2.3.7 Potential impacts of escaped non-salmonid candidates for aquaculture on localized native stocks

Request

This is part of continuing ICES work to assess the potential impact of cultured marine fish species (non-salmonid) escaping from aquaculture sites and their effects on local wild marine fish (native) stocks with the view of developing risk assessment and management strategies.

Recommendations and Advice

ICES recommends that:

- Member Countries continue to conduct research on the interaction between wild and cultured non-salmonid marine fish. Research is also required to develop cost-effective genetic tools to discriminate cultured and wild stocks from the same habitat.
- Member Countries document and report all marine fish aquaculture escape events and submit reports annually to ICES (via the WGEIM).
- Member Countries use reproductively sterile fish in commercial mariculture operations wherever feasible.

Summary

Work continues on five documents dealing with the potential impacts of escaped marine non-salmonid finfish species. The 2005 WGEIM report contains the next draft in the series of reports expected, this one dealing with turbot (*Psetta maxima*) culture.

A standard risk analysis is being used to access the impacts and parallels the approach being taken by the World Organization for Animal Health (OIE) to the risk analysis for diseases of aquatic organisms. As discussed last year, this approach will allow Member Countries to tailor the application to their specific environmental conditions and cultured species under consideration.

Hazards associated with the culture of new exotic species and their associated disease interactions are not discussed as local regulatory authorities are responsible for evaluating the different species being considered for aquaculture using the ICES Code of Practice on the Introductions and Transfers of Marine Organisms and the OIE Protocols.

Scientific Background

The following material is the second in a series of state of knowledge reports being prepared by the WGEIM. Note that this is a draft document for which comments are being solicited.

Source of information

The 2005 report of the ICES Working Group on Environmental Interactions of Mariculture (WGEIM) and ACME deliberations.

1 Draft of "state of knowledge" of the potential impacts of escaped aquaculture turbot (*Psetta maxima*)

Turbot (*Psetta maxima = Scophthalmus maximus*)

1.1 Hazard identification

1.1.1 Distribution

Turbot is distributed throughout the Northeast Atlantic Ocean along the European coastline and is rarer around the Faroe Islands, Iceland and on Rockall Bank. Turbot is also found in the Skagerrak, the Kattegat, and the Belt Sea and in the Baltic Sea, but is very scarce in the Gulf of Bothnia, north of the Åland archipelago, where salinity levels are below 5 psu. The distribution area also extends into the Mediterranean and Adriatic Sea. It is typically found at a depth range of 10 to 70m. Turbot lives on sandy, rocky or mixed bottoms and is one of the few marine fish species that inhabits brackish waters.

1.1.2 Growth and Survival

Turbot is one of the fastest growing flatfish. Only halibut grows faster. During the juvenile phase growth rates are high, through which the turbot can reach 30 cm in three years. Females grow faster than males. During the first years of life females grow from 8 to 10 cm a year. Females older than 10 years still grow 1 or 2 cm a year. In male turbot the growth is already reduced to 2 cm a year at the age of 6 years. Males older than 10 grow less than 1 cm a year. The difference in length between the sexes increases from 3 cm in 3-year-old turbot to 9 cm in 10-year-old turbot.

The maximum growth rates are obtained in 3, 4 and 5-year-old turbot during the summer (May till October). In these months growth can reach between 2 and 2.6 cm per month. This high rate is comparable with the growth in artificial circumstances. In nature the ultimate growth rate (on year basis) is lower due to the slowing-down of metabolism during winter.

Ongenae and De Clerck (1998) concluded that in general no major differences in growth could be found among the areas under study. Males and females have a similar growth rate up to age 3. Hereafter the growth rate slows down in the males while the females continue their growth at a higher rate. Asymptotic lengths (L_∞) varied between 47.4 cm (North Sea) and 51.5 cm (Celtic Sea) for male turbot. For females the L_∞ ranged between 68.0 cm (eastern English Channel) and 74.4 cm (Celtic Sea). The asymptotic length thus attained highest values for both sexes in the Celtic Sea. The highest initial growth rate (characterised by the K-value) for both sexes was in the Bay of Biscay region. (Table 1).

Table 1. The von Bertalanffy growth parameters for turbot - $L(t) = L_\infty \{1\text{-}exp[-K(t\text{-}t_o)]\}$

Location	Sex	L_∞ (cm)	K (year^{-1})	t_0 (year)	Reference
North Sea	Male	55.50	0.23	-0.20	Mengi (1963)
	Female	64.10	0.23	-0.16	
	Male	49.20	0.37	-0.51	Jones (1974)
	Female	64.80	0.26	-0.05	
	Male	50.92	0.33	-1.13	Weber (1979)
	Female	68.65	0.23	-0.67	
	Male	47.4	0.44	-0.20	Ongenae & De Clerck, 1998
	Female	74.2	0.19	-0.85	
Eastern English Channel	Male	49.7	0.47	-0.04	Ongenae & De Clerck, 1998
	Female	68.0	0.26	-0.27	
Bay of Douarne-nez	Male	65.20	0.32	0.09	Deniel (1990)
	Female	73.60	0.28	0.08	
Celtic Sea	Male	51.5	0.41	-0.08	Ongenae & De Clerck, 1998
	Female	74.4	0.21	-0.44	
Irish Sea	Male	49.1	0.46	-0.14	Ongenae & De Clerck, 1998
	Female	71.5	0.22	-.054	
Bay of Biscay	Male	48.5	0.56	-0.01	Ongenae & De Clerck, 1998
	Female	71.5	0.27	-0.26	
Gulf of Lion (Med)	Male	54.3	0.24	-0.22	Robert & Vianet (1988)
	Female	55.6	0.31	-0.12	
Adriatic Sea	Male	67.7	0.27	-0.86	Arneri *et al.*, 1993
	Female	81.4	0.21	-0.99	
	Male	66.2	0.31	-0.14	Arneri *et al.*, 2001
	Female	81.5	0.21	-0.48	

The growth in weight indicated differences between some areas, but they appeared to be sex-dependent. When comparing the males it became clear that North Sea turbot had the slowest growth. Bay of Biscay and eastern English Channel male turbot indicated higher initial growth rates while Celtic Sea and eastern English Channel male turbot reached the highest weights (2400 g). For the females, the highest final weights were recorded in turbot from the Celtic and North Sea (8000 g). The asymptotic weight was least for females from the eastern English Channel stock (6300 g) (Table 2).

Table 2. The von Bertalanffy growth parameters for turbot - $W = W_\infty \{1\text{-}exp[-K(t\text{-}to)]\}^b$

Location	Sex	W_∞ (kg)	K (year^{-1})	t_0 (year)	b	Reference
North Sea	Male	1.91	0.44	-0.20	2.85	Ongenae & De Clerck, 1998
	Female	8.53	0.19	-0.85	3.11	
Eastern English Channel	Male	2.43	0.47	-0.04	3.04	Ongenae & De Clerck, 1998
	Female	6.33	0.26	-0.27	3.04	
Celtic Sea	Male	2.43	0.41	-0.08	3.10	Ongenae & De Clerck, 1998
	Female	8.04	0.21	-0.44	3.18	
Irish Sea	Male	2.14	0.46	-0.14	2.87	Ongenae & De Clerck, 1998
	Female	7.29	0.22	-0.54	3.10	
Bay of Biscay	Male	2.15	0.56	-0.01	3.22	Ongenae & De Clerck, 1998
	Female	6.93	0.27	-0.26	3.15	

Overview of the parameters of the length/weight relationships for turbot from different regions, it became apparent that the females show a higher allometric coefficient than the males, as this phenomenon occurs in almost every region. Male turbot from the English Channel and Celtic Sea has somewhat higher b-values, which means a slightly higher body weight for the same length compared to other regions. Male turbot from the Bay of Biscay have the highest allometric coefficient and thus the highest weight/length ratio (Table 3).

