ICES CM 1999/U: 05 – Theme session U:

"M-74 SYNDROME AND SIMILAR REPRODUCTIVE DISTURBANCES IN MARINE ANIMALS"

Some possible explanations to Baltic cod (*Gadus morhua* L.) reproduction disturbances with special emphasis on lipids - an overview

Jana Pickova¹, Per-Olov Larsson² and Anders Kiessling³

¹ Department of Food Science, SLU, P.O. Box 7051, S-750 07 Uppsala, Sweden,.

² Institute of Marine Research. P.O.Box 4, S-453 21 Lysekil, Sweden;

³ Matre Aquaculture Station, Institute of Marine Research, N-5984 Matredal, Norway.

Abstract

2

Disturbances in Baltic cod reproduction, additional to the basic problems related to low salinity and oxygen content, are discussed. Abnormal cod embryos appeared first in 1979 and have been observed since. Still recruitment has been relatively well correlated to the "reproduction volume" (sufficient salinity and oxygen content) until year-class 1991. Since that year, year-class strengths have been lower than expected considering available reproduction volume. Many experimental investigations on Baltic cod reproduction have shown a lower hatching success compared to other cod stocks. Simultaneously with cod, other fish species in the Baltic Sea have experienced reproduction problems. Several hypotheses regarding the causes of the reproductive disturbances are reviewed, including a mis-match of hatching and environmental factors, including food availability and predator abundances. Also the impact of xenobiotic substances on peroxidation of unsaturated lipids (polyunsaturated fatty acids and cholesterol) as well as reactive intermediates, as DNA adducts, and their effects on the quality of spawning products are discussed. Also other fatty acid xenobiotic interactions are highlighted. Insufficient hormone level for induction of maturation and spawning, causing a delay in spawning activities, is also suggested to be caused by anthropogenic substances. We conclude that the causes of reproductive problems in Baltic Sea cod include factors other than overfishing and salinity - oxygen interactions.

Key words: Baltic Sea, egg quality, fatty acids, Gadus morhua, larval quality, lipid oxidation, xenobiotics

Fishery and other mortalities

Several investigations showing a decreased reproductive success in the Baltic cod population (Grauman and Sukhorukova 1982, Westernhagen et al. 1988, reviewed by Norrgren et al. 1998) have been performed during the 1980 - 90s. A strong decline in the population size from the mid 1980s until today is partly caused by poor management resulting in over-fishing. The peak yearly catch was approximately 400,000 tonnes in 1984. The variation of population size is large, lowest yearly catch being about 45,000 tonnes in the latest years (1993, Figure 1), partly due to the impact of low salinity and often restrictingly low oxygen content. Baltic cod is an important resource giving the most important fishery in many Baltic countries. The Baltic cod population consists of two stocks, the eastern Baltic stock which is the larger both in size and space and a western stock (Brander 1994). A reproduction volume, defined as the water volume having sufficient levels of salinity (>ca 11.5 psu) and oxygen (>ca 2 ml/l) (Larsson 1994), determines to a high degree the year-class strength and subsequent recruitment of Baltic cod. Since 1991 the relation between reproduction volume and corresponding year-class strength has been changed, with generally smaller year-classes than expected (Larsson 1994; reviewed by Norrgren et al. 1998).

Simultaneously with the low recruitment since 1991 a delay (approximately two months) of the peak of and spreading in time of the spawning was observed (Wieland 1995). Larsson (1996) discussed if this shift in timing could have led to an increased mortality by deteriorating environment e.g. lower oxygen content and higher temperatures later in the summer and fall, and/or increased predation, e.g. high abundance of medusae during late summer, and/or low food availability during the cod larvae first feeding. He found no conclusive evidence for effects on cod larvae mortality by any of these factors. Also the quality of the food available has to be considered as stressed by Bell and Sargent (1996) based on lipid and fatty acid composition of plankton during different periods. They suggest the content of essential fatty acid in different plankton species to be of importance.

It has been suggested (Nissling and Vallin 1996) that a low proportion of large females in the present Baltic cod population is detrimental for the reproductive success. This should be due to larger cod females having a higher fecundity than first time spawners and at least in some species larger eggs/larvae (generally from larger females) have a higher survival expectancy (e.g. Miller et al. 1988; see however below). On the other hand the proportion of large, older females is mainly determined by relative size of year-classes. In Figure 2 the relation between proportion of older females (5+) in the spawning stock and the subsequent recruitment for the yearclasses 1979 – 1996 is shown. The regression of recruitment on proportion old females is very poor ($r^2 = 0.02$) for the period chosen, which at least shows that there are other important factors involved in the recruitment process.

