 1998). The lack of  $\Delta$ -5-desaturase activity in marine fish, preventing conversion of linoleic acid, 18:2n-6, to arachidonic acid (AA), 20:4n-6, which is essential for the production of eicosanoids, implies the essentiality of providing AA in larvae diets (Sargent *et al.*, 1997, 1999). McEvoy *et al.* (1998) noted, though, that normal pigmentation in halibut larvae was negatively correlated with the AA concentration in the brain. The best pigmentation occurred in the larvae fed *Artemia* enriched with oil containing EPA : AA ratios above 4 : 1 and no correlation between the DHA : EPA ratio in the oil and pigmentation was found (McEvoy *et al.*, 1998). In contrast, Shields *et al.* (1999a) suggested that the poorer pigmentation of halibut larvae fed *Artemia* compared with those fed copepods (*Eurytemora velox*) was related to a relative deficiency of DHA in the *Artemia* and was not related to the levels or ratio of EPA and AA. The copepods also contained a higher ratio of phospholipids to triacylglycerols (TG) than the *Artemia*. As reported by Shields *et al.* (1999a), phospholipids may be more easily digested than TG and may also be required in the diet of fish larvae because of low *de novo* synthesis.

In early trials with grow-out Atlantic halibut, the digestibility of fat was found to be in the 78–94% range (Berge and Storebakken, 1991; Berge *et al.*, 1991), whereas more recent studies show consistently higher digestibility (95–99%) (Grisdale-Helland and Helland, 1998; Berge *et al.*, 1999; Haugen, 1999).

## **Carbohydrates**

---

Gawlicka *et al.* (2000) noted that the amylase activity level was low in halibut larvae before metamorphosis compared with that found for other marine fish larvae. The amylase activity in metamorphic larvae was considerably higher than that found in earlier stages (Gawlicka *et al.*, 2000).

The digestibility of carbohydrate in 1 kg Atlantic halibut is reduced when the dietary level is increased (Grisdale-Helland and Helland, 1998; Grisdale-Helland and Helland, unpublished; Helland and Grisdale-Helland, unpublished results; Fig. 7.1). Brudeseth (1996) found that, of the disaccharidases, only maltase activity in halibut was affected by the dietary carbohydrate level. Comparing Atlantic halibut and Atlantic salmon, Brudeseth (1996) found that these species exhibited similarities in both the total capacity of the disaccharidases and the enzymatic capacity of similar gut segments.

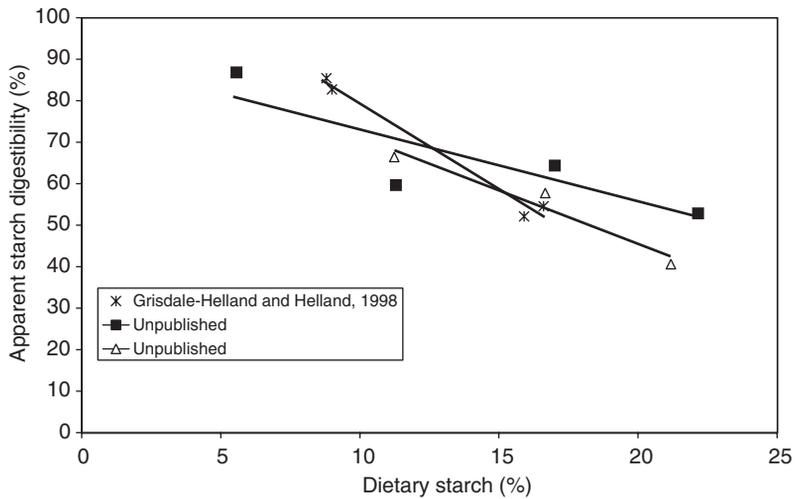
Metabolic adaptation to increasing dietary carbohydrate has been shown. Garcia-Riera and Hemre (1996) reported that halibut fed diets containing 5% or 8% carbohydrate showed rapid removal of glucose from the blood and deposition in the liver compared with fish fed a diet containing 2% carbohydrate. The latter fish had the highest plasma glucose levels 16 h after a peritoneal injection of glucose, and exhibited greater retention of glucose in muscle, heart, gills, kidney and the gastrointestinal tract.

## **Vitamins**

---

Before the development of dry diets, it was common to feed large halibut whole or parts of fish, such as herring, or moist diets. This is still being practised with brood stock. A preliminary study by Goff and Lall (1989) indicated that the liver content of vitamins A, E and C and total flavin could be increased in Atlantic halibut through vitamin supplementation of these types of diets.