Table 3. Weight-length relationships ($W = a \, . \, L^b$) for different areas and for each of the sexes (Ongenae and De Clerck, 1998).

Location	**Sex**	**a**	**b**	$\mathbf{R^2}$
North Sea	Male	0.0325	2.8525	0.84
	Female	0.0133	3.1136	0.97
Eastern English Channel	Male	0.0173	3.0403	0.93
	Female	0.0168	3.0366	0.94
Celtic Sea	Male	0.0121	3.1016	0.97
	Female	0.0089	3.1845	0.98
Irish Sea	Male	0.0302	2.8714	0.96
	Female	0.0131	3.0998	0.98
Bay of Biscay	Male	0.0082	3.2182	0.94
	Female	0.0104	3.1538	0.96

1.1.3 Diet

Turbot is a typical visual feeder and feeds mainly on other bottom-living fishes (common gadoids, sand-eels, gobies, soles, dabs, dragonets, sea breams and boarfish), small pelagic fish (sprats, pilchards) and also, to a lesser extent, on larger crustaceans and bivalves. Large turbot (40 to 70 cm) feed from March till May excessive on herring and sprat (Rae & Devlin, 1972; Wetsteijn, 1981), to build up enough reserve for the subsequent spawning season. During the other nine months 50 to 70 % of the animals were found to have empty stomachs. This percentage was much higher than for most flatfish species. For example, a complete time of fasting, which is characteristic in the life cycle of lemon sole, *Microstomus kitt* is not observed in turbot (Rae & Devlin, 1972). The diet of the juveniles has been shown to consist of copepods, shrimps, barnacle larvae and gastropod mollusc larvae (Jones, 1973).

1.1.4 Abundance

Ongenae and De Clerck (1998) observed from the annual catches per unit effort, that the CPUEs for the North Sea and Celtic Sea with 1.0-1.2 kg/hour fishing were higher than for the English Channel, Bay of Biscay and the Irish Sea, with 0.5-0.8 kg/hour fishing.

Data from the annual Beam Trawl Surveys indicated a high abundance of turbot along the continental coast from Belgium to Denmark, with strong concentrations at the Dogger Bank and near the Wadden Sea and in the German Bight, and to a lesser extent the Scheldt estuary. In the English Channel, Celtic and Irish Sea, the overall abundance of turbot appears to be lower than in the North Sea. Other flatfish, such as sole mostly appear very abundant in the Thames estuary on the UK coast, but this was not the case for the turbot. It could be noted that turbot mainly occurred along the continental coasts of the North Sea. In the central and western part of the North Sea, turbot was much less abundant or even absent. Mainly in the central part of ICES-region Ivb, no turbot were caught. Catches by the International Bottom Trawl Surveys showed pronounced occurrence of turbot in the central parts of the North Sea and a lower abundance in the German Bight. Another remarkable difference between both survey types lies in the number of turbot caught per rectangle. These were substantially lower for the bottom trawl surveys. For these surveys, the occurrence of turbot along the east coast of the UK was observed in the years 1991-1995. This was not the case for the beam trawl surveys. Year to year comparisons for both surveys pointed out that overall abundance has decreased significantly over the years.

1.1.5 Migration

In general, turbot is rather a sedentary species, but there are some indications of migratory patterns. For example in the North Sea, migrations from the nursery grounds in the south-eastern part to the more northern areas have been recorded, since adult turbot is more tolerant of in the colder conditions in the northern areas of the Sea where temperatures are too low for juveniles to survive.,. A study in the northern Baltic of Aneer & Weston (1990) also indicated that adult turbot might be considered to be very stationary. In this project a large number of turbot were tagged and released. After recapture the average distance between first capture and recapture appeared to be very short: only 6 km. Furthermore, more than 90% of the recaptured turbot were caught less than 20 km away from the point of first capture.

1.1.6 Reproduction and spawning

Turbot exhibit no sexual dimorphism. The cyclical pattern of reproduction is characterised by massive gonad development and morphological changes (volume and colour), particularly of the ovaries, immediately before the emission of the gametes. In late spring to early summer, males and females gather on spawning beds, which are generally situated above gravel bottoms on the continental shelf. Fish with ripe gonads have been taken in trawls on the North Sea during the months April to July; ripe eggs have been found in the plankton from April to August (Malm, 1877; Möbius & Heincke, 1883; Brook, 1886; Ewart & Fulton, 1889; Fulton, 1892; Holt, 1892). Jones (1974) indicated the occurrence of ripe gonads between May and August. In the English Channel, the spawning season is rather long, viz. from May to September (Lahaye, 1972; Deniel, 1990). The eggs are released during the night in one batch and the fertilisation is external and at random.

The fertilised eggs are buoyant and their diameter varies between 0.9 and 1.2 mm. These eggs are extremely numerous: depending on the size of the female, their number ranges from 5 million up to 10 million per individual. The size-specific fecundity is rather constant. After spawning and feeding season, the turbot moves again to deeper waters.

First maturity for turbot in the North Sea is between ages 4 and 5 for females and age 3 for the males. This conclusion is drawn from a range of studies. Kyle (1926) determined maturity at age 6 or 7 for males as well as females. This (false) result went of course hand in hand with an incorrect age-length key. Ehrenbaum (1936) estimated first maturity at age 5 for both sexes. Length at maturity was determined by this author at 28 cm for the males and 35 cm for the females. Mengi (1963) estimated maturity at age 3, which corresponds to a length of 29-31 cm for males and 35-38 cm for females. In Rae's study (1972), maturity of the females was attained between 31 and 45 cm between the age of 4 or 5. Age of maturity for the males was set between age 3 and 4. Jones (1974) determined length, weight and age at which 50% of the females reached maturity as follows: 46,01 cm; 2001g and 4,46 years. For males a length at maturity of 30 cm was recorded. Deniel (1990) determined age and length at first maturation for the females in the English Channel at age 4 and 49 cm.

1.1.7 Further development

The fertilised eggs are carried to the shores by the currents. After more or less 7 days, the eggs hatch. At hatching, the larvae are 2.1-2.8 mm (Barnabé); 2.7-3.0 mm (Jones, 1972), 2.14-2.80 mm (Russell, 1976); 2.3-2.8 mm (Al-Maghazachi & Gibson, 1984) in length. Newly hatched turbot larvae possess a large yolk sac containing a single oil globule. This results in the larvae floating upside-down near the water surface during their first 6-12 h of life. At this time the larvae are largely inactive but may occasionally perform energetic wriggling movements. Larval growth and yolk utilisation are affected by temperature. The pelagic phase lasts around 60 days at 16°C (early summer). At the end of the larval phase the fish undergo metamorphosis, develop asymmetry, and descend to the bottom. Metamorphosis takes place at a length between 13-25 mm (NN); 23 mm (Jones, 1972); 27-39 mm (Jones et al, 1974); 38-45 mm (Al-Maghazachi & Gibson, 1984); 19.8 mm (Fukuhara *et al*, 1990). The rates at which morphological changes occur during larval development are partly under genetic control and partly reflect the influence of environmental factors such as temperature, diet and water quality.
Five major developmental stages can be recognised and are characterised as follows:

Stage 1: Larvae symmetrical, yolk sac present
Stage 2: Larvae symmetrical, development of spines and air bladder
Stage 3: Appearance of fin rays, notochord straight
Stage 4: Asymmetry and eye migration, notochord posteriorly slanted dorsally
Stage 5: Completion of eye migration, spines and swim bladder resorbed

There is no sharp distinction between the successive stages; in general at least half of the features characteristic of a particular stage must be developed before the onset of the next stage. For example, the right eye does not commence its migration until most of the fin rays have formed and the notochord within the caudal fin is inclined dorsally by 45° or more (Al-Maghazachi & Gibson, 1984).