Possible explanations for the spawning time shift could be an alteration in endocrine hormone levels caused by unknown factors, as for example inhibition of prostaglandins

and/or steroid hormones by hormone-like substances. Also impact on spawning behaviour by hormone-like substances and pollutants are suggested (reviewed by Jones and Reynolds 1997).

Simultaneously with cod, other fish species in the Baltic Sea have experienced reproduction problems. For example the M74 syndrome in Baltic salmon (*Salmo salar*) and totally failing reproduction of pike (*Esox lucius*) and perch (*Perca fluviatilis*) in some coastal areas as well as sterile burbot (*Lota lota*) in areas which have a high load of anthropogenic substances have been reported in the Baltic Sea (reviewed by Norrgren et al. 1998).

Egg quality

During 1992-1997 several attempts were done to culture Baltic cod for experimental and research purposes in Denmark, Germany and Sweden (Buchman et al. 1993, Nissling and Westin 1991, Pickova and Larsson 1992). Several studies indicated that Baltic cod eggs have a higher frequency of irregular early blastomere cleavages followed by lower hatching rates (Kjörsvik 1994, Kjörsvik et al. 1999, Pickova et al. 1997). Åkerman et al. (1998) observed that the Baltic cod eggs had dramatically lower hatching rates than the eggs from the Barent Sea. Investigations on protist infections in Baltic cod eggs (Pedersen et al. 1994, Kjörsvik et al. 1999) did not show any dramatic effects on egg and larval survival.

The suggestion above by Nissling and Vallin (1996) that relatively fewer large, old cod females in later years result in overall higher mortality of larvae, is based on the assumption that larger larvae from larger eggs have a higher survival. This conclusion can however be discussed. In a study on haddock (*Melanogrammmus aeglefinus*) Trippel et al. (1999) showed that total dry matter and total lipid composition did not affect egg quality. McEvoy et al. (1993) showed in turbot (*Scophthalmus maximus*) that there was a decline in size in late season eggs but the survival of larvae was not affected. These studies are in contrary to the suggestion of Nissling and Vallin. The conclusions in their study are based on the length of larval survival prior to first feeding. This method can be discussed since no investigation on point of no return was performed and does therefore not evaluate survival during the important stage until feeding has succeeded.

The large importance of fatty acid composition for successful early development has been shown in many fish species and was reviewed by Sargent et al. (1995) and Wiegand (1996). Pickova et al. (1997) investigated fatty acid composition of eggs from different cod stocks. A significant difference in content of arachidonic acid (AA, 20:4n-6) in Baltic cod eggs was correlated to a lower hatching rate and higher degree of blastomere cleavage asymmetries. In addition, a higher content of docosahexaenoic acid (DHA, 22:6n-3) was found in those eggs. Since DHA is the most unsaturated fatty acid, its susceptibility to peroxidation is the highest (Cosgrove 1987). Similar results in fatty acid composition affecting survival were obtained in a study on Baltic salmon (Pickova et al. 1998). Impact of toxic substances on reproduction of fish and larval survival by hormone-like substances and direct toxicity is well recognised (reviewed by Kime 1995). The load of known and unknown anthropogenic substances has been frequently discussed in connection with Baltic cod reproduction. A study by Petersen et al. (1997) indicates that there is a correlation in ovary burdens of lipophilic xenobiotics with i.a. cod cgg and larval survival. This might also be the most plausible explanation for the increase in DNA adducts observed in Baltic cod at early life stages as suggested by Ericson et al. (1996).

