European Food Safety Authority

*The EFSA Journal* (2009) 1073, 1-34

# SPECIES-SPECIFIC WELFARE ASPECTS OF THE MAIN SYSTEMS OF STUNNING AND KILLING OF FARMED TURBOT[1]

## Scientific Opinion of the Panel on Animal Health and Welfare

**(Question N° EFSA-Q-2008-442)**

**Adopted on 30th April 2009**

**Panel Members**

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1 For citation purposes: Scientific Opinion of the Panel on Animal Health and Welfare on a request from the European Commission on welfare aspect of the main systems of stunning and killing of farmed turbot. The EFSA Journal (2009) 1073, 1-34.

## Summary

Following a request from the European Commission, the Panel on Animal Health and Welfare was asked to deliver a scientific opinion on welfare aspect of the main systems of stunning and killing of farmed European Turbot *(Psetta maxima).*

A semi-quantitative risk assessment approach was used to rank the risks of poor welfare associated with the different commercially applied stunning / killing methods for European Turbot and to identify areas of concern, as well as to provide guidance for future research. The risk assessment was based on expert opinion, due to the limited amount of quantitative data and published peer reviewed data on the effects of the hazards associated with the killing of turbot. Pre-slaughter stages, immediately before killing, which had a direct impact on welfare were included in the risk assessment. Stunning methods such as electrical stunning and percussion that are not commercially used in Europe were also described but not included in the risk assessment. The two methods assessed were: exsanguination and asphyxia on ice, the latter being the most commonly used method in the EU.

The pre-slaughter stages considered were common to all killing methods: i) Feed withdrawal; ii) Crowding iii) Removal from water and iv) chilling in ice water slurry. The pre-slaughter procedure of chilling turbot in ice water slurry represents a welfare risk because it can cause cold shock in conscious fish which is known to cause distress due to involuntary muscle contractions. Live chilling is an immobilisation method and not a stunning method since it does not induce unconsciousness.

At present, turbot are not stunned prior to slaughter under commercial farming conditions. Existing methods of killing turbot, exsanguination and asphyxia on ice, involve prolonged periods of consciousness during which stress responses have been observed, and they constitute a considerable welfare risk. Trials involving alternative methods, especially electrical stunning which induces immediate loss of consciousness followed by chilling in ice water slurry, have shown promising results for turbot welfare and meat quality. As a matter of urgency, industry should be encouraged to test and develop commercially viable alternative methods such as electrical stunning followed by chilling or percussive methods, which induce immediate loss of consciousness. Standard operating procedures to improve the control of the slaughter process to prevent impaired welfare should be introduced and relevant practical monitoring welfare indicators developed.

Although turbot are susceptible to a notifiable disease Viral Haemorrhagic Septicaemia (VHS) specific operating procedures and detailed contingency plans are lacking. Large scale killing methods developed for other species of fish can be applied under disease outbreak situations in turbot but they need to be evaluated in turbot and in their developmental stages.

At present there are no validated and robust indicators available to evaluate in practice the welfare of turbot associated with slaughter procedures and their development is recommended.

Further research is also recommended regarding temperature tolerance limits in live chilling. The existence of nociceptors specific to temperature shock and pain are not established and need to be investigated.


**Key words:** Fish**,** European turbot, *Psetta maxima*, animal welfare, risk assessment, pre-slaughter, slaughter, stunning, killing.

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## Background as provided by European Commission

Directive 93/119/EC[2] provides conditions for the stunning and killing of farm animals. Fish are legally part of the scope of the EU legislation but no specific provisions were ever adopted.

Following a previous request from the Commission, EFSA issued in 2004 a scientific opinion on the welfare aspects of the principal methods for stunning and killing the main commercial species of animals[3], including farmed fish. As regards farmed fish, this opinion concluded that *"Many existing commercial killing methods expose fish to substantial suffering over a prolonged period of time."* Furthermore, *'for many species, there is not a commercially acceptable method that can kill fish humanely"*.

Moreover, this EFSA report[4] highlighted that different methods for stunning and killing of farmed fish must be developed and optimised according to the species specific different needs and welfare aspects:

*"Fish are often treated as one species when it comes to regulations and legislation governing welfare during farming or at slaughter. But, it is important to realise that a very wide number of species of fish are farmed, with an equally wide variety of ecological adaptations and evolutionary developments. These differences mean that different species fish reacts differently to similar situations. For example, at a given environmental temperature, some species like trout die relatively quickly when removed form water into air, whilst others like eel or marine flatfish can take several hours. Similarly, in electrical stunning situations, eel require a much larger amount of stunning current than trout or salmon to render them unconscious species differences need to be taken into account when adopting particular procedures. Processes must be developed and optimised with respect to welfare specifically for each species. For example, it would be as unreasonable to assume that a process developed for killing trout in freshwater would be suitable for killing tuna in the sea as it would be to assume that a system developed for quail would be effective on ostriches."*

## Terms of Reference as provided by European Commission

In view of the above, the Commission requests EFSA to issue a scientific opinion on the species-specific welfare aspects of the main systems of stunning and killing of farmed fish. The opinion should assess whether the general conclusions and recommendations of the 2004 opinion apply to the species of fish specified below. Furthermore, the above mentioned conclusions and recommendations should be updated in a species specific approach, integrating where possible reference to welfare indicators and to new scientific developments. Where relevant, the animal health and food safety aspects should be taken into account.
The following species should be considered:

- Atlantic salmon *(Salmo salar)*
- Rainbow trout *(Oncorhynchus mykiss)*
- European eel *(Anguilla anguilla)*
- Gilthead seabream *(Sparus auratus)*
- European seabass (Dicentrarchus labrax)
- European turbot *(Psetta maxima)*
- Common carp *(Cyprinus carpio)*
- Farmed tuna *(Thunnus* spp*)*

2 OJ L 340, 31.12.1993, p. 21–34

3 http://www.efsa.europa.eu/cs/BlobServer/Scientific_Opinion/opinion_ahaw_02_ej45_stunning_en.pdf?ssbinary=true

4 http://www.efsa.europa.eu/cs/BlobServer/Scientific_Opinion/opinion_ahaw_02_ej45_stunning_report_v2_en1,1.pdf?ssbinary=true

**Acknowledgements**

The European Food Safety Authority wishes to thank the members of the Working Group for the preparation of this opinion: Mohan Raj (Panel member, chairman), David Morton (Panel member), Bjorn Roth and Ed Peeler (Risk assessor).

Albert Imsland and Ana Riaza who were invited as Hearing experts are gratefully acknowledged as well as Hans Van De Vis for his contribution on the section on electrical stunning.

The AHAW Panel also would like to thank the Member States and stakeholders organisations for their valuable comments which were taken into consideration by the WG Members in producing the Scientific Opinion.

The scientific coordination for this Scientific Opinion has been undertaken by the EFSA AHAW Panel Scientific Officers Oriol Ribó, Franck Berthe, Sofie Dhollander, Tomasz Grudnik, Jordi Tarrés-Call and particularly Ana Afonso.

## Scope and objectives of the scientific opinion

The scope of this report is the animal welfare aspects of the stunning and killing of farmed European turbot, *Psetta maxima* (L.)[5] .
Pre-slaughter procedures should only be considered if evidence exists for a direct impact on welfare at stunning and killing. Where fish welfare immediately before and during killing or stunning is affected, it is also considered as part of the process. Therefore, the welfare aspects of the farming phase of these species as well as the transport are not included in this report.

The impact on meat quality is not part of this assessment however, references are provided in the text that could be used and evaluated for further socio-economic study on stunning and killing methods for turbot.

Emergency killing for disease control or other reasons is included in the report. However, humane killing of individual fish, in the course of farming operations (i.e. sorting, grading, or background morbidity) is not included.

Food safety issues are to be dealt with by the BIOHAZ panel.

In drafting this Scientific Opinion, the panel did not take into consideration any ethical, socio-economic, human safety, cultural or religious or management issues, the emphasis has been to look at the scientific evidence and to interpret that in the light of the terms of reference. Nevertheless, it is acknowledged that such aspects can have an important impact on animal welfare.