The young fish, which were carried by the currents towards the shore, start a benthic existence. The juvenile turbot gather together on intertidal nursery grounds, where they remain throughout the summer months. In autumn they migrate from the coastal areas to deeper waters in the more Northern regions. The juvenile phase is characterised by a high growth rate.

1.1.8 Genetic structure of the populations

Only limited research on genetic stock analysis on turbot has been performed. In 1986, Renaud *et al*. (1986) showed in a study on allozymes of the cestode parasite, *Bothriocephalus gregarious*, a significant differentiation between the parasites from Atlantic and Mediterranean host turbot. The separation between these two forms was located in southern Portugal, between Lisbon and Faro. Allozyme analysis on 17 loci revealed almost no genetic differences within the complete distribution area turbot, only samples from the Aegean Sea were different from the others (Mediterranean to Kattegat), but with a negligible genetic distance as a result (Blanquer *et al*., 1992). Also Bouza *et al*., 1997 found, by the use of 14 allozyme markers a low genetic variability (P = 0.012) in both natural and hatchery populations. Imsland *et al*. (1994) did research on blood samples from turbot caught along the Norwegian coast, in Kattegat, and from the Southwest coast of Iceland. They found some genetic differentiation (P<0.01 for Hb-1) based on haemoglobin polymorphisms between Norwegian/Icelandic turbot and turbot from the Kattegat. Studies done with three microsatellite loci on wild and farmed turbot originating from two different locations (Norway and Ireland – Celtic Sea and the Western Approaches) also revealed a lack of significant differentiation between the two wild populations (Coughlan *et al*., 1998). Which is consistent with the low level of genetic differentiation found in the allozyme studies (Blanquer *et al*.,1992; Bouza *et al*., 1997). However, Coughlan *et al*. (1998) stressed the importance of further genetic analysis with more microsatellite loci to screen wild turbot across its distribution area. Bouza *et al*., 2002 found, employing 12 microsatellite and 28 allozyme loci, no differentiation between turbot from the Atlantic Ocean (Burela – 43°40'N, 7°22'W) and the Cantabric Sea area (Vilagarcia – 42°36'N, 8°45'W), areas which are separated by a major oceanographic discontinuity (Harden Jones, 1968). Recent studies carried out by Nielsen *et al.* (2004) on turbot from the Northeast Atlantic and the Baltic Sea (from the Bay of Biscay to the Aaland archipelago) suggests that the presence of multiple hybrid zones in the transition zone (Skagerrak, Kattegat and Belt Sea) between the high saline North Sea and the low saline Baltic Sea. The differentiation between turbot from the North Sea and the Baltic Sea was also observed by Karås and Klingsheim (1997) based on the effects of temperature and salinity on embryonic development of turbot from the two areas. Further research on population structure in the distribution area of turbot was undertaken by Boon *et al*. (2000). The preliminary study showed, using four microsatellites, that turbot from the English Channel appears genetically indistinguishable from the Bay of Biscay. Also turbot from the North Sea was not indistinguishable from the Celtic Sea. While turbot from the Irish Sea appeared to be genetically different from turbot from all other areas under research (Table 4).

Table 4. Matrix of genetic distance (DA) estimates above the diagonal and P-values below the diagonal, between turbot from different fishing grounds (Boon et al., 2000).

	North Sea	English Channel	Celtic Sea	Irish Sea	Bay of Biscay
North Sea	-	0.169	0.151	0.220	0.171
English Channel	0.052	-	0.196	0.220	0.120
Celtic Sea	0.111	0.005	-	0.235	0.208
Irish Sea	0.002	0.000	0.000	-	0.195
Bay of Biscay	0.019	0.367	0.002	0.001	-

Although samples sizes were small (20 samples per area) and these estimates must be considered as very preliminary, it appears likely according from the results of the statistical analysis that there exists a turbot population in the Irish Sea, which would be genetically different from the other areas under research. This was also noticed by Ongenae and De Clerk (1998) analysing the fishing and landing parameters. There was also a difference found (although not so significant) between turbot from the Celtic Sea and the North Sea, and turbot from the English Channel and the Bay of Biscay. The low genetic differentiation between the North Sea/Celtic Sea and English Channel/Bay of Biscay is caused by the low genetic differentiation between the samples from the English Channel and those from the North Sea. This could mean that the English Channel acts as a transition zone between the Bay of Biscay and the North Sea. Tagging experiments on several flatfish species (turbot, plaice, dab and sole) indicated migrations of small portions from the North Sea into the English Channel (De Clerck & Cloet, 1975; De Clerck, 1984; Delbare and De Clerck, 2000). A similar transition or hybrid zone was found between the North Sea and the Baltic (Nielsen *et al*., 2004) (Figure 1).

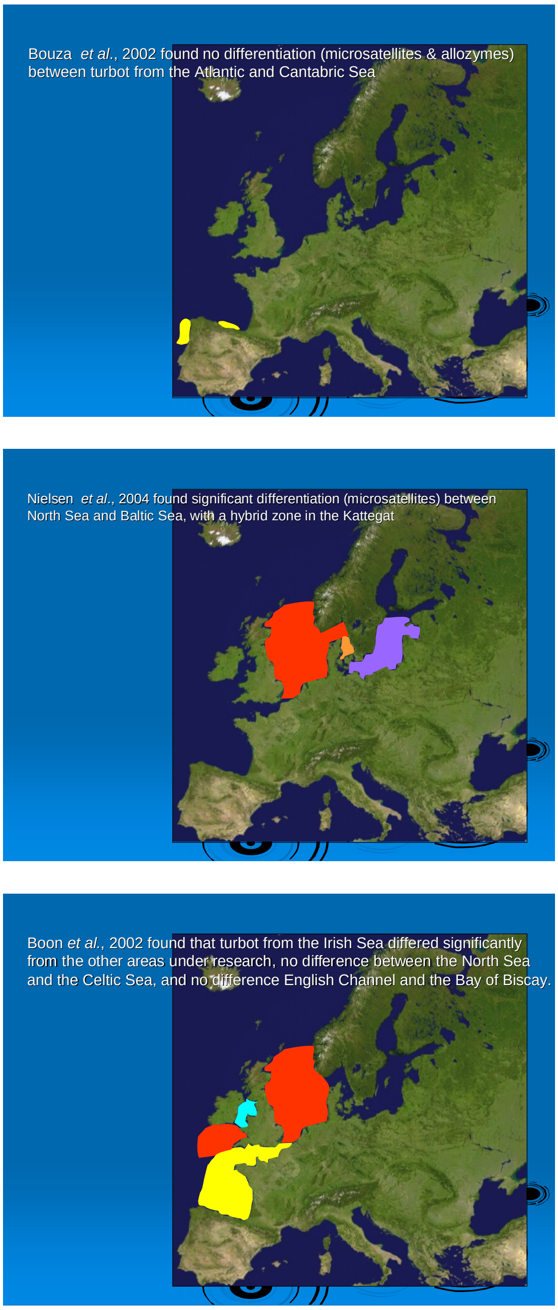

Figure 1. Areas which were studied and showed genetic differentiation.