0

Impact of xenobiotics on lipid quality

We suggest that lipids have to be considered to a higher degree as they are the fraction which is not only carrier of the halogenated substances but as a fraction which will get seriously affected by those. Chlorinated fatty acids have been detected in Baltic fish species (Wesén et al. 1995). Some shorter chlorinated fatty acids, probably as results of βoxidation seem to be stable and therefore it was concluded in Wesén et al. (1995) that they are able to disturb the reproductive processes. Despite an overall decrease in PCB load it was shown that coplanar PCBs have increased in salmon tissue (reviewed by Norrgren et al. 1998). The polyunsaturated fatty acids eicosanoid fatty acid and arachidonic fatty acid are precursors of eicosanoids which are the base for prostaglandins and other biologically important molecules (Sargent et al 1995). Mercure and Van Der Kraak (1995) have shown that different fatty acid content influence steroid hormone production in teleosts. In a study on arachidonic acid Mercure and Van Der Kraak (1996) concluded that it stimulated testosterone production and this action was mediated by prostaglandin (PGE-2). Cholesterol is a base of all steroid hormone synthesis in fish as well as in mammals (Teshima 1990). Peroxidation of unsaturated fatty acids and of cholesterol results in toxic products which are harmful to the organisms (Pettersson and Lignell 1996, Pickova 1998). In addition, the highly polyunsaturated fatty acids are the most susceptible to oxidative reactions (Cosgrove et al. 1987) and therefore the molecules which usually initiate the free radical induced chain reactions. Therefore to understand the role of lipid based hormonal complex and lipoproteins in the reproduction of fish in relation to xenobiotics has significance not only for fish but also for the lower animals in the Baltic Sea food chain. Further, it has been shown that polychlorinated biphenyls (PCBs) are able to cause changes in lipid metabolism by changing fatty acid composition in livers of different animals (Borlakoglu et al 1990, Kamei et al. 1996, Matsusue et al 1997). In experiments groups treated by injection of PCB showed an increased linolenic acid (18:3n-3) proportion in all glycerophosholipids of liver. On the other hand the content of arachidonic acid decreased significantly. Changes in the activity of desaturase isozymes have been postulated to explain this unusual lipid metabolism and also its contribution to toxicity (Matsusue et al. 1997). In general a decrease in antioxidant levels is observed in fish exposed to PCBs, dioxins and other halogenated substances (Otto and Moon. 1996, Winston and Di Giulio 1991). In a preliminary study on Arctic charr (Salvelinus alpinus) fed with diet added the commercial PCB mixture Aroclor 1254, we found a different fatty acid composition of the liver when compared to the control group fish (own unpublished results).

Conclusions

2

Many different factors have been investigated in order to explain the low reproductive success in Baltic cod. No single factor responsible for the decline in reproduction has so far been identified. Many studies indicate that factors other than exploitation, salinity and oxygen are influencing reproductive success. The free radical induced peroxidation, combined with a low antioxidative defence might well be one of the underlying factors. Other effects of lipid - halogenated xenobiotic interaction might also be of importance. We suggest that maturation and spawning of cod in the Baltic Sea are also influenced by the load of anthropogenic substances. It can be concluded that human activities resulting in environmental pollution are involved in the reproductive disorders in Baltic fish species.

References

- Åkerman, G. and Balk, L. 1998. Descriptive studies of mortality and morphological disorders in early life stages of cod and salmon originating from the Baltic Sea. Am. Fish. Soc. Symp. 21: 41-46.
- Bell, M. V. and Sargent, J. R. 1996. Lipid nutrition and fish recruitment. Mar. Ecol. Prog. Ser. 134: 315-316.
- Borlakoglu, J.T., Edwards-Webb, J.D. and Dils, R.R. 1990. Polychlorinated biphenyls increase fatty acid desaturation in the proliferating endoplasmatic reticulum of pigeon and rat livers. Eur. J. Biochem. 188: 327-332.
- Brander, K., (Ed.) 1994. Spawning and life history information for North Atlantic cod stocks. ICES Coop. Res. Rep. No. 205.
- Buchmann, K, Larsen, J. L. and Dalsgaard, I. 1993. Discases and injuries associated with mortality of hatchery reared Baltic cod (*Gadus morhua*), larvae. Acta Veterinaria Scandinavica 3: 385-390.
- Cosgrove, J.P., Church, D.F. and Pryor, W.A. 1987. The kinetics of the autooxidation of polyunsaturated fatty acids. Lipids 22:299-304.
- Ericson, G., Åkerman, G., Liewenborg, B. and Balk, L. 1996. Comparison of DNA damage in the early life stages of cod, *Gadus morhua*, originating from the Barents Sea and Baltic Sea. Mar. Environ. Res. 42 (1-4): 119-123.
- Grauman, G. and Sukhorukova, L. 1982. On the emergence of sprat and cod abnormal embryos in the open Baltic. ICES C. M. 1982/J: 7.
- Jones, J. C. and Reynolds, J.D. 1997 Effects of pollution on reproductive behaviour of fishes. Rev. Fish. Biol. Fish. 7: 463-491.
- Kamei, M., Ohgaki, S., Kanbe, T., Shimitzu, M., Morita, S., Niiya, I., MatsuiYuasa, I. and Otani, S. 1996. Highly hydrogenated dietary soybean oil modifies the responses to polychlorinated biphenyls in rats. Lipids 31(11): 1151-1156.
- Kime, D. E. 1995. The effects of pollution on reproduction in fish. Rev. Fish. Biol. Fish. 5: 52-95.