## 1. Introduction

Turbot is a marine demersal carnivorous flatfish of the Scophthalmidae family. It is relatively abundant in Europe, from Iceland (66ºN) and western Norway in the north to Morocco (30ºN) in the south. It is also abundant in the Mediterranean Sea as far as Turkey.

The studies of Aneer and Westin (1990), Déniel (1990), Iglesias and Rodríguez-Ojea (1994), and Bergstad and Folkvord (1997) indicate that turbot do not undergo long migrations but is a stationary species. Different spatial distribution between juveniles and adults is seen as only large fish migrate to colder areas (Aneer and Westin, 1990; Iglesias and Rodríguez-Ojea, 1994). This difference may be partly explained by the decreasing temperature sensitivity with size increase and a downshift in temperature optimum with size (Imsland et al., 1996, 2001a, 2006a); it might also be a strategy to reduce predation risk. Lack of long migration, together with the fact that this species is found in different environments (e.g. different salinities), makes it reasonable to believe that turbot in European waters belong to more than one genetically diverse population.

Following the initial commercialisation of turbot farming in the UK and France during the 1980s (Jones et al., 1981) the emerging industry became centred in northern Spain, owing to favourable water temperatures for on-growing. The industry has subsequently consolidated and output has risen gradually to approximately 6000 tonnes in Europe in 2005. Accurate numbers for production in other parts of the world are difficult to obtain, but is estimated to be around 3000 tonnes in 2005 (400 t in Chile, 2500 t in China). The farmed product has increasingly gained commercial acceptance especially in the Spanish and French markets due, in part, to feed improvements and to the greater availability of larger-sized fish. Consolidation of

[5] European turbot, *Psetta maxima* (L.) is often referred in the scientific literature as *Scophthalamus maximus* (Rafinesque 1810).

production has taken place both in the on-growing and in the hatchery sectors. Juvenile supply is dominated by one company, which produced approximately 5 million intensively reared turbot in 2001.

Applying optimal rearing temperatures, a 2 kg fish can be produced in 18-22 months. However, the majority of aquaculture production of turbot is now in land-based flow-through systems in Spain, which is based on the ambient temperature cycle of the Spanish coast. Farms located in North Western Spain and Portugal contribute to 75% of European turbot production. The remaining production is from: France (Brittany, Bay of Biscay) approximately 1000 tonnes).Warm water and re-circulation systems are also in use in Iceland, Norway, Wales, Netherlands, Denmark (1 farm recirculation) and Germany (1 farm recirculation). Turbot are nursed in square or circular tanks (10-30 $m^3$) with open-circuit pumped seawater. Aeration systems are usually used to maintain the water at oxygen saturation. Juveniles are fed with dry pelleted feed, introduced manually or automatically. Turbot juveniles grow from 5 to 100 g in the pre-fattening period (duration 4-6 months).

For on-growing square or circular cement tanks (25-100 $m^3$) are used, with open-circuit pumped seawater. Aeration or oxygenation systems are normally used to maintain the water at oxygen saturation. Feeding consists of extruded pellets, introduced manually or automatically. The elements that determine productivity are temperature and fry quality (survival and deformity rates). The optimum temperatures for feeding range from 14-18 C (Imsland et al., 1996, 2001), while the extreme range for the culture of turbot is 10-24 C (Imsland et al., 1996, 2001).

Fish stocking density can be up to 100 kg /$m^2$ (Danielssen and Hjertnes, 1991) but normal production density is around 40-60 kg/$m^2$.

The normal production cycle is based on hatching in June/July and weaning (when juveniles start being fed exclusively on dry feed) in August/September of the same year. During the second summer of on-growing, ambient temperatures become too high for optimal growth, the fish lose appetite, and are usually slaughtered. Farmed turbot can be marketed from about 0.7 kg (one year) to 3 kg (>2 years) or more, with larger fish commanding higher prices. Size demand has changed as formerly it ranged from 1.5-2.0 kg, but now smaller sizes are acceptable and range between 0.7 kg and 2.0 kg

Turbot are slaughtered in just few locations with the smaller producers having on-site slaughtering facilities. The sale of commercial size fish, packed live in ice is rare but can be done for restaurants and local markets . Fish are slaughtered all year but with a peak around Christmas when prices are higher.

After killing turbot are packed in polystyrene boxes, covered with a layer of ice and plastic film. In Spain, turbot are generally marketed whole and fresh, while in the rest of Europe they are generally gutted before sale. Spain has begun to produce filleted turbot to meet other European market demands.

The most common steps that occur in the killing process of turbot and in Europe are summarized in Figure 1. Stunning and killing methods that are not commercially used in the EU MS are described in Sections 4.1.3. (percussion) and 4.1.4. (electrical stunning). These methods were not included in the risk assessment.

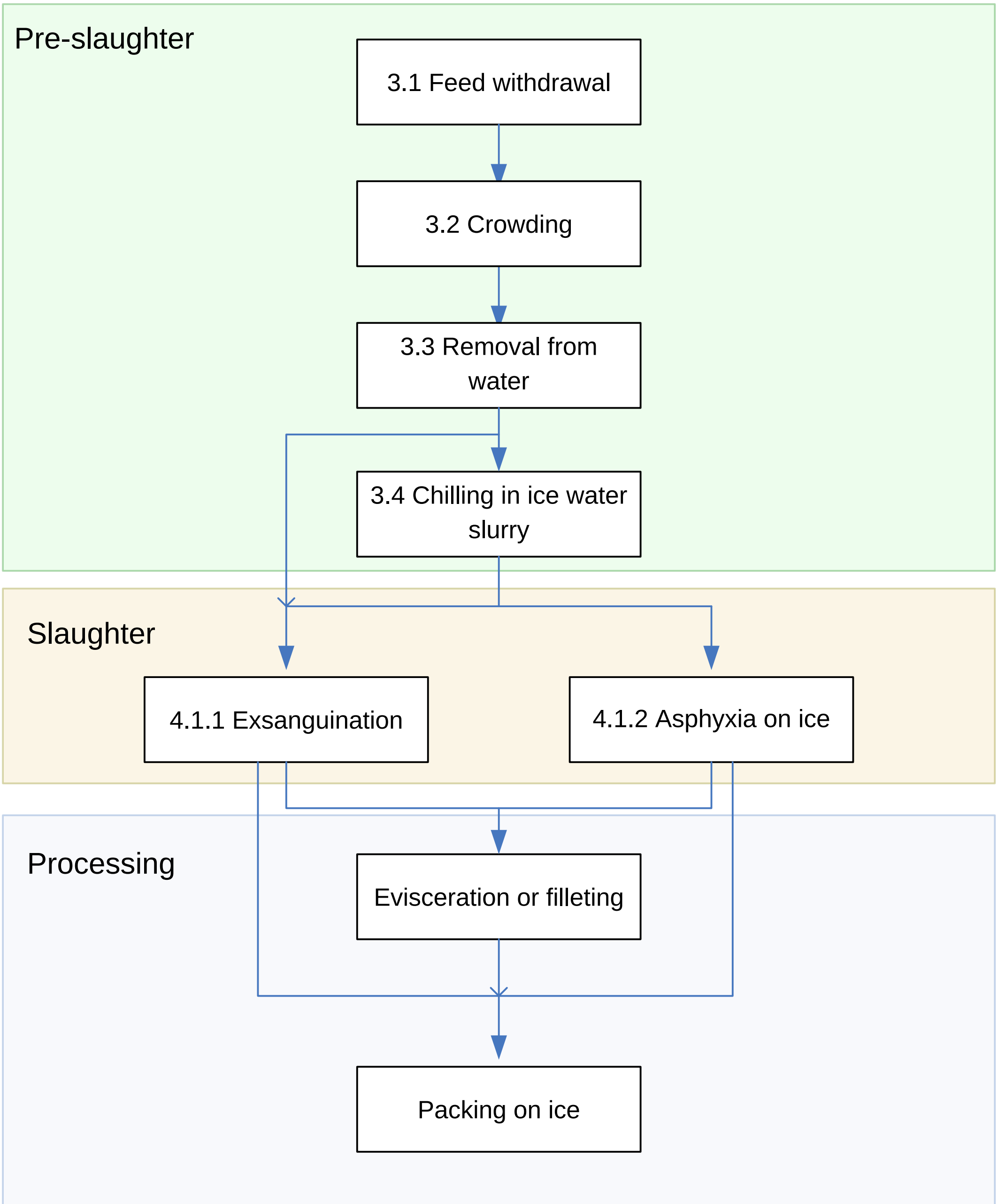



Figure 1**: Turbot pre-slaughter and slaughter process**

## 2. Pre-slaughter process

### 2.1. Pre slaughter feed withdrawal

Pre-slaughter feed withdrawal (3 to 5 days at average temperature of 15 C) to empty the gut is primarily carried out for food hygiene reasons and is not known to have a negative effect on the fish welfare.