Compiling all data from different studies, it becomes clear that there are distinct turbot populations in the Baltic Sea and in the Irish Sea. Furthermore there are indications that turbot from the North Sea, the southern coast of Iceland, the western coast of Scotland and Ireland, and the Celtic Sea (including the Western Approaches - 51°N, 10°W) forms another stock, the northern Atlantic stock. Which is different from the stock originating from the Bay of Biscay and the Atlantic site of southern Europe, the southern stock? Transition zones between the northern stock and the southern stock is found in the English Channel and between the northern stock and the Baltic Sea in Kattegat and the Belt Sea. The situation of turbot stocks in the Mediterranean is still unclear, although there are indications that samples from the Aegean Sea are genetic different from those originating from other areas (Figure 2).

Figure 2. Preliminary map of the population structure of turbot

1.2 Known effects of cultured populations

1.2.1 Genetic variability in broodstock

Only limited evidence exists for reduced variability in farmed strains of turbot, as was described for other cultivated fish species (Cross & King, 1983; Verspoor, 1988). Bouza *et al.* (1997) observed a reduction in heterozygosity in farmed strains of turbot in comparison with wild populations taken of the Norwegian coast and the Celtic Sea. This was also noticed by Coughlan *et al.* (1998) for farmed turbot from Norway and Ireland. Bouza *et al.* (2002) observed lower allozyme heterozygosity and loss of genetic variation in comparison with samples from the wild. The decrease in differentiation and divergence found in the farmed strains was believed to be caused by genetic drift during culture, due to the use of a limited number of broodstock animals. These results, however, can not be generalized, since broodstocks from other turbot farms in Galicia (Bouza, unpublished data) and France (Estoupe *et al.*, 1998) show much higher genetic diversity values, which were not different from the wild stocks.

Imsland and Jonassen (2001) observed that turbot was sensitive to the length of the light period, with longer light periods showing enhanced growth. But authors also revealed that growth in some cases was enhanced at lower temperatures and longer day lengths. Usually, warmer temperatures enhance growth. They concluded that a strong genotype by environmental interaction must be present.

1.2.2 Behaviour in the wild of released turbot

In the past, introductions of turbot have been carried out in the former USSR (1930) (FAO, 1997), in Iran (period 1930 – 1931) (Coad, 1995), and in Chile for aquaculture purposes (FAO, 1997, Pérez *et al*., 2003), but with no successful recapture or establishment of breeding populations. Turbot, however, was successfully introduced (self reproducing) into waters around New Zealand (Muus and Nielsen, 1999). Experimental releases of cultured fry for stock enhancement purposes have been performed in Spain (Iglesias & Rodriguez-Ojea, 1994), Denmark (Nicolajsen, 1993; Støttrup and Paulsen, 1998), and Norway (Bergstad & Folkvord, 1998).

The Sea Fisheries Department in Belgium has started to investigate the possibilities of restocking commercial important North Sea flatfish species, e.g. turbot and sole (*Solea solea*). Turbot was chosen as the first candidate, as reproductive biology and rearing techniques for all life stages are fully understood and under control. Delbare & De Clerck (2000) obtained 3000 juveniles from a commercial fish farm: France Turbot – Adrien Group (Noirmoutier, France) and reared for another 6 months in the pilot nursery system of the Department Sea Fisheries – CLO (Ostend, Belgium). Before release, the juveniles were conditioned for two months to natural live prey organisms, e.g. brown shrimp (*Crangon crangon* L.) and sand gobies (*Pomatoschistus* sp.). Next to that, all juveniles were tagged with a Petersen disk (Petersen, 1893). The tagged turbot were released in a for fisheries closed area (release position: 51°12’000 N and 02°45’600 E). Approximately 16% of the released turbot was reported back after a period of 1.5 years. At the end of 2004 more than 30% of the released turbot was reported. The migration pattern of the released turbot juveniles is presented in Figure 3. During the first two months after release, the juveniles remained in Belgian coastal waters following the main current towards the Dutch coast. The direction in the two following months (August-September’98) was clearly north – north-east, with the centre of capture on the Flemish sand banks. The same situation was found in October-November’98, although a portion of the animals was migrating into deeper water, i.e. the central part of the Southern North Sea. In the months December’98-January’99 some of the turbot were captured in the proximity of the “Thornton Bank”, while most migrated into deeper waters. Such an off-shore migration pattern, from shallow water during late spring and summer into deeper water during autumn and winter was also observed by Bagge (1987) for turbot in the Kattegat. In February-March’99, the major part of tagged turbot was still captured in deeper waters, with some found in more coastal waters (the Netherlands and the United Kingdom), but also into the Dover Straits, in the proximity of Bologne sûr Mer (France) and Port Rey (United Kingdom). This situation continued in the periods April-May’99, June-July’99 and August-September ’99. However, in the latter period, a concentration of turbot was seen again in the area around the “Thornton Bank”. Further captures (more than 30% of the released juveniles) were found scattered throughout the southern en central North Sea and the English Channel.