- Kjörsvik, E. 1994. Egg quality in wild and broodstock cod *Gadus morhua*. J. World aquacult. Soc. 25 (1): 22-29.
- Kjörsvik, E., Pedersen, B. H. and Wold, P. A. 1999. Egg quality and endoparasitic infection in eggs of Baltic cod, *Gadus morhua*. Nordic Council of Ministers, TemaNord 530: 143-160.

5

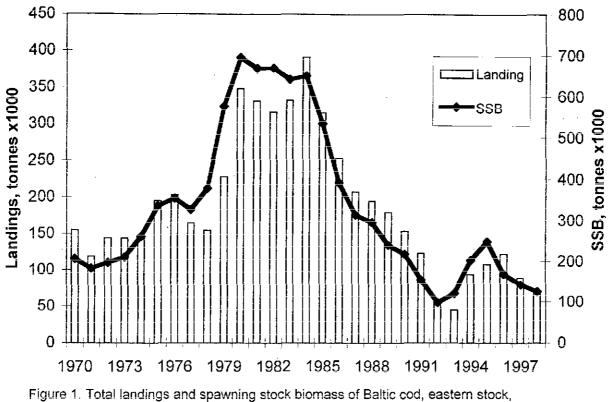
- Larsson, P-O. 1994. Recent development of the cod stocks around Sweden and possible reproduction disturbances. *In* Report from the Uppsala Workshop on Reproduction Disturbances in Fish (Norrgren, L., ed.) Swedish Environment Protection Board, Report no 4346: 26 – 34.
- Larsson, P-O. 1996. Can variation in spawning time affect the recruitment of Baltic cod? .
 In Report from the Second Workshop on Reproduction Disturbances in Fish (Bengtsson, B-E., Hill, C., Nellbrink, S. eds.) Swedish Environment Protection Board, Report no 4534: 70.
- Matsusue, K. Ishii, Y., Ariyoshi, N. and Oguri, K. 1997. A highly toxic PCB produces unusual changes in fatty acid composition of rat liver. Toxicology letters. 91: 99-104.
- McEvoy; L.A. and McEvoy, J. Multiple spawning in several commercial fish species and its consequences for fisheries management, cultivation and experimentation. J. Fish Biol. 41: Suppl B 125-136.
- Mercure, F. and Van Der Kraak, G. 1995. Inhibition of gonadotropin-stimulated ovarian steroid production by polyunsaturated fatty acids in teleost fish. Lipids 30 (6): 547-554.
- Mercure, F. and Van Der Kraak, G. 1996. Mechanisms of action of free arachidonic acid on ovarian teroid production in the goldfish. General and Comp. Endocrin. 102(1): 130-140.
- Miller, T.J., Crowder, L.B., Rice, J.A. and Marshall, E.A. 1988. Larval size and recruitment mechanisms in fishes: Toward a conceptual framework. Can J. Fish. Aquat. Sci., 45: 1657-1670.
- Nissling, A. and Westin, L. 1991. Egg mortality and hatching rate of Baltic cod in different salinities. Mar. Biol. 111: 29-32.
- Nissling, A. and Vallin, L. 1996. The ability of Baltic cod eggs to maintain neutral buyoancy and the opportunity for survival in fluctuating conditions in the Baltic Sea. J.Fish.Biol. 48:217-227.
- Norrgren, L, Amcoff, P., Börjesson, H. and Larsson, P-O. 1998. Reproductive disturbances in Baltic fish: A review. *In* Early life stage mortality syndrome in fishes of the Great Lakes and Baltic Sea. (McDonald, G., Fitzimons, J. D. and Honeyfield, D. C., eds.) Am. Fish. Soc. Symp. 21: 8-17.
- Otto, D.M.E. and Moon T.W. 1996. Phase I and II enzymes and antioxidant responses in different tissues of brown bullheads from relatively polluted and non-polluted systems. Arch. Environ. Contam. Toxicol. 31: 141-147.
- Pedersen, B. H. and Köie, M. 1994. A protistan endoparasite in embryo and yolk sac larvae of cod Gadus morhua and turbot Scophthalmus. maximum. Dis. Aquat. Org. 17: 57-59.
- Petersen, G., Gerup J., Nilsson, G., Larsen, J R. and Scheider, R. 1997. Body burdens of lipophilic xenobiotics and reproductive success in Baltic cod. ICES C.M. 1997/U: 10.