Upon reaching slaughter weight (>1 kg), turbot intended for slaughter may be transferred by netting, pumping or sliding into a holding tank, and kept at identical rearing conditions but no feed is given until slaughter. Normally all turbot in the holding tank are slaughtered within a limited amount of time (one week).

## 2.2. Crowding

Turbot are reared at high densities in aquaculture (> 160 kg/m$^2$) (Danielssen and Hjertnes, 1991) and there is no real crowding during the initial transfer and they appear to show no behavioural signs of distress during the transfer from the rearing tank or raceway to a holding tank or slaughter facility. Social hierarchies are common in farmed turbot (Imsland et al., 1998) and the species preference is to lie on top of each other, with dominant ones preferring to remain at the bottom of the pile. Usually growth of subordinates is affected by dominants due to numbers, space or size of individuals, the relative size difference between members of the population usually increases. This is thought to be an adaptive strategy to optimise survival in a restricted space (Volpato and Fernandes, 1994). Turbot show a preference for high densities. Crowding was not considered as a risk for poor welfare in turbot culture.

## 2.3. Removal from water

Turbot are removed from their rearing or holding tanks to a slaughter facility and it is customary to transfer them in batches. The number of fish slaughtered in one day depends on the number of workers. The fish are hand netted (tank) or slid (raceways) out of the rearing tank to a transfer or holding tank. The hand netting is done on an individual or in batches (20 to 100 depending upon their weight) and the process is usually gentle. Since turbot are scale-less fish and have a thick epidermis, skin lesions very rarely occur due to hand netting. The exposure to air associated with the netting out of the tank (**hazard 1**) is very short and, in commercial conditions, is 10 sec or less. Although no observable behavioural reactions are caused by this procedure, some studies indicate that physiological stress responses occur (Staurnes 1994, 2001; van Ham et al., 2003, Roth et al., 2009, Waring et al. 1996).

## 2.4. Live chilling in ice water slurry

Turbot are commonly netted and placed in containers with a mixture of ice and sea water (0 to 4 C) at densities of approximately 300kg / m$^3$ and kept in these containers for a minimum of 30 min. The objective of chilling is to reduce muscle temperature and the metabolic activity in order to improve meat safety and quality.

When turbot are slaughtered by asphyxia they are kept in ice water slurry for approximately 30 min to 1h and subsequently the water is drained and the containers kept in cold chambers at 4 C until transported in refrigerated vehicles to the processing plant. When slaughtered by exsanguination,the waiting time in the containers will vary between 10 and 30 min.

In general, turbot show a low responsiveness to stress (van Ham et al., 2003) although there are indications that stress responsiveness is temperature dependent (van Ham et al., 2003). Turbot have the capacity to adapt to relatively low ambient oxygen concentrations (< 5.0 mg/L at 17 C) (Pichavant et al. 2000) without negative long term effects on normal growth and metabolism. However, due to the high density of fish in the transport containers, there is reason to believe that the water quality will deteriorate over time leading to a negative welfare effect in the short term (**hazard 2**). No skin colour changes in turbot are associated with lowering of dissolved oxygen in transport tanks but early onset of rigor mortis is observed after slaughter (Morzel et al. 2003, Roth et al. 2007). Acute stress responses lead to catecholamine release which will affect the chromatophores but turbot have the ability to suppress this physiological stress response as part of their camouflage/survival strategy (Bonga, 1997).

Signs of stress as determined by elevated plasma glucose and ionic disturbances ($Na^+$, $K^+$, $Ca^{++}$ and $Cl^-$) have been reported in turbot during simulated transport and live chilling (Staurnes, 1994, 2001; van Ham et al., 2003, Roth et al. 2009).

Low levels of dissolved oxygen (DO) may cause hyperventilation, resulting in an increase in arterial pH (respiratory alkalosis) in turbot, whereas high levels (above normal saturation) may cause hypo-ventilation and result in a marked drop in arterial pH (respiratory acidosis) (Foss et al., 2007). In general, fish are able to tolerate and rapidly compensate for short-term changes in ambient DO concentrations within well-defined species and life stage-specific limits. Beyond these limits, however, too high or too low oxygen concentrations may lead to acid-base and metabolic disturbances, indirectly causing reduced growth or even death (Claireaux and Dutil, 1992). The oxygen level below which juvenile turbot are reported to display depressed growth is 5.0 mg/L (Pichavant et al., 2000).

Poor mixing of ice water slurry can result in stratification of water quality and dead space volumes, particularly as flatfish have a tendency to remain inactive at the bottom (Reig et al., 2006) and the subsequent lack of mixing of tank water through fish movement. The formation of layers of different water quality within a tank is also dependent on the tank water renewal rate as higher flow rates promote mixing (Rasmussen et al., 2005).

In general, exposure to cold water of fish acclimated to higher temperatures leads to physiological disturbances which may cause death if they are excessive (Staurnes, 1994, Donaldson et al., 2008). If the exposure causes death ('primary chill coma'), this is normally as a result of ultimate respiratory failure because of disturbances in the respiratory centre in the central nervous system (Fry 1971; Staurnes 1994). Delayed death ('secondary chill coma') is usually the result of iono-osmoregulatory failure (Fry 1971) which in sea water manifests itself in the form of dehydration and an increase in body ion content (Lega et al., 1992; Staurnes, 1994, 2001).

When juvenile (70 g) turbot were transferred from water at 16 C to 1 C, they showed rapid, spasm-like response, characterized by quivering of the mouth, opercula and fins, and contraction of the dorsal muscles, indicating that the turbot experienced an initial cold shock. These responses were not seen if the juvenile turbot had been acclimated to lower temperatures (6-10 C) prior to chilling, but iono-osmoregulatory disturbances still occurred. Low temperature has also been found to impair the intestinal uptake of water in other fish species (Lega et al., 1992) resulting in dehydration and a fall in body moisture content and an increase in blood osmolarity and ion concentration. In the wild, adult turbot have a preference for cooler water temperatures compared with juveniles (Aneer and Westin, 1990; Iglesias and Rodríguez-Ojea, 1994) and data indicate a size-dependent drop in temperature optima for optimal growth and metabolism in turbot (Burel et al., 1996; Imsland et al., 1996; 2001).

Recent studies involving commercial size turbot showed that they do not exhibit secondary stress responses when chilled from 15 C to 1 C while responses were observed when turbot is exposed to -1 C suddenly (**hazard 3**).  This indicates that there is a certain temperature tolerance for turbot as reported for other species (Donaldson et al., 2008). However thermal nociceptors for cold have not yet been identified in fish (Ashley et al 2007).

Immobility and lack of response to painful stimuli have been interpreted in two different ways in turbot, either it signifies a state of unconsciousness or is due to cold shortening of muscles in a conscious fish. Cold shortening is explained by the seroplasmic reticulum membrane losing its ability to retain $Ca^{2+}$ and this, combined with the inhibition of both $Mg^{2+/}Ca^{2+}$ and $Na^{+}/Ca^{2+}$ ATPases, leads to the formation of actin and myosin bonds and muscle contraction (Ushio et al., 1991). When such contractions occur in the head and operculum muscles, some fish may display mouth gaping (Roth et al., 2009). The period of involuntary muscle contraction caused by live chilling will have a major negative impact on the welfare of turbot and may even be confused with rigor mortis (Roth et al., 2009).

## 3. Recognition of consciousness, unconsciousness and death

Stunning methods are expected to induce immediate (e.g. less than 1 second) unconsciousness. If loss of conscious does not occur immediately then induction of unconsciousness should occur without causing avoidable pain and distress. It is important for people involved in fish slaughtering operations to be able to recognise whether a stunning operation has rendered a fish immediately unconscious.