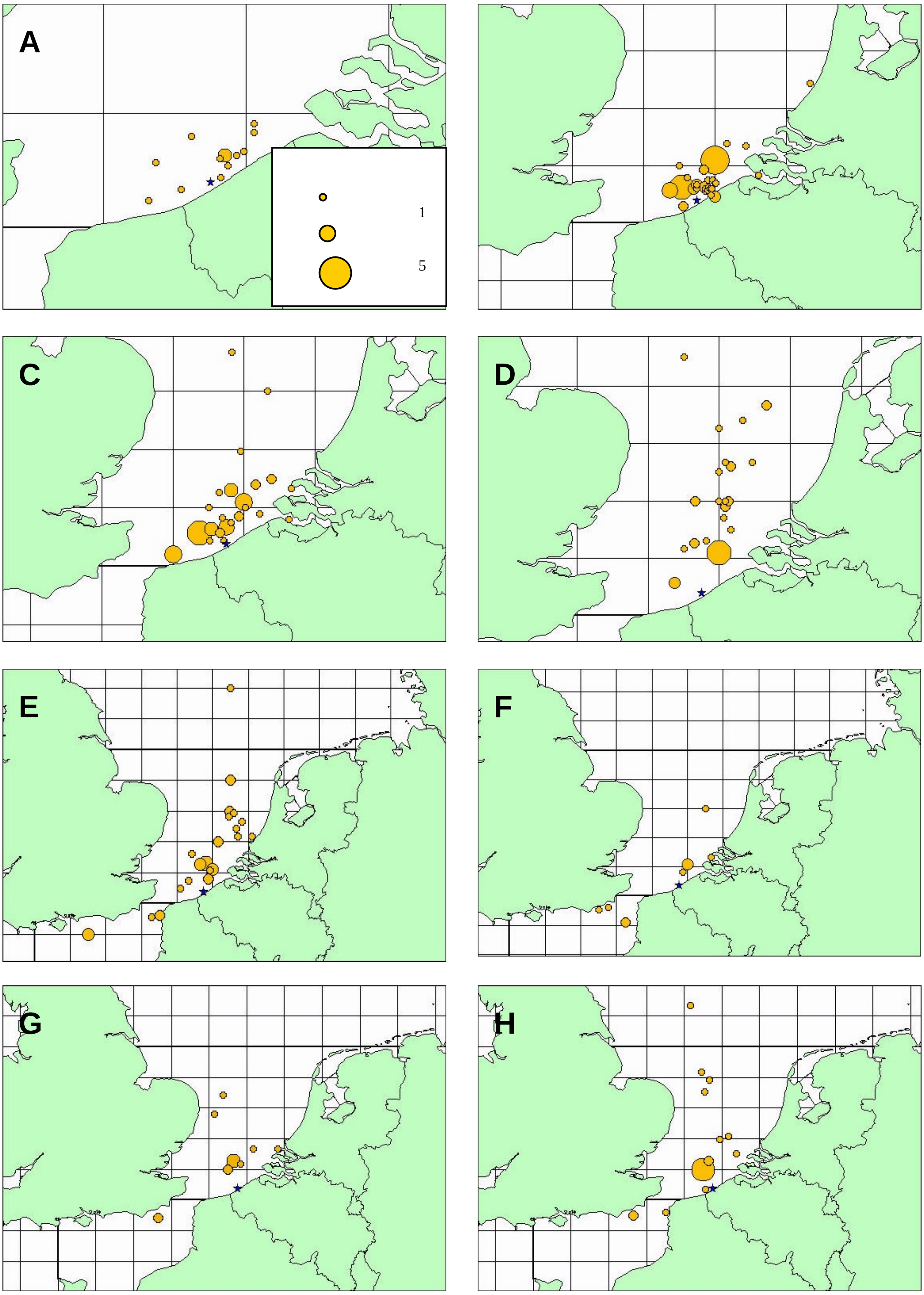

Figure 3. Distribution of the released turbot in time : A. June-July'98 ; B. August-September '98 ; C. October-November ; D. December '98-January '99; E. February-March '99 ; F. April-May '99 ; G. June-July '99 ; and H. August-September '99.

The general migration pattern of the released juvenile turbot followed a north – north-west direction into deeper waters of the North Sea, but with a migration to more coastal waters in late-spring and summer. Only a small portion migrated in south - south-western direction into the English Channel. Migration in northern direction started from October 1998 onwards, while tagged turbot in the English Channel were reported from February 1999 onwards. In tagging experiments with other flatfish species (plaice, dab and sole), it was also observed that a small portion migrated from the North Sea into the English Channel (De Clerck & Cloet, 1975; De Clerck, 1984). Growth rate was similar in comparison with the turbot in the wild, although these animals were initially bigger due to the high culture temperatures and *ad libitum* feeding. Other studies on released turbot revealed no differences in growth rate with their wild counterparts (Støttrup and Paulsen, 1998; Støttrup *et al.*, 1998a & b). The stomach analyses showed that the released turbot were able to adapt to the natural food sources. Turbot of the length class 21-23.9 cm fed exclusively on gobies (*Pomatoschistus* sp.). With increasing length, there is a change in prey spectrum, in which other bottom dwelling fish (e.g. lesser weaver, *Trachinus vipera* and dragonet, *Callionymus* sp.) and brown shrimp (*Crangon crangon*) were eaten. From 30 cm onwards a significant change in feeding habit occurs, raging from consumption of benthic organisms to hunting for pelagic fish, e.g. bib, *Trisopterus luscus.* The monthly variation in condition factor showed that the animals well adapted to the natural conditions, with a condition factor between 1.8 and 2.2, which was comparable with the range in wild turbot populations (Ongenae & De Clerck, 1998). Furthermore, no major differences in condition factor were noticed between released and wild turbot in the research period.

Several restocking experiments with turbot showed that survival rate of reared turbot in the wild were very high. Survival can, however, be further enhanced by conditioning the reared juveniles to natural conditions. Reared turbot were found to exhibit lower cryptic behaviour compared to their wild counterparts. After conditioning the reared animals to a sand bottom, the juveniles exhibited an improved cryptic behaviour and a more efficient burying technique (Støttrup and Nielsen, 1998). Stomach analyses on newly released turbot showed within two months after release lower stomach weights than wild fish of the same size. However, conditioning reared turbot to natural food increased the feeding success after release in the wild (Støttrup and Paulsen, 1998). Studies undertaken to estimate the carrying capacity of habitats along the European coastline revealed that the carrying capacity is rarely reached (van der Veer et al., 1990; van der Veer et al., 1991; Rijnsdorp et al., 1992; Henderson & Seaby, 1994) and could therefore sustain small quantities of released or escaped fish.

1.2.3 Effect of interbreeding between wild and escaped/released fish

At present no studies have been carried out on the interactions between wild and reared turbot. But for other species extensive data on interbreeding between escaped and wild individuals are available.

Among the main concerns is the loss of genetic variability within and among populations, with a reduction in flexibility to respond to environmental changes. This becomes a serious problem when the genetic variation within a hatchery population is reduced due to inbreeding, selective breeding, or domestication. Even one generation of artificial spawning and hatchery rearing can cause shifts in the genetic make-up (genetic variability and composition), with often detrimental effects to fitness (Allendorf and Ryman, 1987; Cross, 1999).

Interbreeding between wild and escaped domesticated salmon has been observed by Crozier (1993), Webb *et al.* (1993) and Clifford *et al.* (1998). Carr *et al*. (1997) and Saegrov *et al*. (1997) even noticed that in some cases the majority of the fry production in a population was produced by escaped cultured females. Other studies show that for salmon in certain Scottish rivers at least 7% of the spawnings are attributed to farmed female salmon (OSPAR QSR, 2000). Studies with Atlantic salmon demonstrated a significant superior survival of wild strains compared to farmed and hybrid strains under the same natural stream conditions, which means that there is a reduced fitness of the progeny from interbreeding. Fleming and Einum (1997) reported that farming of Atlantic salmon generated rapid genetic change that altered important fitness-related traits relating to behaviour and growth. Skaala *et al*. (1996) reported that survival of young juveniles was nearly three times higher in wild brown trout than in hybrids of wild and introduced (and genetically distinct) trout. Reisenbichler and Rubin (1999) reviewed a number of studies on Pacific salmon and concluded that they provide strong evidence that fitness for natural spawning and rearing can be rapidly and substantially reduced by interbreeding between wild salmon and those produced by artificial propagation.