The content of vitamin A in halibut larvae during the initial first-feeding period appear to be affected by the type of live food used. Rønnestad *et al.* (1998) observed that larvae fed only *Artemia* enriched with DHA Selco (INVE, Belgium) contained lower levels of all-trans retinal and all-trans retinol and had a higher incidence of malpigmentation than larvae initially fed copepods (mainly *Temora longicornis*). High contents of lutein and astaxanthin were found in *Temora*, whereas cryptoxanthin/cantaxanthin and an unknown retinoid compound were found in *Artemia*.



**Fig. 7.1.** Relationship between dietary starch concentration and apparent starch digestibility in Atlantic halibut fed diets containing extruded wheat.

Halibut juveniles do not exhibit L-gulonolactone oxidase activity, which is involved in the synthesis of ascorbic acid (Mæland and Waagbø, 1998). Halibut larvae (10 mg) fed zooplankton (mainly *T. longicornis* and containing 750 mg ascorbic acid  $\text{kg}^{-1}$  dry weight (DW)) were observed to maintain stable tissue concentrations of ascorbic acid, suggesting that the requirement for this vitamin is met at this level (Rønnestad *et al.*, 1999a). Young halibut (500–1000 mg) weaned on to formulated diets showed increasing whole-body ascorbic acid levels in response to increasing dietary ascorbate polyphosphate (300–3000 mg ascorbic acid equivalents  $\text{kg}^{-1}$  diet), although retention of the vitamin was estimated to be very low (Mæland *et al.*, 1999). The dietary level of ascorbic acid has not been shown to affect growth or mortality of young halibut (Mæland and Waagbø, 1998; Mæland *et al.*, 1999).

Vitamin B<sub>6</sub> in halibut larvae feeding on zooplankton containing 4–5 mg B<sub>6</sub>  $\text{kg}^{-1}$  DW stabilized during development at about 5 mg  $\text{kg}^{-1}$  DW (Rønnestad *et al.*, 1997). The decline in the vitamin B<sub>6</sub> content in whole larvae per g growth, from the yolk-sac stage until first feeding, was 3.5 mg, or about 17 nmol (Rønnestad *et al.*, 1997), similar to the requirement exhibited by many different species of animals (15 nmol  $\text{g}^{-1}$  weight gain) (Coburn, 1994).

## Practical Diets

In semi-intensive systems, zooplankton is still necessary for the production of acceptable halibut juveniles (Berg, 1997). As mentioned above, feeding with marine copepods generally improves pigmentation (Shields *et al.*, 1999a) and also influences the vitamin A status, eye rod/cone ratio and eye migration of halibut larvae (Rønnestad *et al.*, 1998; Shields *et al.*, 1999a,b). However, because

of the limited supply of copepods, studies are continuing on enrichment of *Artemia* for use as halibut larval diets (Olsen *et al.*, 1999; Shields *et al.*, 1999b).

Inclusion of free amino acids in first-feeding diets for halibut is difficult because of leaching. Inclusion in liposomes may be possible, but only small particles can be made. Alternatively, either liposomes may be included in microdiets fed to larvae or free amino acids may be used to enrich *Artemia* (Rønnestad *et al.*, 1999b).

Some commercial diets are produced specifically for halibut larvae and grow-out fish. The protein content in these diets (DM basis) range from 62–68% for particles less than 1 mm to 48–67% for 8–20 mm pellets. Concomitant with the decrease in protein content with increasing pellet size is an increase in fat from 12–20% to 16–27%.

## Feeding Practices

---

The production technology used in halibut farming was initially based on experience from turbot and cod, whereas the nutrition and feeding of juvenile and grow-out halibut have had their base mostly in salmon production. In Norway, most of the larvae are produced semi-intensively and fed zooplankton and *Artemia* nauplii, whereas the rest are produced in intensive systems (Olsen *et al.*, 1999), as is the case in Scotland (Shields *et al.*, 1999b) and Iceland. After first feeding, the majority of halibut in Norway are raised on land in tanks with circular water flow (with or without shelves) (Engelsen, 1998). Octagonal concrete tanks and raceways are also in use. Salmon cages, modified to give a stiff bottom, also have good potential for use with halibut, but although they may be cheaper to establish than a land operation, they require greater production and technical control (Engelsen, 1998). Automatic feeders, supplemented with hand-feeding, are normally used in halibut production.