Turbot rely heavily on camouflage and immobility as defence mechanisms. Behavioural reactions to stressful situations are, therefore, not as evident as in other fish species. In turbot, field recognition for unconsciousness or death is made on the basis of absence of opercular movement, eyes fixed, and absence of a response to painful stimuli (pin-prick or touching the gill arch). In some processing plants turbot carcasses that have been chilled in ice water slurry are labelled by stapling a tag on the operculum, and the absence of response to this painful stimulus is used in the field to recognize death. However, lack of a response can also be due to immobilisation occurring as a result of chilling and so does not necessarily indicate death.

Other tests have been used experimentally to determine consciousness, unconsciousness and death such as ability to right itself and response to a 6V electric shock to the lips (Morzel et al., 2003).

Table 1: **Tests used to assess consciousness and unconsciousness of turbot. (Morzel et al. 2003)**

| **Test** | Breathing | Ability to right itself | Response to tail pinching | Response to needle scratch | Vestibulo - ocular reflex | Response to a 6V electric shock |
|---|---|---|---|---|---|---|
| **Protocol** | Observe the fish undisturbed | Place the fish with its blind side facing up | Grasp the tail firmly and drag the fish to the waterline while scratching its tail | Scratch firmly along the imaginary middle line of the right ocular side of the fish | Grasp the fish firmly and rotate it from a horizontal to a vertical position; the eyes must always face the observer | Apply the electrodes to the lips |
| **Location** | In water | In water | In water | In water | In air | In air, fish on a board |
| **Observation** | Movement (existence and rhythm) of the gill opercula | Ability or attempt to return to a natural position | Attempt to escape | Attempt to escape | Rolling of the eyes to compensate for changes in body posture | Eye retraction into the sockets |

In laboratory conditions, Electroencephalograms (EEGs) including visual evoked responses (VERs) and somatosensory evoked responses (SERs) may also be used (StunFishFirst 2005). Demonstration of the correlation between EEG data and physical responses to painful stimuli is lacking.

## 4. Specific stunning and killing systems

EFSA sent a questionnaire to all Member states enquiring about the methods in use for the slaughter of turbot. Chilling in ice water slurry was reported as the most common method (Questionnaire EFSA, 2009).

The methods used to kill turbot commercially are: 1. Exsanguination; and 2. Asphyxia on ice The later being the most common. Stunning is not practised in turbot only killing methods. Prototypes are available for electrical stunning in field conditions. Other methods such as percussion have been tested but they are not used on a commercial scale.

## 4.1. Killing methods used for turbot

### 4.1.1. Exsanguination

Turbot are exsanguinated straight after netting or after chilling in ice water slurry. Exsanguination is performed to remove the blood and for improving the visual quality of the meat (Roth et al., 2007a) as well as killing the animal prior to further processing such as evisceration and filleting. Turbot that have been previously chilled are taken out of the container in batches and placed on a table for exsanguination (**hazard 4**). Exsanguination is done by slicing the gill arches on one side **(hazard 5)** and normally 5 gill arches are cut but poor slicing (a blunt or an incorrectly positioned knife) may lead to inadequate exsanguination for death (**hazard 6).** There is a possibility that some turbot may not have their gills cut and remain alive until evisceration or packing in ice. After exsanguination, the turbot is left for 2 h or more to bleed-out in the ice water slurry prior to processing. It is likely that bleeding to death in a blood stained water is distressing to turbot. A study by Morzel et al. (2003) showed that behavioural responses in turbot were not lost within 90 min after exsanguination in water at 15 C (**hazard 7**). In the same study, it was shown that responses were lost earlier when turbot were exsanguinated in ice water slurry at 1 to 3 C. This raises the welfare concern that chilling might have inhibited any response to pain and distress caused by exsanguination. Roth et al. (2007) showed that exsanguinated turbot placed in ice water slurry showed escape behaviour and other responses to physical handling, and had to be killed by a percussive blow to the head 1h after exsanguination. During this period the muscle pH dropped from 7.2 to 6.8-6.9 indicating physical activity after exsanguination.

### 4.1.2. Asphyxia on ice

This method is the most commonly used method for killing turbot. Turbot are chilled in ice water slurry for approximately 30-60 min and subsequently the water is drained and the fish is killed by asphyxia on ice (**hazard 8**). In commercial practice, turbot are found to be dead in 4 h, which is the minimum time between the commencement of chilling and processing. Field experience indicates that turbot respond to painful stimuli before 4 h. The time to onset of death seems to be temperature dependent. Nevertheless, turbot lying at the bottom can be subjected to considerable pressure due to the weight of ice and fish until death occurs. This may cause pain and distress (**hazard 9**).

### 4.1.3. Electrical stunning

This method has been tested experimentally and needs evaluation under commercial conditions. Several studies have demonstrated that electrical stunning can stun fish unconscious within 1s (Lambooj et al., 2003, 2008; Robb and Roth, 2003). EEG recordings in turbot showed that a 5 sec electrical stun followed by chilling of the unconscious and insensible fish in ice water slurry for at least 15 minutes is sufficient to prevent recovery following stunning (StunFishFirst 2005). Several types of commercial electrical stunning equipment, both in water and dry stunners, are available but have not been evaluated for turbot. Studies show that turbot meat quality is not adversely affected by electrical stunning (Knowles et al., 2007; Morzel et al., 2003; Roth et al., 2007).

#### 4.1.4. Percussion

Experimentally percussion has been tested in turbot and is commonly used to stun/kill other flat fish species such as halibut. The results reported by Morzel et al. (2003) indicate an immediate loss of consciousness using an air gun. In commercial conditions, the method is man-power demanding as it can only be applied manually to turbot as the position of the brain in relation to the position of the eyes varies between individuals according to degree of rotation during metamorphosis, and so hitting the brain consistently is difficult. The destruction of the whole head is feasible but at the moment is not an alternative for commercial reasons since turbot is usually sold whole and the freshness of a fish is often evaluated by the appearance of the eyes. However, percussive equipment capable of stunning and killing turbot without causing these potential problems could be developed, according to the expert opinion.

## 5. Processing

### 5.1. Evisceration or filleting

Most turbot are sold as whole fish. Evisceration, when performed, involves removal of liver and intestinal tract and not the heart. The risk for eviscerating or filleting of conscious turbot (**hazard 10**) will depend on the time interval between the end of bleeding and the duration of chilling and the onset of the processing, and also on the temperature. There is no direct evidence for these time intervals however it is likely to be the same time as reported for killing by exsanguination, i.e. approximately 90min (Morzel et al., 2003). The normal commercial practice is to pack eviscerated turbot in ice. The time to onset of death due to asphyxia is dependent on the temperature and is reported to be prolonged at lower temperatures in trout (Kestin et al. 1991) but similar data are lacking for turbot.

## 6. Methods of stunning and killing for disease control

Emergency killing of fish is necessary in several circumstances. Moribund and diseased growing fish can require killing on production farms but this aspect has not been included. Fish can require culling on farms for disease control purposes and emergency killing of illegal imports may be required.

Methods listed are either in use or could be developed for mass killing on-farm.

Turbot is susceptible to Viral Haemorrhagic Septicaemia, (VHS), one of the listed diseases in Annex II CD 2006/88.

Emergency killing for disease control has occurred in the past and the method used was the same as commercial slaughter, which is asphyxia on ice.

The following methods could be used for killing for disease control purpose as they are used for other species but there are no standard operating procedures for the effective use of any of the methods. The choice of method will vary depending on the amount of fish being killed, and availability of facility and equipment. Such methods should be considered as part of contingency plans. Stunning should be carried out prior to killing. Signs of consciousness in fish should be monitored before disposal.

#### 6.1.1. Percussive stunning

For practical reasons this method cannot be applied to mass killing of fish.

#### 6.1.2. Overdose of anaesthetic

Better for welfare but the method results in carcasses not fit for human consumption.

#### 6.1.3. Electrical stunning

Described in section 4.1.3.

#### 6.1.4. Maceration

#### 6.1.5. Exsanguination

Described in section 4.1.1.