A difficulty with demonstrating outbreeding depression is that the severity of the action becomes evident in the second and subsequent generation hybrids. Only few studies have continued to monitor the interactions over longer time periods, e.g. Jorstad *et al*. (1994) with cod *Gadus morhua*, and McGinnity *et al*. (1997) with Atlantic salmon *Salmo salar*. Perez-Enriquez *et al*. (2001) studied the genetic diversity of red sea bream (*Pagrus major*) in western Japan, in order to investigate the effects of stock enhancement programs around Shikoku Island on the genetic differentiation among wild stocks. They found significant departures from Hardy-Weinberg equilibrium and significant pair wise F_{st} among locations that indicated genetic instability within this region. It was suggested that stock enhancement caused this genetic instability. For Pacific salmon, Reisenbichler and Rubin (1999) also observed genetic changes from stock enhancement, which affected the productivity and viability in wild stocks. The effect of interbreeding between wild and cultured could cause catastrophic results to wild population in the long run. High numbers of escapees that interbreed with small populations, like in salmonids, can cause genetic incompatibilities between parents, that does only occurs in the second generation, when recombination of the parental genes takes place (Smoker *et al*., 2004). This, however, provides the possibility of increased hybrid formation until the second generation.

2 Risk Assessment

The specific risk under examination in this section is the consequences of releases (accidental or intentional) of cultured fishes on Fitness of Wild Populations of turbot due to Genetic Intergradation.

Our evaluation is based on the following set of conditions leading to the expression of a significant decline in survival in wild turbot populations is likely due to interbreeding with escaped cultured turbot

e.g.
1. Some turbot will escape captivity and,
2. will interact with wild stock by interbreeding and,
3. there will be significant differences between composition of the will and cultured stock, and,
4. those differences are such that the cultured population genome would be less well adapted to survival in the wild, and
5. the intergradation event will rapidly effect a large portion of the wild stock in question and,
6. hybrid cultured wild fish would lower survival of the fish population below the level to which the cultured fish enhance the number of fish in the population and,
7. the occurrence of intergradation will be repeated every year until …. and,
8. the duration of the depression in survival is likely to last for a number of generations after cessation of escapes

2.1 Release Assessment

2.1.1 Turbot in aquaculture

Turbot culture has developed rapidly in the last two decades, growing from 4 mt in 1984 to 6748 mt in 2003 in Europe. In China production is estimated at 3000 mt (approximately 33% of total turbot production) and 350 mt in Chile (approximately 4% of total turbot production). The majority of production systems for turbot are land-based recirculation systems for juveniles and on growing. Tank volumes can differ according to the farm and depends on the holding system in use. For example, small water volumes are used in “shallow raceway” systems or very high volumes of 3600 m^3 in Puraq`s Sunfish aquaculture (Cambados, Galicia). Maximum stocking densities are presented in Table 5.

Table 5. Maximum stocking densities (kg.m^{-2}) for turbot (Cachelou, 1992; Kamstra, 1992)

Start weight of the fish (g)	**1 End weight of the fish (g)**										
	1	5	10	40	75	125	300	600	1000	2000	5000
1		5	5	5	5	5	5	5	5	5	5
5			5	5	5	5	5	5	5	5	5
10				10	10	10	10	10	10	10	10
40					20	20	20	20	20	20	20
75						20	20	20	20	20	20
125							30	30	30	30	30
300								40	40	40	40
600									50	50	50
1000										60	60
2000											60

No information is available on the actual number of escaped fish from the turbot farms.

2.2 Exposure Assessment

Turbot is a widespread species (from Morocco to Norway and into the Mediterranean Sea), but only in low abundances. Total annual turbot production equals the total landing (approximately 7000 mt) of this species, but is concentrated in only a few areas. This means that an accidental release could mean a very sharp increase in turbot numbers in one area.

From a study carried out on turbot by Boon *et al*. (2000) the turbot population size in the North Sea for the period 1981-1989 was estimated at approximately 11000000 individuals. In 1990, however, there was a strong recruitment estimated at 60000000 one year old turbot with a total stock number of 68000000 individuals. The mean CPUE for the North Sea increased after 1990 (Ongenae and De Clerck, 1998).

Taken into account that almost 75% from European turbot landings originates from the North Sea and the CPUE data from the Beam Trawl Survey showing for the English Channel, the Celtic Sea and the Irish Sea rarely 5 ind. per hour fishing in certain ICES rectangles, total stock numbers must be much lower in these areas than in the North Sea and are somewhere in the range of:

- 1000000 individuals for the eastern English Channel (0.7 kg per hour fishing)
- 660000 individuals for the Celtic Sea (1.0 kg per hour fishing)
- 275000 individuals for the Irish Sea (0.6 kg per hour fishing)
- 770000 individuals for the Bay of Biscay
- No estimation available for stock numbers for the Atlantic coast of the Iberian Peninsula or the Mediterranean Sea.

Note of caution: these numbers are very crude approximations with considerable uncertainty but could reasonably be expected to be within 3 orders of magnitude.

No studies have been carried out on the interactions between wild and reared turbot, but interbreeding is most likely when the escaped/released turbot have matured, although it is not certain if these turbot have the sensory clues to migrate to spawning areas.

Turbot is a predator high on the trophic pyramid and release experiments have shown that reared turbot juveniles are very successful in adapting to conditions in the wild and have no problem in finding prey items (Støttrup and Paulsen, 1998, Støttrup et al., 1998a; Delbare & De Clerck, 2000). The natural predator avoidance strategy in flatfishes is to flee to the bottom, bury into the sediment and remain motionless. It is expected that such cryptic behaviour is not as effective in reared fish as in their wild counterparts, since turbot is cultured in bare bottom tanks. In some cases even lengthy off-bottom behaviour is displayed by cultured Japanese flounder (Tsukamoto *et al*., 1997). Reared turbot were found to exhibit lower cryptic behaviour compared to their wild counterparts. Avoidance of predators through burying in the sand is lower in reared turbot than for their wild counterparts. After conditioning to sandy bottoms, cryptic behaviour can be improved significantly (Støttrup and Nielsen, 1998). Conditioning is only carried out prior to controlled release in the wild. According to Iglesias and Rodriguez-Ojea (1994), however, cultured turbot buried immediately in the sand upon release in their stock enhancement experiments.

Studies on released turbot in the North Sea showed that juveniles dispersed through the North Sea and for a lesser portion moving into the English Channel. Off-shore migration was seen during autumn and winter, while near-shore migration took place in spring and summer. Similar migration patterns of turbot were observed by Bagge (1987) for wild turbot in the Kattegat.

2.3 Consequence Assessment

2.3.1 Establishment of turbot farms

Turbot farming is a well established mariculture activity in Europe and growth in production is still foreseen in the near future.

Conclusion: Many farms are active in turbot production

2.3.2 Differences between the genome of wild and cultured turbot

In many turbot farms it is the practice to use wild-caught mature adults as broodstock (Bouza, unpublished data; Estoupe *et al*., 1998). There is however evidence that more and more turbot farmers are selecting juveniles with high growth rates and less mal-pigmentation, in order to increase production outputs. Furthermore, several turbot farmers are obtaining fish from a select few hatcheries. For turbot, some evidence of lower allozyme heterozygosity and loss of genetic variability exists in farmed strains of turbot (Bouza *et al*., 1997; Coughlan *et al*., 1998; Bouza *et al.*, 2002).

Conclusion: Genetic differences have been observed between wild and cultured turbot.