Little research has been done on optimal feeding practices for halibut. Feed intake of halibut is influenced by pellet size and/or mixture of pellet sizes (Helland *et al.*, 1997). In a pilot trial, growth of 390–600 g halibut was found to be significantly better when the fish were fed in excess daily or every second day compared with every third day. Feed intake measurements showed no effect of feeding interval on feed efficiency and it was concluded that this size of halibut could be fed every second day (Helland *et al.*, 1993). Our results from a trial with 23 g halibut fed from one to 72 times per day revealed only small differences in growth and feed efficiency (Grisdale-Helland and Helland, unpublished).

## References

---

- Aksnes, A. and Mundheim, H. (1997) The impact of raw material freshness and processing temperature for fish meal on growth, feed efficiency and chemical composition of Atlantic halibut (*Hippoglossus hippoglossus*). *Aquaculture* 149, 87–106.

- Aksnes, A., Hjertnes, T. and Opstvedt, J. (1996) Effect of dietary protein level on growth and carcass composition in Atlantic halibut (*Hippoglossus hippoglossus* L.). *Aquaculture* 145, 225–233.
- Berg, L. (1997) Commercial feasibility of semi-intensive larviculture of Atlantic halibut (*Hippoglossus hippoglossus*). *Aquaculture* 155, 333–340.
- Berge, G.M. and Storebakken, T. (1991) Effect of dietary fat level on weight gain, digestibility, and fillet composition of Atlantic halibut. *Aquaculture* 99, 331–338.
- Berge, G.M., Krogdahl, Å., Strømsnes, Ø., Grønseth, F.A., Myhre, P. and Austreng, E. (1991) Digestibility determination in Atlantic halibut (*Hippoglossus hippoglossus*). *Fiskeridirektoratets Skrifter, Serie Ernæring* 4, 117–125.
- Berge, G.M., Grisdale-Helland, B. and Helland, S.J. (1999) Soy protein concentrate in diets for Atlantic halibut (*Hippoglossus hippoglossus*). *Aquaculture* 178, 139–148.
- Brudeseth, L.K. (1996) Hydrolyse av karbohydrater i tarmmucosa hos atlantisk laks (*Salmo salar*), kveite (*Hippoglossus hippoglossus*) og mink (*Mustela vison*). Effekt av variasjon i førets innhold av stivelse og antinæringsstoff fra soya. (Hydrolysis of carbohydrates in the intestinal mucosa of Atlantic salmon (*Salmo salar*), halibut (*Hippoglossus hippoglossus*) and mink (*Mustela vison*). Effect of variation in dietary content of starch and antinutrients from soya.) Cand. Scient. thesis, University of Oslo, Oslo, Norway (in Norwegian).
- Coburn, S.P. (1994) A critical review of minimal vitamin B<sub>6</sub> requirements for growth in various species with a proposed method of calculation. *Vitamins and Hormones – Advances in Research and Applications* 48, 259–300.
- Davenport, J., Kjorsvik, E. and Haug, T. (1990) Appetite, gut transit, oxygen uptake and nitrogen excretion in captive Atlantic halibut, *Hippoglossus hippoglossus* L. and lemon sole, *Microstomus kitt* (Walbaum). *Aquaculture* 90, 267–277.
- Engelsen, R. (1998) *Matfiskoppdrett av kveite. En økonomisk betraktning.* (Production of grow-out halibut. An economic consideration.) Report to the Norwegian Research Council, Rolf Engelsen AS, Bergen, 74 pp. (in Norwegian).
- Finn, R.N., Rønnestad, I. and Fyhn, H.J. (1995) Respiration, nitrogen and energy metabolism of developing yolk-sac larvae of Atlantic halibut (*Hippoglossus hippoglossus*). *Comparative Biochemistry and Physiology* 111A, 647–671.
- Fonds, M., Cronie, R., Vethaak, A.D. and van der Puyl, P. (1992) Metabolism, food consumption and growth of plaice (*Pleuronectes platessa*) and flounder (*Platichthys flesus*) in relation to fish size and temperature. *Netherlands Journal of Sea Research* 29, 127–143.
- Fraser, K.P.P., Lyndon, A.R. and Houlihan, D.F. (1998) Protein synthesis and growth in juvenile Atlantic halibut, *Hippoglossus hippoglossus* (L.): application of <sup>15</sup>N stable isotope tracer. *Aquaculture Research* 29, 289–298.
- Garcia-Riera, M.P. and Hemre, G.-I. (1996) Organ responses to <sup>14</sup>C-glucose injection in Atlantic halibut, *Hippoglossus hippoglossus* (L.), acclimated to diet of varying carbohydrate content. *Aquaculture Research* 27, 565–571.
- Gawlicka, A., Parent, B., Horn, M.H., Ross, N., Opstad, I. and Torrissen, O.J. (2000) Activity of digestive enzymes in yolk-sac larvae of Atlantic halibut (*Hippoglossus hippoglossus*): indication of readiness for first feeding. *Aquaculture* 184, 303–314.
- Goff, G.P. and Lall, S.P. (1989) An initial examination of the nutrition and growth of Atlantic halibut (*Hippoglossus hippoglossus*) fed whole herring with a vitamin supplement. *Bulletin of the Aquaculture Association of Canada* 89(3), 56–58.
- Grisdale-Helland, B. and Helland, S.J. (1998) Macronutrient utilization by Atlantic halibut (*Hippoglossus hippoglossus*): diet digestibility and growth of 1 kg fish. *Aquaculture* 166, 57–65.