#### 6.1.6. Asphyxia on ice

Described in section 4.1.2

## 7. Reference to welfare indicators

Welfare indicators for turbot have not been satisfactorily assessed and validated so far. Nevertheless, observation of fish behaviour such as escape behaviour and flapping were taken into account in this opinion and may be used for field monitoring of welfare. Further validation of input and outcome measures is needed.

## 8. Risk Assessment

### 8.1. Risk assessment: discussion and results

#### *8.1.1. Application of the risk assessment approach to stunning and killing of turbot*

The Risk assessment method used to assess the risk to welfare of farmed fish at the time of killing is described in Appendix A.

The definitions of intensity and the categories for duration of the adverse effect used for the assessment of welfare risks on the killing of turbot are described in Tables 2 and 3.

Different definitions of severity for hazards that occurred pre-slaughter and those arising during stunning and slaughter were needed.

Table 2**: Intensity categories for adverse effects arising from hazards associated with pre-slaughter / slaughter for turbot**

| Evaluation | Score | Pre slaughter /Slaughter |
|---|---|---|
| **MILD**<br>The animal is minimally affected as evidenced by minor changes in behaviour and physiological stress responses.(a) | 1 | Behavioural reactions (such as escape behaviour and flapping) may or may not occur. |
| **MODERATE** | 2 | Not in the mild or severe category |
| **SEVERE**<br>The animal is affected greatly, as evidenced by marked changes from normal behaviour, physiological stress responses and the presence of physical injury.(a) | 3 | Escape behaviour, flapping, disturbed osmoregulation, changes in physiological stress indicators, |

Table 3**: Duration categories for adverse effects**

| Duration (minutes)[1] | Score |
|---|---|
| < 1 | 1 |
| 1 – 10 | 2 |
| >10 – 30 | 3 |
| > 30 | 4 |

### *8.1.2. Pre-slaughter hazards*

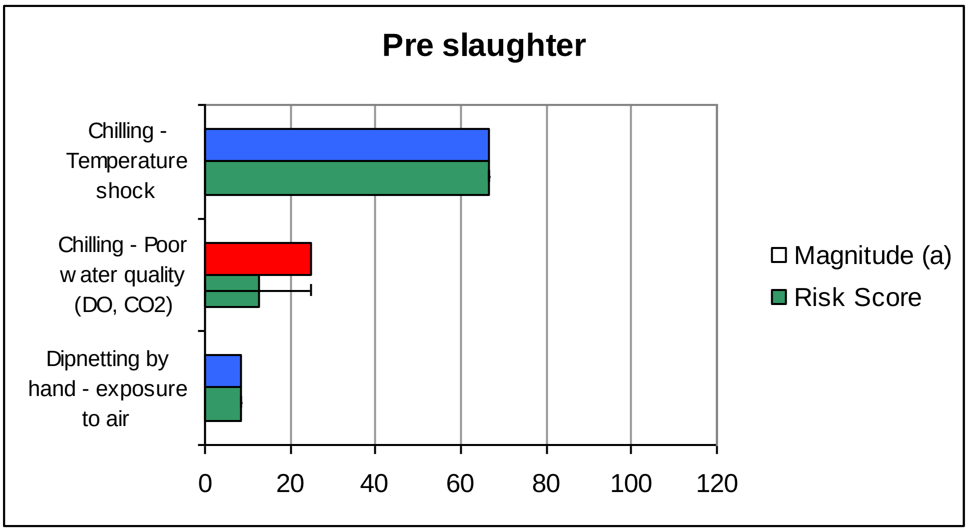
Three hazards were identified (see Table 4) (details in Appendix B) that may occur in pre-slaughter. Dip netting by hand was assessed as having a low magnitude of the adverse effect and 100 % exposure, all fish are exposed to air prior to slaughter. The adverse effects associated with deterioration of water quality were estimated as not severe but uncertainty regarding probability of exposure was very high. Temperature shock was the highest risk score hazard

Table 4: **Risk and magnitude scores for pre-slaughter hazards**

| | ***Hazard*** | **Description of the adverse effect** | ***Risk score*** | ***Magnitude*** |
|---|---|---|---|---|
| 1 | Exposure to air - Dip netting by hand | Distress, escape behaviour, flapping, changes in physiological stress indicators. | 8.3 | 8.3 |
| 2 | Live chilling - Poor water quality (DO, $CO_2$) | No behavioural signs, only measurable by physiological stress indicators and water quality parameters changes, early onset of rigor mortis. | 12.5 | 25.0 |
| 3 | Live chilling - Temperature shock | Escape behaviour, disturbed osmoregulation, changes in physiological stress indicators, cellular failure, cold shortening. | 66.7 | 66.7 |
| | | | **87.5 (a)** | |

(**a**) The sum of risk scores for the hazards of pre- slaughter

Figure 2: **Pre-slaughter hazards – risk and magnitude scores**




(a) The uncertainty associated to magnitude of the adverse effect was represented by: red =High, yellow=Medium and blue=Low

Hazards are ranked by risk score. Error bars show the estimated minimum and maximum values for the risk score, reflecting the uncertainty about the probability of exposure to the hazard. .

The uncertainty scores related to the hazard's magnitude were low for the hazard of exposure to air and high for the transport possible adverse effects since these are dependent on the duration of transport, density and external environmental conditions.

### *8.1.3. Slaughter hazards*

No stunning methods are commercially available at the moment. Two methods of killing were assessed: 1) Exsanguination and 2) Asphyxia on ice (details in Appendix B). Both methods may be followed by evisceration and filleting depending on the practices of each farm. If further processing is practised the risk associated to it was also estimated

For each method the risk scores for the hazards were summed (Table **5**). The risk scores range from 0.3 to 100. Asphyxia in ice and exsanguination while conscious had the highest risk scores since all fish killed by this method are exposed to the hazards. The magnitude of the adverse effect caused by these hazards was estimated as severe with low uncertainty, good data and consistent observations in commercial practice. Although the risk score is the same it is important to note that asphyxia in ice has a very long duration, turbot may remain alive when chilled for up to 2 days and the state of consciousness /unconsciousness has not been determined. The worst case scenario was assessed

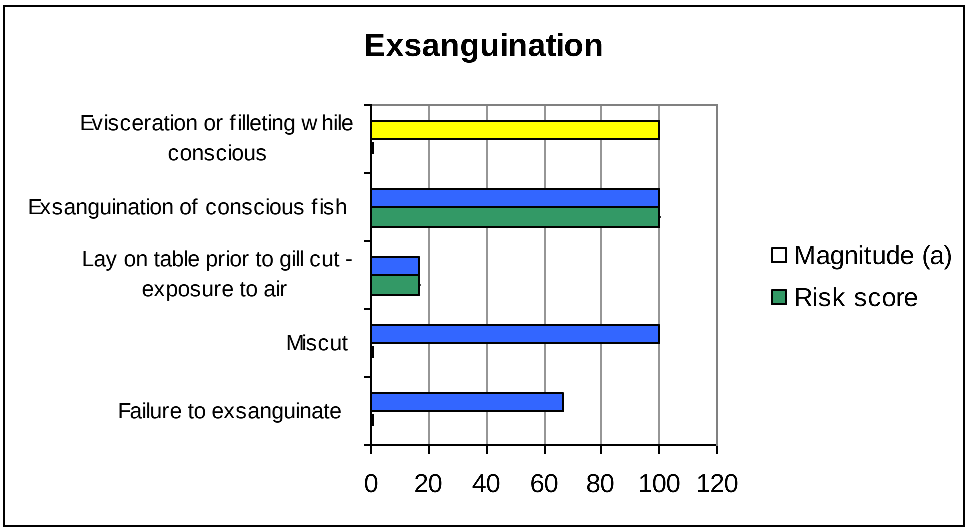
Evisceration while conscious, mis-cut, failure to exsanguinate are all hazards with a low risk score because probability of exposure is low, but very high magnitude since the welfare risk for the individual fish is very high. The experts had high uncertainty related to the estimates for probability of exposure to evisceration while conscious. Consciousness time after exsanguination is temperature dependent and no precise data exist on this issue.