2.3.3 Turbot escapees

Although turbot is mainly cultured in land based systems on recirculation, escapes are possible through outlets in flow through systems (when used) or by getting into dewatering channels by accident during sorting and handling of turbot and taken to the sea. Further impact on wild stocks could be expected through accidental release of fertilized eggs in the environment, since most incubation tanks are run in an open flow through system. The risk on escape will increase when culture systems are changed from on-land based systems to sea cage culture. In the latter, it is more likely that escapes could form a significant route for genetic interaction with the wild stock. Net cages can be damaged due to heavy weather conditions (storms), persistent predators such as seals that try to get at the fish, industrial accidents (human error or equipment malfunction), and even vandalism. So far, no information is available on the actual number of escaped fish from land based turbot farms, but the number is likely to be very small. However, with the use of sea cages for turbot, the risk on escapes could increase substantially, since accidents do happen. But it is predicted that the losses in net cage culture would be much lower than, for example the 20-25 incidents per year reported from 1998-2003 in salmon net pen aquaculture in Scotland (the escape rate is estimated at 0.1-1.0% of the stocked smolt; I.M. Davies, pers. comm).

Conclusion: escaped turbot from land based farms is likely, but in very small numbers, especially in land based flow through systems and during sorting and handling. The risk becomes much higher for net cage cultured turbot.

2.3.4 Interbreeding

Although there have been no studies carried out on the interactions between wild and reared turbot, interbreeding is most likely when escaped turbot have matured, Studies of released juvenile turbot in the wild have shown to exhibit only small differences in feeding and cryptic behaviour (for a short period after release). Migratory patterns of released turbot showed a similar off-shore migration during autumn and winter, and a near-shore migration in spring and summer, as seen in wild turbot (Bagge, 1987). But it is not certain that escaped turbot have the necessary sensory clues to migrate to spawning areas.

As a precautionary approach, one can look to other examples of interactions between reared and wild fish. In Atlantic salmon there is clear evidence of interbreeding between wild and escaped domesticated individuals.

Conclusion: evidence for interbreeding in turbot is not given but is known in other species, for example Atlantic salmon.

2.3.5 Reduced fitness caused by interbreeding

There exists no evidence for a reduced fitness caused by interbreeding between wild and cultured turbot. As a precautionary approach, one can look to other examples of interactions between reared and wild fish. In brown trout, Pacific and Atlantic salmon, there is clear evidence of reduced fitness of the progeny from interbreeding.

Conclusion: there is no evidence for reduced fitness by interbreeding in turbot, but there reduction of fitness was observed in salmonids.

2.3.6 Risk on affecting population fitness

The knowledge about the population structure throughout the distribution area of turbot is still incomplete. Currently, the situation in the north-western part of the Atlantic Ocean is that there are two distinct populations in the Baltic Sea and in the Irish Sea. Furthermore, there are indications that there is a northern (sub) population (North Sea/Celtic Sea, including the Western Approaches - 51°N, 10°W) and a southern (sub) population (English Channel/Bay of Biscay/Atlantic coast of Spain). Between the northern (sub)population and the Baltic population there is a transition zone, situated in Kattegat and the Belt Sea, and a second between the northern and southern (sub)population in the English Channel. Turbot is a wide spread species (from Morocco to Norway and into the Mediterranean Sea), but is found in low abundances. These abundances have decreased significantly over the last years. However, with the reduction in TAC of sole and plaice, in order to protect certain sole and plaice stocks, but also cod in general, fisheries mortality of the wild stocks will decrease, as turbot is a by catch product of beam trawling on sole and plaice. It is expected that accidental escapes of small numbers of reared turbot have a limited negative impact on wild populations, but could be substantial in areas with explicit stock characteristics, e.g. the Baltic and the Irish Sea.

Conclusion: wild populations with a limited distribution area and under high fishing pressure can be affected by interbreeding.

2.3.7 Decline in survival in wild turbot populations

There is no evidence to support this contention for turbot. As a precautionary approach, one can look to other examples and in Pacific salmon lower productivity and viability in wild stocks were observed after inbreeding with domesticated salmon.

Conclusion: there is no evidence in turbot for a loss in fitness after interbreeding, but reduction in productivity and viability in offspring was observed in Pacific salmon.

2.3.8 Escapes of farmed turbot cause significant decreases in wild/feral turbot stocks

There is no evidence to support this contention for turbot. As a precautionary approach, one can look to other examples and in salmon indications were found that interbreeding between wild and cultured could cause catastrophic results to wild populations.

Conclusion: there is no evidence in turbot for decrease in wild stocks due to interbreeding, but genetic changes leading to reduced survival in the wild is a feature of all domesticated salmon and consequently in hybrids from farmed and wild fish.

3 Risk evaluation

Risk evaluation based on a set of conditions (see above) leading to the expression of a significant decline in survival in wild turbot populations is likely due to interbreeding with escaped cultured turbot (Table 6).

Table 6. Risk evaluation for interbreeding between escaped cultured and wild turbot risk.

	Severity	Probability	Uncertainty
1. Establishment of turbot farms	N	H	N
2. Differences between the genome of wild and cultured turbot	N	H	L
3. Turbot escapees	L	EL	M
4. Interbreeding	M	EL	M
5. Reduced fitness caused by interbreeding	M	EL	H
6. Risk on affecting population fitness	H	N	H
7. Decline in survival in wild turbot populations	H	N	H
8. Escapes of farmed turbot cause significant decreases in wild/feral turbot stocks	C	N	H

Severity: C-Catastrophic, H-High, M-Moderate, L-Low, EL-Extremely low, N-Negligible

Catastrophic: The occurrence of a risk that would be expected to cause serious irreversible harm to ecosystem performance at the faunal level

High: The expression of a risk that would have serious biological consequences

Moderate: The change that has a less prorated biological consequence.

Low: The expression of a risk has mild consequences and would be amendable to control or mitigate.

Negligible: The measurable changes are not significant at an ecosystem level and are readily amendable to control or mitigation.

Probability: H-High, M-Moderate, L-Low, EL-Extremely low, N-Negligible

High: There is high probability that the event will take place

Moderate: There is a reasonable probability that the event will take place

Low: There is a chance that the event could take place

Negligible: Chances are rare that the event will take place

Uncertainty: H-High, M-Moderate, L-Low, N-Negligible

High: The chance of the risk being expressed is so small that it can be ignored

Moderate: There is reasonable uncertainty as to whether the risk will be expressed

Low: The risk is more likely than not to be expressed

Negligible: the event is very likely

4 Effect of Infrastructure on Risk

4.1 Regulation

- There is some indication that there exists several (sub) populations, which probably have there own optimal growth temperature and salinity range, especially for the Baltic and the Irish Sea. However, it is advisable to use as broodstock animals from those stocks that are best fitted for that specific culture location. Decrease in genetic differentiation and divergence was found in the farmed strains, therefore special breeding programs must be set up to guarantee a high level in heterozygosity in farmed strains. Furthermore, it is important to use broodstock animals for restocking that are related to the local turbot (sub) population, in order to minimize adverse genetic interactions with the wild stock.

- The use of triploid turbot would reduce the risk of interactions with the wild stock. Experiments with hybrids between turbot and brill have been carried out to produce only female offspring (Purdom and Thacker, 1980).

- To limit escapees, physical barriers must be installed in all outlets of open flow through systems. Double mesh screens must be installed in the outlet of broodstocks at all times, toe prevent fertilized egg loss. Closed recirculation techniques can further reduce the risks of escapes.
- Particular attention should be paid to robust containment technologies for sea cages, when cage culture of turbot would become feasible.