- Hallaråker, H., Folkvord, A. and Stefansson, S.O. (1995) Growth of juvenile halibut (*Hippoglossus hippoglossus*) related to temperature, day length and feeding regime. *Netherlands Journal of Sea Research* 34, 139–147.
- Harel, M. and Place, A.R. (1998) The nutritional quality of live feeds for larval fish. *Bulletin of the Aquaculture Association of Canada* 98(4), 6–11.
- Haugen, T. (1999) Growth, feed conversion efficiency and digestion in Atlantic halibut: shorttime effects of different levels of temperature and fat/protein ratio in feed. Cand. Scient. thesis, University of Bergen, Bergen, Norway.
- Helland, S.J. and Grisdale-Helland, B. (1998) Growth, feed utilization and body composition of juvenile Atlantic halibut (*Hippoglossus hippoglossus*) fed diets differing in the ratio between the macronutrients. *Aquaculture* 166, 49–56.
- Helland, S.J., Grisdale-Helland, B. and Holm, J. (1993) Feeding regimes and compensatory growth of halibut. In: *6th International Symposium on Fish Nutrition and Feeding*. Hobart, Australia, 4–7 October, 1993. Abstract.
- Helland, S.J., Grisdale-Helland, B. and Berge, G.M. (1997) Feed intake and growth of Atlantic halibut (*Hippoglossus hippoglossus*) fed combinations of pellet sizes. *Aquaculture* 156, 1–8.
- Helland, S.J., Grisdale-Helland, B., Berge, G.M. and Bekkevold, K.R. (2000) Replacement of fish meal with wheat gluten in diets for Atlantic halibut: implications for lysine supplementation. In: *Ninth International Symposium on Nutrition and Feeding in Fish*. Miyazaki, Japan, 21–25 May, 2000, p. 68.
- Hjertnes, T. and Opstvedt, J. (1989) Effects of dietary protein levels on growth in juvenile halibut (*Hippoglossus hippoglossus* L.). In: *Proceedings of the Third International Symposium on Feeding and Nutrition in Fish*. 28 Aug–1 Sept, 1989, Toba, Japan, pp. 189–193.
- Hjertnes, T., Gulbrandsen, K.E., Johnsen, F. and Opstvedt, J. (1993) Effect of dietary protein, carbohydrate and fat levels in dry feed for juvenile halibut (*Hippoglossus hippoglossus* L.). In: Kaushik, S.J. and Luquet, P. (eds) *Fish Nutrition in Practice*. Les Colloques, no. 61, INRA, Paris, pp. 493–496.
- Jonassen, T.M., Imsland, A.K., Kadowaki, S. and Stefansson, S.O. (2000) Interaction of temperature and photoperiod on growth of Atlantic halibut *Hippoglossus hippoglossus* L. *Aquaculture Research* 31, 219–227.
- Kim, J.-D. and Lall, S.P. (2000) Amino acid composition of whole body tissue of Atlantic halibut (*Hippoglossus hippoglossus*), yellowtail flounder (*Pleuronectes ferruginea*) and Japanese flounder (*Paralichthys olivaceus*). *Aquaculture* 187, 367–373.
- McEvoy, L.A., Estevez, A., Bell, J.G., Shields, R.J., Gara, B. and Sargent, J.R. (1998) Influence of dietary levels of eicosapentaenoic and arachidonic acids on the pigmentation success of turbot (*Scophthalmus maximus* L.) and halibut (*Hippoglossus hippoglossus* L.). *Bulletin of the Aquaculture Association of Canada* 98(4), 17–20.
- Mæland, A. and Waagbø, R. (1998) Examination of the qualitative ability of some cold water marine teleosts to synthesise ascorbic acid. *Comparative Biochemistry and Physiology Part A* 121, 249–255.
- Mæland, A., Rosenlund, G., Stoss, J. and Waagbø, R. (1999) Weaning of Atlantic halibut *Hippoglossus hippoglossus* L. using formulated diets with various levels of ascorbic acid. *Aquaculture Nutrition* 5, 211–219.
- Mangor-Jensen, A., Harboe, T., Shields, R.J., Gara, B. and Naas, K.E. (1998) Atlantic halibut, *Hippoglossus hippoglossus* L., larvae cultivation literature, including a bibliography. *Aquaculture Research* 29, 857–886.
- Nortvedt, R. and Tuene, S. (1995) Multivariate evaluation of feed for Atlantic halibut. *Chemometrics and Intelligent Laboratory Systems* 29, 271–282.