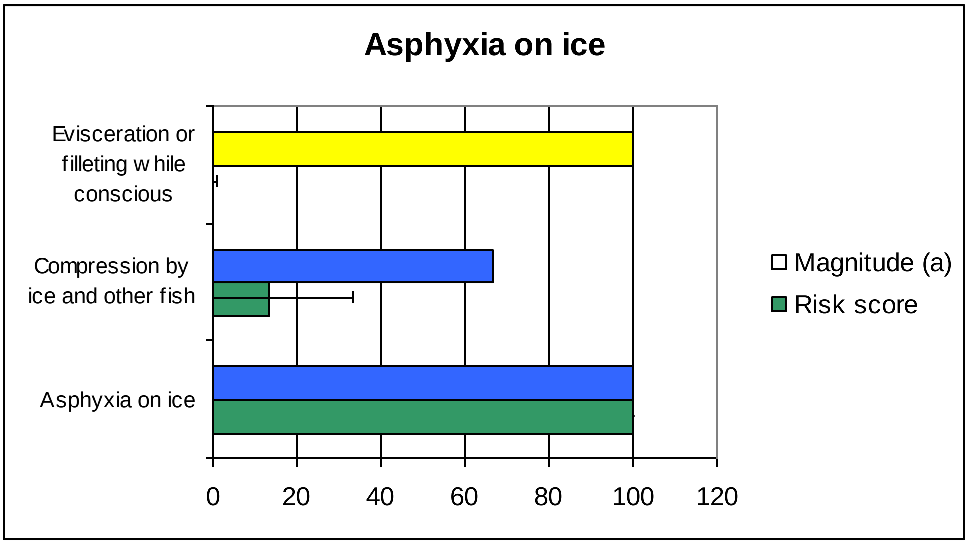
Table 5: **Risk and magnitude scores for the hazards associated with each slaughter method**

| | Slaughter method | Description of the adverse effect | Risk score | Magnitude |
|---|---|---|---|---|
| | **Exsanguination** | | | |
| 4 | Lay on table prior to gill cut - exposure to air | Distress, escape behaviour, flapping, changes in physiological stress indicators. | **16.7** | **16.7** |
| 5 | Exsanguination of conscious fish | Pain, escape behaviour, flapping, death due to asphyxia | **100** | **100** |
| 6 | Mis-cut | Pain, escape behaviour, increased probability of being eviscerated alive. | **0.5** | **100** |
| 7 | Failure to exsanguinate | Escape behaviour, splashing, some behaviours are inhibited by low temp. | **0.3** | **66.7** |
| 10 | Evisceration or filleting while conscious | Pain, trauma, flapping, death due to asphyxia. | **0.5** | **100** |
| | | | **118** | |
| | **Asphyxia on ice** | | | |
| 8 | Asphyxia on ice | Asphyxia, disturbed osmoregulation, dehydration, changes in physiological stress, cold shortening | **100** | **100** |
| 9 | Compression by other fish and ice | Pain, Distress | **13.3** | **66.7** |
| 10 | Evisceration or filleting while conscious | Pain, trauma, flapping, death due to asphyxia. | **0.5** | **100** |
| | | | **113.8 (a)** | |

**(a)** The sum of risk scores for the hazards of that slaughter / stun method;

**Figure 3: Magnitude and risk scores for stunning and slaughter methods**






(a) The uncertainty associated to magnitude of the adverse effect was represented by: red =High, yellow=Medium and blue=Low

Hazards were ranked by risk score. Error bars show the estimated minimum and maximum values for the risk score, reflecting the uncertainty about the probability of exposure to the hazard. In cases where the probability of exposure was equal to 1 there was no associated uncertainty (no error bars).
The uncertainty scores related to the hazards' magnitude were from one to three. Field observations are consistent and clear but there are often no published data.

### 8.1.4. *Overall comparison of methods of stunning and killing*

The total scores for the killing methods (i.e. summed pre-slaughter and slaughter hazards) are given in Table 6. The scores for the pre-slaughter hazards do not vary with the slaughter method. Evisceration is sometimes performed after killing with either of the two methods described.

**Table 6: Ranking of methods for turbot killing**

| Method | Exsanguination | Asphyxia on ice |
|---|---|---|
| Pre-slaughter score | 87.5 | |
| Slaughter score | 118 | 113.8 |
| **Total** | **205.5** | **201.3** |

## Conclusions

1. At present, turbot are not stunned prior to slaughter under commercial farming conditions.
2. The pre-slaughter procedure of chilling turbot in ice water slurry represents a welfare risk because it can cause cold shock in conscious fish which is known to cause distress due to involuntary muscle contractions. Live chilling is an immobilisation method and not a stunning method since it does not induce unconsciousness.
3. Turbot exposed to cold shock can suffer secondary stress responses with osmolarity and respiratory disturbances.
4. Water quality during pre-slaughter live chilling may have adverse effects on turbot welfare.
5. Existing methods of killing turbot, i.e. exsanguination and asphyxia on ice, involve prolonged periods of consciousness during which stress responses have been observed, and they constitute a considerable welfare risk.
6. Trials involving alternative methods, especially electrical stunning which induces immediate loss of consciousness followed by chilling in ice water slurry, have shown promising results for turbot welfare and meat quality.
7. Although turbot are susceptible to a notifiable disease Viral Haemorrhagic Septicaemia (VHS) specific operating procedures and detailed contingency plans are lacking.
8. Large scale killing methods developed for other species of fish can be applied under disease outbreak situations in turbot but they need to be evaluated in turbot and for the developmental stages.
9. At present there are no validated and robust indicators available to evaluate in practice the welfare of turbot associated with slaughter procedures.

## Recommendations

1. According to the farming system, location, species etc, appropriate pre-slaughter procedures and equipments should be identified.
2. As a matter of urgency, industry should be encouraged to test and develop commercially viable alternative methods such as electrical stunning followed by chilling or percussive methods, which induce an immediate loss of consciousness.
3. Standard operating procedures to improve the control of the slaughter process to prevent impaired welfare should be introduced.
4. For each species, practical welfare indicators relevant to stunning and slaughter should be developed.
5. A surveillance (monitoring) programme should be initiated for all the fish species so that data is available in the future for an improved risk assessment and for determining improvements over time and also for benchmarking for those involved in the slaughter of fish.

6. Humane methods of stunning or killing that do not cause avoidable pain or suffering should be developed for turbot.
7. Valid, robust and practically feasible indicators to evaluate the welfare of turbot during slaughter procedures need to be developed.
8. Persons involved in killing fish should be trained and hence skilled in handling and welfare.

**Recommendations for further research and development**

1. The temperature tolerance limits of turbot with regard to pre-slaughter live chilling are not clearly understood and need investigation.
2. Physiological stress responses to netting and removal from water need to be investigated further.
3. Relationship between the EEG data and behavioural responses to external stimuli needs to be established and appropriate welfare indicators developed.
4. The existence of nociceptors specific to temperature shock and pain are not established and needs investigation.
5. The critical oxygen levels and impact of water quality deterioration in live chilling tanks need further investigation.
6. The existing electrical and percussive stunning and killing methods need to be evaluated under commercial conditions in order to develop standard operating procedures.
7. This Scientific Opinion on turbot killing evaluated the methods currently used in farmed turbot in Europe. Methods used in other fish species other than those described in this Opinion may be applicable to turbot.