4.2 Code of practice – certification

- In all cases, the training of operators should be an essential preoccupation by the fish farmer. The maintenance and cleaning of tanks and in case of cage culture the replacement and monitoring of nets is of the outmost importance to limit accidental escapes. Periodic inspection of tanks (outlets and physical barriers) and nets should be compulsory. Special attention should be paid to the procedures of sorting and treatment operations.
- At present, declaration of turbot escapees is not compulsory in any country. To reduce uncertainty, the need for regulatory enforcement, and improved mandatory reporting should be introduced. Since there is no additional cost inferred to it, it would be profitable to both the industry and the environment. In Ireland salmonid farmers is obliged immediately following any escape of reared salmonids from a freshwater or marine installation, to fill out a *Reared Fish Escapees — Incident Report Form* and contact the Department of Communications, Marine and Natural Resources (DCMNR), Marine Institute and relevant Regional Fisheries Board(s). The operator is required to report the number of escapees and cause of the escape, if known. The DCMNR collates this information with a view to making recommendations to try and prevent other incidences from happening. Nevertheless, there are no accurate data available for the number of escapes in Ireland. Voluntary Codes concerning escapes: aquaculture industry self-regulation and environmental safeguards through voluntary Codes are effectively worthless forms of governance in the absence of binding legal obligations to enforce rules (See Regulation of Marine Aquaculture). Concerning stock health management, it is a recommended action under the Code to implement the Irish Salmon Growers' Association (ISGA) Code of Practice for the prevention of stock escapes of Irish farmed salmonids (reproduced in Annex III of ECOPACT). The ECOPACT document also annexes the Federation of European Aquaculture Producers (FEAP) Code of Conduct for European Aquaculture.
- EU policy on escapes: in its Communication A strategy for the sustainable development of European aquaculture (COM (2002) 511 final, 19/9/02) the European Commission states that 'escaped fish inter-breeding with native populations may induce long-term damage by the loss of genetic diversity'. The Commission proposes developing instruments to tackle the impact of escapees as part of the EU Aquaculture Strategy and states that it 'has financed research on the threats to the diversity of wild Atlantic salmon caused by farm escapees, but further studies are needed. The process started in February 2000 by NASCO and the North Atlantic salmon farming industry to develop guidelines to minimise salmon escapees is particularly worthy of support. The Commission will examine whether such guidelines should be implemented by way of compulsory rules and may extend them to other fish species and strains.'

5 Risk Management

Identify a list of options for controlling risk e.g.

5.1 Keep all culture on land

Some experiments have been carried out in Scotland for cage culture turbot, but with limited success. Recirculation techniques are improved to be able to culture turbot on full recirculation in order to enhance the control and reduce the dependence on natural water resources and heating costs. Nowadays, most of the turbot is farmed at 75-80% recirculation. It is expected that in the next five years all turbot are farmed at 100% recirculation. The severity on escapees and probability would be reduced and subsequently the interaction between wild and farmed turbot. With land based culture of turbot on full recirculation, the uncertainty of escapes would be negligible

5.2 Use sterile fish

Manipulation of sex and ploidy are being introduced in fish farming. All-female production is economically advantageous, because in many species female growth rate is higher compared to that of the males and first maturation takes place at older age. The market value declines drastically with maturity. For example, all female rainbow trout *On-*

corhynchus mykiss are cultured in Europe (Ingram, 1988) in order to reach greater market size before maturing. Tests with hybrids have been carried out with brill, *Scophthalmus rhombus*. These hybrids showed a higher survival rate during larval development and metamorphosis. Hybrids between these two species can also been found in nature. Holt (1892) noted three hybrids caught in the North Sea differed significantly from turbot or brill in body form, colour, scale and number of fin rays. Hybrids formed between a female turbot and a male brill, were all females and could reach a weight of 382 g at natural temperatures. While hybrids formed between a male turbot and a female brill resulted in an all male stock and grew to 289 g in 20 months at the same temperatures (Purdom and Thacker, 1980). Successful experiments to obtain an all-female stock were carried out in the UK and are interesting for the farmer since they exhibit a 12% high growth rate and only rudimentary development of the ovaries. In turbot females, ovaries can take up 15% of the total body weight (Bye, 1981) as gonad development can divert much energy from somatic growth. Induced triploidy is also used to produce sterile fish, which continue to grow somatically (Ingram, 1988). The severity on interaction between a wild and all-female turbot would remain the same, but the probability would be reduced to extremely low to negligible. The uncertainty would be extremely low to negligible. In Europe experiments with trout and turbot (Vázquez *et al.* 1996; Cunado *et al*., 2002; Terrones *et al.*, 2004.) have been carried out, although for turbot this technique is not used in commercial farms.

5.3 Create dependence on specific food supplements that are not readily available in the wild

Another possible technique is to produce genetically modified turbot, that are incapable to synthesize certain nutritional components and which are not available in nature. Reared turbot would therefore be totally relying on the artificial diets given in captivity providing these essential nutrients. Once a fish has escaped from the rearing system, it could not survive in the wild due to deficiency. This technique would reduce the severity of escapes. The probability that the escapee would find food containing the essential nutrients depends on the chosen component, but would be very low. The uncertainty could be low to moderate, taken into account that one has a very good knowledge about the natural prey items. This feeding technique is still highly hypothetic and needs substantial theoretical developments (animal welfare, technical feasibility, GMO regulations, human welfare, etc.).

5.4 Contingency Planning

The degree of monitoring all activities on the farm must be in function of the degree of risk in relation to the farming system. In this respect there is a decreasing need for intensive control from sea ranching to inshore sea cages, flow through land based systems to closed recirculation culture systems.

Recovering large number of escapees in a certain area could be carried out by using gill nets in the area shortly after the accident has occurred.

5.5 Research

- Better information on total numbers, spawning areas and the genetic structure is needed to evaluate the severity, probability and uncertainty about the interaction of domesticated escapees on wild populations.
- Studies are needed to determine the number and survival of escapees in the natural environment (in function of season and space, impact of escaped turbot in winter may be different from winter, due to the presence of natural predators, e.g. cod migrating to the north in winter). This is needed to evaluate the severity, probability and uncertainty about the interaction of domesticated escapees on wild populations.
- Development of tools to distinguish wild fish from wild population. For turbot, the morphology of reared turbot is slightly different in comparison with their wild counterparts. In turbot, as in other flatfish species, morphological differences are primarily seen as mal-pigmentation on the blind side to patches with lack of pigment and white pigment on the eyed side. Such mal-pigmented turbot are found in the wild but at much lower frequencies than in cultured turbot, as it is determined through the larval diet and possibly the rearing conditions. Further morphological characteristics, like the general form of the turbot, is highly influenced by the stocking densities in the culture tanks. Tagging could be a method, but due to high stocking densities in the tanks, these tanks could harm other turbots. The most common chemical compounds used to mark otoliths are alizarin compounds (Beckman & Schultz, 1996; Tsukamoto, 1985), calcein (Brooks et al. 1994; Wilson et al. 1987) and oxytetracycline (Dabrowski & Tsukamoto, 1986; Nagiec et al. 1988; Schmitt, 1984). Studies that address what levels of escapees will cause problems for local populations and their impact on the different life-cycle stages of their wild counterparts. Discrimination

between reared and wild turbot is useful to act after an accidental release of farmed turbot in the wild, in order to reduce the severity. Probability and uncertainty of escapees stays the same.

Final evaluation of unmanaged risk should be located on an Acceptable Level of Protection matrix with annotation to show the effect of uncertainty. Also plotted should be the level of risk associated with each of the risk management options.

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