- NRC (National Research Council) (1993) *Nutrient Requirements of Fish*. National Academy Press, Washington, DC, 114 pp.
- Olsen, Y., Evjemo, J.O. and Olsen, A. (1999) Status of the cultivation technology for production of Atlantic halibut (*Hippoglossus hippoglossus*) juveniles in Norway/Europe. *Aquaculture* 176, 3–13.
- Rønnestad, I. (1993) No efflux of free amino acids from yolk-sac larvae of Atlantic halibut (*Hippoglossus hippoglossus* L.). *Journal of Experimental Marine Biology and Ecology* 167, 39–45.
- Rønnestad, I. and Naas, K.E. (1993) Routine metabolism in Atlantic halibut at first feeding – a first step towards an energetic model. In: Walther, B.T. and Fyhn, H.J. (eds) *Physiology and Biochemistry of Marine Fish Larval Development*. University of Bergen, Bergen, Norway, pp. 279–284.
- Rønnestad, I., Lie, Ø. and Waagbø, R. (1997) Vitamin B<sub>6</sub> in Atlantic halibut, *Hippoglossus hippoglossus* – endogenous utilization and retention in larvae fed natural zooplankton. *Aquaculture* 157, 337–345.
- Rønnestad, I., Helland, S. and Lie, Ø. (1998) Feeding *Artemia* to larvae of Atlantic halibut (*Hippoglossus hippoglossus* L.) results in lower larval vitamin A content compared with feeding copepods. *Aquaculture* 165, 159–164.
- Rønnestad, I., Hamre, K., Lie, Ø. and Waagbø, R. (1999a) Ascorbic acid and  $\alpha$ -tocopherol levels before and after exogenous feeding. *Journal of Fish Biology* 55, 720–731.
- Rønnestad, I., Thorsen, A. and Finn, R.N. (1999b) Fish larval nutrition: a review of recent advances in the roles of amino acids. *Aquaculture* 177, 201–216.
- Sargent, J.R., Bell, J.G., Bell, M.V., Henderson, R.J. and Tocher, D.R. (1995) Requirement criteria for essential fatty acids. *Journal of Applied Ichthyology* 11, 183–198.
- Sargent, J.R., McEvoy, L.A. and Bell, J.G. (1997) Requirements, presentation and sources of polyunsaturated fatty acids in marine fish larval feeds. *Aquaculture* 155, 117–127.
- Sargent, J.R., McEvoy, L., Estevez, A., Bell, G., Bell, M., Henderson, J. and Tocher, D. (1999) Lipid nutrition of marine fish during early development: current status and future directions. *Aquaculture* 179, 217–229.
- Shields, R.J., Bell, J.G., Luizi, F.S., Gara, B., Bromage, N.R. and Sargent, J.R. (1999a) Natural copepods are superior to enriched *Artemia* nauplii as feed for halibut larvae (*Hippoglossus hippoglossus*) in terms of survival, pigmentation and retinal morphology: relation to dietary essential fatty acids. *Journal of Nutrition* 129, 1186–1194.
- Shields, R.J., Gara, B. and Gillespie, M.J.S. (1999b) A UK perspective on intensive hatchery rearing methods for Atlantic halibut (*Hippoglossus hippoglossus*). *Aquaculture* 176, 15–25.
- Simensen, L.M. (1999) Growth, activity and oxygen consumption in juvenile halibut (*Hippoglossus hippoglossus* L.) reared under four different photoperiods. Cand. Scient. thesis, University of Bergen, Bergen, Norway.