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## Glossary / Abbreviations

| | |
|---|---|
| Adverse effect | The welfare consequences for an animal in terms of pain and distress when exposed to a hazard. |
| Asphyxia | A process where fish die from hypoxia. This may happen in some species by: taking them out of water; by partially bleeding animals out; by preventing gill movements e.g. crushing; and by reducing oxygen content of the water. |
| Cold shock | Shock induced in muscles by exposure to low water temperatures to which the fish have not been acclimated. |
| Crowding | Keeping animals at stocking densities that are high or that reduce swimming volume e.g. by hoisting a net. |
| Demersal fish | Living near the bottom |
| Depopulation (Emergency killing for disease control) | A process of killing animals for public health, animal health, animal welfare or environmental reasons, sometimes under the supervision of the competent authority. |
| Dip-net | A net used to dip into a tank or cage to catch fish for the purpose of transfer of fish to another pond or facility or to market or for slaughter. |
| Duration | Specifically used with ‘intensity’ in the context of evaluating the magnitude of the adverse effect. |
| Emergency killing | The killing of animals that are injured or have a disease associated with severe pain or suffering and where there is no other practical possibility to alleviate this pain or suffering. |
| Exposure Assessment | The quantitative and qualitative evaluation of the likelihood of hazards to welfare occurring in a given fish population. |
| Mouth Gaping | Opening of mouth and operculum resulting from muscles contraction. |
| Hazard | Any factor with the potential to cause an adverse welfare effect on fish. |
| Hazard characterisation | The qualitative and quantitative evaluation of the nature of the adverse effects associated with the hazard. |

| | |
|---|---|
| Hazard Identification | The identification of any factor capable of causing adverse effects on fish welfare. |
| Hypercapnia | A condition with a raised level of carbon dioxide in blood. |
| Hyperoxia | A condition with oxygen saturation above 100% of the normal atmospheric equilibrium for a given temperature and salinity. |
| Hypoxia | A condition with low oxygen saturation in the water or a condition with low oxygen saturation in the water (blood). |
| Intensity | The quality of pain or distress per unit time |
| Involuntary muscle contraction | Muscle contractions occurring as a result of cold shock, which is equivalent to 'cramps' in humans with no control over their occurrence. |
| Killing | Any intentionally induced process that causes the death of an animal. |
| Magnitude of the adverse effects | A function of intensity and duration of welfare impairment for fish. |
| Percussive stunning | A blow in the head is applied with a club, less often with a spring-loaded or pneumatic device. |
| Pre-slaughter | Anything happening just before stunning, killing or slaughter. |
| Risk | A function of the probability of an adverse effect and the intensity of that effect, consequent to a hazard for fish. |
| Risk Assessment | A scientifically based process consisting of the following steps: i) hazard identification, ii) hazard characterisation, iii) exposure assessment and iv) risk characterisation. |
| Risk Characterisation | The process of determining the qualitative or quantitative estimation, including attendant uncertainties, of the probability of occurrence and severity of known or potential adverse effects on welfare in a given fish population based on hazard identification, hazard characterisation, and exposure assessment. |
| Severity | Sometimes used to denote intensity. |
| Slaughter | The killing of animals for human consumption. |
| Slaughterhouse | Any establishment used for slaughtering fish. |

| | |
|---|---|
| Starvation | A period of food deprivation such that the animal metabolises tissues that are not food reserves but are functional tissues. |
| Stocking density: | Number of fish in a defined volume of water. |
| Stunning | Any intentionally induced process that causes loss of consciousness and sensibility without pain, including any process resulting in instantaneous death. |
| Uncertainty Analysis | Uncertainty refers to the extent to which data are supported by published evidence. A method used to estimate the uncertainty associated with model inputs, assumptions and structure/form. This includes also uncertainty, due to the lack of reliable publications, uncertainty in the scientific results etc. |
| Variability | The natural biological variation that occurs in a population of animals. Not to be confused with uncertainty as it cannot be reduced by simply decreasing uncertainty. |
| Vestibulo-ocular reflex (VOR) | A reflex where eye movement occurs in a conscious fish when rocked from side to side (commonly called eye roll). |
| Visual evoked reflexes (VER) | Evoked EEG activity in the brain with a visual stimulus. |

# Appendices

## APPENDIX A

## RISK ASSESSMENT APPROACH

### Introduction

Overall the risk assessment was constrained due to limited scientific data and consequently a semi-quantitative assessment was carried out often based on expert opinion. Because of this lack of data, the Panel on Animal Health and Welfare recommends that a surveillance / monitoring programme should be initiated for all the fish species so that in the future it may be possible to carry out a quantitative risk assessment.

In this section, the risk assessment method used to assess the risk to welfare of farmed fish at the time of killing is described.

Risk assessment is a systematic, scientifically based process to estimate the probability of exposure to a hazard, and the magnitude of the effects (consequences) of that exposure. A hazard in animal welfare risk assessment may be defined as a factor with the potential to cause a negative animal welfare effect (adverse effect). Risk is a function of both the probability that the hazard and the consequences (characterised by the adverse effect) occur.

Three parameters were scored to assess the importance of a hazard; the intensity of the adverse effect that the hazard causes, the duration of the adverse effect and the probability of exposure to the hazard. The population in question is the fish killed in the EU by the selected method of stunning and slaughter.

The probability of exposure to the hazard corresponds to the percentage of all fish exposed to the hazard. Thus if 4% of the all the fish killed by a particular method are exposed to a hazard there is a probability of 0.04 that any randomly selected fish within that population is exposed. The consequence of exposure can be assessed by scoring the intensity and the duration of the adverse effect in the individual. The risk assessment was based on two assumptions;

1. all fish exposed to the hazard experienced the same intensity and duration of the adverse effect.
2. in the absence of any evidence to the contrary, it is assumed that all fish exposed to the hazard experience the adverse effect[6].

Factors which adversely affect fish welfare are considered in the risk assessment. In absence of reliable data, the volume of fish slaughtered by each method is not taken into account. Thus the results are not weighted by the volume of fish slaughtered by each method.

The definitions of intensity and the categories for duration of the adverse effect used for the fish species considered in this scientific opinion are in the relevant section in each Scientific Opinion.

[6] if this assumption was not found to be sound for a particular hazard an additional parameter (probability that exposure resulted in the adverse effect) was used.

In the following paragraphs the risk assessment process for hazard identification and characterization and the probability of exposure to the hazard are described as well as the way they were scored. Finally the risk scoring process is described.

The general risk assessment is in line with the approach previously used in the EFSA welfare reports (EFSA, 2007a; EFSA, 2007b; EFSA 2007c; EFSA, 2008a; EFSA, 2008b; EFSA, 2008c; EFSA, 2008d; EFSA, 2008e) with some modifications according to the risk question posed.

**Hazard identification**

The objective of the hazard identification is to identify potential welfare hazards associated with each stunning and killing method. The identification was based on a review of the literature and field observations. The scope of the risk assessment included the period leading up to killing (which may be the time spent in lairage for fish killed in a slaughterhouse). The adverse effect caused by each hazard is described. In order to consistently identify hazards associated with stunning and killing, the relationship between the time from applying a stun method, unconsciousness and the point at which the killing method was applied are illustrated graphically (Figure 1). Various scenarios (A to E) in which hazards may arise were identified as follows:

'A' where a fish is killed in some potentially painful way (asphyxia, bleeding out) while it is conscious i.e. before it has been made unconscious; and

'B' represents a fish that has been stunned and is killed or it dies after it is unconscious;

'C' where a fish has been stunned but it recovers consciousness and is killed in some potentially painful way (asphyxia, bleeding out).

'D' represents a fish that, like A is killed in some potentially painful way (asphyxia, bleeding out) while it is conscious but has also suffered from the aversive nature of the stunning method; and

'E' represents a fish that has been stunned and is killed or it dies after it is unconscious but has also suffered from the aversive nature of the stunning method.

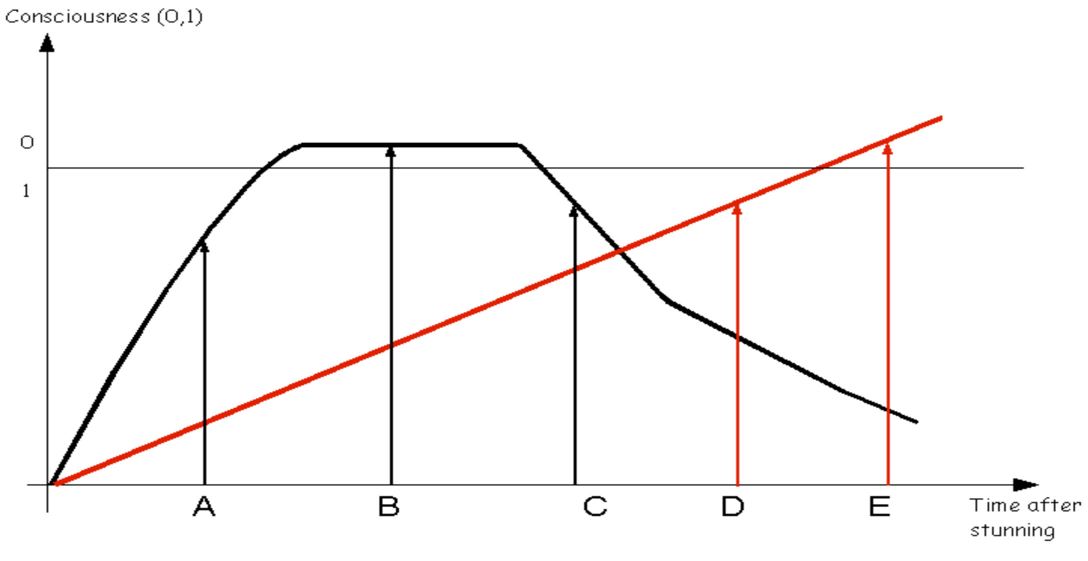


**Figure 1. Time to unconsciousness (insensibility) following stunning / killing** (horizontal grey line indicates consciousness threshold above which killing takes place without an adverse effect).