- Pettersson, A. and Lignell, Å. 1996. Decreased astaxanthin levels in the Baltic salmon and the M74 syndrome. *In* Report from the Second Workshop on Reproduction Disturbances in Fish (Bengtsson, B-E., Hill, C., Nellbrink, S., eds.) Swedish Environment Protection Board, Report no 4534: 28-29.
- Pickova, J. and Larsson, P-O. 1992. Rearing experiments with cod. Comparison between Baltic cod and Skagerrak coastal cod. ICES C. M. 1992/F: 12.

Pickova, J., Dutta, P., Larsson, P-O. and Kiessling, A. 1997. Early embryonic cleavage pattern, hatching success and egg-lipid fatty acid composition: Comparison between two cod stocks (*Gadus morhua*). Can. J. Fish. Aquat. Sci. 54: 2410-2416.

Pickova, J. 1998. Lipids in cod and Atlantic salmon eggs, with special emphasis on reproduction and quality. PhD thesis, SLU; Uppsala Sweden, Agraria no 123.

- Pickova, J., Kiessling, A., Pettersson, A. and Dutta, P. C. 1998. Comparison of fatty acid composition and astaxanthin content in healthy and by M74 affected salmon eggs from three Swedish river stocks. Comp. Biochem. Physiol. 120B: 265-271.
- Sargent, J.R., Bell, J.G., Bell, M.V., Henderson, R.J. and Tocher, D.R. 1995. Requirement criteria for essential fatty acids. J. Appl. Ichthyol. 11:183-198.
- Sjöstrand, B. 1999. Resurs 2000 Del 1. (In Swedish). Fiskeriverket Information Nr 6.
- Teshima, S. 1990. Sterols of crustaceans, molluscs and fish. *In*: Physiology and biochemistry of sterols. Glenn W. Patterson and W. David Nes (Eds.)
- Trippel, E.A., Castell, J.D., Neil, S.R.E. and Blair, T.J. In press. Assessment of egg quality of Haddock (*Melanogrammmus aeglefinus*) in paired matings. In: Proceedings 6th International Symposium on the Reproductive Physiology of Fish. Bergen 4-9 July 1999, Norway (Taranger, G.L. et al. eds.).
- Wesón, C., Sundin, P., Kvernheim, A. L., Mu, H. and Odham, G. 1995. Chlorinated fatty acids the major organochlorine compounds in fish. Scand. J.Nutr. 39: 170-174.
- Westernhagen Von, H., Dethlefsen, V., Cameron, P., Berg, J. and Furstenberg, G. 1988. Developmental defects in pelagic fish embryos from the western Baltic. Helgoländer Meeresuntersuchungen 42: 13-36.
- Wiegand, M. D. 1996. Composition, accumulation and utilization of yolk lipids in teleost fish. Rev. Fish Biol. Fisheries. 6 (3): 259-286.
- Wieland, K. and Horbowa, K. 1996. Recent changes in peak spawning time and location of cod in the Bornholm Basin, Baltic Sea. ICES C.M. 1996./J: 15.
- Winston, G.W. and Di Giullio, R.T. 1991. Prooxidant mechanisms in aquatic organisms. Aquatic Toxicol. 19: 137-161.



1970 - 1998. After Sjöstrand, 1999.

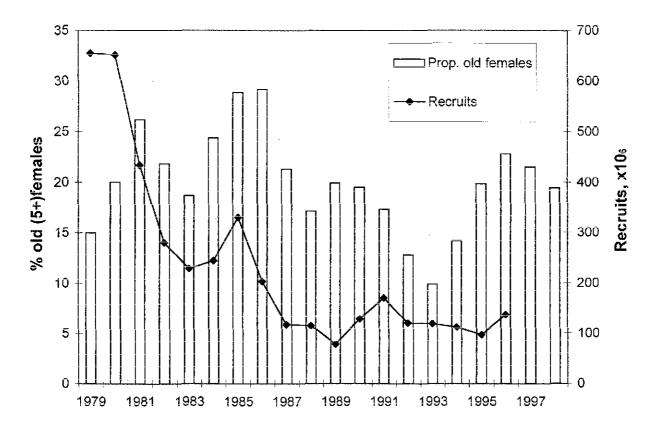


Figure 2. Proportion of old (5+) females of spawning stock and subsequent recruitment 1979 - 1996 of Baltic cod, eastern stock. Data from ICES Baltic Fisheries Assessment Working Group reports.