The scenarios above do not take into account hazards arising from gathering animals during pre-slaughter or killing without stunning.

## Hazard characterisation

### Intensity

If a fish is unconscious, by definition there is no adverse welfare effect at that time. Therefore, before assessing the intensity of any adverse effects, consideration must be given as to whether the fish is conscious or not; this is a binary judgement (i.e. degrees of un/consciousness are not assessed). There is evidence that signs associated with consciousness and unconsciousness at the time of killing apply to all fish species as they do for general anaesthesia (Kestin et al., 2002). If it is conscious, the appropriate score for the degree of intensity of the adverse effect must be selected: mild, moderate or severe. If unconsciousness is achieved or induced with no suffering, or any pain or distress is for less than one second, then it is assumed that there was no welfare hazard. The issue of consciousness is mainly relevant to hazards associated with the killing method. If unconsciousness was achieved immediately (less than one second) then it is assumed that there was no hazard associated with the proper and effective application of that method and so this was not included in the risk assessment.

Generic guidelines for defining intensity categories for pre-slaughter hazards and slaughter hazards are given in Table 1. The approach taken has been to define only the mild and severe categories; the moderate is defined as being neither mild nor severe. Thus, by default hazards which are considered to have welfare consequences which are not in the severe or mild category fall into the moderate category. This approach was taken as scientists are reasonably confident in recognising the extreme states of intensity but as these states are on a continuum, allocating a distinct moderate banding is more difficult and contentious. Appropriate descriptions for the categories of intensity will vary between species and are given for each species in the Scientific Opinion.

Additionally, different definitions of intensity for the same species may be required for hazards that occur before killing, compared with at the time of killing. The descriptions of intensity for these pre-slaughter adverse effects are given for each species in the Scientific Opinion.

Table 7: **Observable signs considered by experts when scoring the intensity of an adverse effect in farmed fish arising from hazards associated with the pre-slaughter or slaughter period**

| Evaluation | Score | Description |
|---|---|---|
| Mild | 1 | The animal is minimally affected as evidenced by minor changes in behaviour (e.g. rapid swimming away from stimulus and then slowing down, eye position normal). |
| Moderate | 2 | The animal is affected as evidenced by behaviour changes which can be considered moderate (more pronounced than minor but not severe). |
| Severe | 3 | The animal is affected greatly, as evidenced by marked changes from normal behaviour (e.g. energetic and purposeful escape behaviour, eyes rolling, rapid and erratic swimming, swimming upside down or tilted, colliding with the net, stopping swimming for more than 5 secs, crowding of fish) |

Finally, each hazard was assessed and ranked by magnitude and occurrence independently of other hazards. For some hazards there may be more than one adverse effect. For example, all fish netted will be exposed to air, but in addition they may be injured e.g. skin lesions due to contact with the net or other fish.

**The duration of the adverse effect**

The time during which an animal will on average experience the adverse effect was estimated in minutes. The duration of an adverse effect can be longer than the duration of the hazard, for example a mis-stun takes a fraction of a second but the adverse effect lasts until the animal is unconscious or dies. Thus the duration of the hazard is included in the duration of the adverse effect.

Different time periods may be used for the adverse effects arising from pre-slaughter hazards compared with the hazards associated with slaughter. The definitions of duration used are given in the relevant section of the Scientific Opinion (Table 2 and 3).

## Exposure assessment

The exposure assessment is performed by assessing the proportion of the population of interest (i.e all fish in the EU being killed by the method in question) that is likely to experience the hazard. This proportion is equal to the probability of exposure to the hazard (P_hazard). It is recognised that the proportion of the population exposed to a selected hazard will vary depending on the farm of origin and slaughterhouse. Estimates of the most likely, maximum and minimum values for this proportion are required. The range of values provides an indication of the uncertainty of the estimate (see next section).

## Uncertainty and variability

The degree of confidence in the final estimation of risk depends on the uncertainty and variability (Vose, 2000). Uncertainty arises from incomplete knowledge and/or when results are extrapolated from one situation to another (e.g. from experimental to field situations) (Vose, 2000). Uncertainty can be reduced by carrying out further studies to obtain the necessary data, however this may not always be a practical possibility. It can also be appraised by using expert opinion or by simply making a judgment.

Variability is a statistical and biological phenomenon and is not reducible by gathering further information. The frequency and severity of welfare hazards will inevitably vary between farms and countries and over time, and fish will vary individually in their responses. However, it is not always easy to separate variability from uncertainty. Uncertainty combined with variability is generally referred to as total uncertainty (Vose, 2000).

Total uncertainty associated exposure to the hazard was captured by estimates of the maximum and minimum estimates of the most likely value of the proportion of the population exposed to the hazard. For the other parameters (intensity and duration of the adverse effect) total uncertainty was scored on a scale of 1-3 (Table 8).

Table 8: **Scoring system for total uncertainty in intensity and duration of effect**

| Evaluation | Score | Description |
|---|---|---|
| low | 1 | Solid and complete data available; strong evidence in multiple references with most authors coming to the same conclusions, or<br>Considerable and consistent experience from field observations. |
| medium | 2 | Some or only incomplete data available; evidence provided in small number of references; authors' or experts' conclusions vary, or<br>Limited evidence from field observations, or<br>Solid and complete data available from other species which can be extrapolated to the species being considered |
| high | 3 | Scarce or no data available; evidence provided in unpublished reports, or<br>Few observations and personal communications, and/or<br>Authors' or experts' conclusions vary considerably |

**Risk Characterisation**

**The scoring process**

The scoring was undertaken by the working group in plenary. The estimates were based on current scientific knowledge, published data, field observation and experience (as summarised in this report).

**Calculation of the risk score**

All three factors (probability of exposure to the hazard; intensity of adverse effect; duration of adverse effect), were included in calculating the final risk score of a hazard. The score for each parameter was standardised by dividing the score by the maximum possible score for that parameter. Thus all parameters have a maximum value of one. The risk score is the product of the standardised scores multiplied by 100 (for ease of comparison) and thus has a maximum value of 100.

Risk score = [*(I_adverse_effect /3)* * *(D_adverse_effect / 4)* * *(P_hazard)*] * 100

Where the following are defined:

the intensity of the adverse effect *(I_adverse_effect)*

the duration of the adverse effect *(D_adverse_effect)*

the probability of exposure to the hazard *(P_hazard)*

The minimum, most likely and maximum values for *P_hazard* were used to generate minimum, most likely and maximum estimates of the risk score. If only one risk score is given it refers to the most likely. It is also assumed that hazards usually occur independently of each other.

**Calculation of magnitude of adverse effect**

The magnitude of the adverse effect is the product of the scores for intensity and duration according to the following formula:

Magnitude score = [(*I_adverse_effect* /3) * (*D_adverse_effect* / 4)] * 100

It has a maximum score of 100. The magnitude provides an indication of the impact of the hazard on the fish which are exposed to the hazard and experience the adverse effect. Thus a hazard that causes a prolonged and severe adverse effect but which affects only a small proportion of the population will have a low risk score but a high magnitude of severity score**.**

### Worked example – mis-stun

Mis-stun may result when a concussive stunning method is used. This will give rise to an adverse effect. It was estimated that the adverse effect had a intensity score equal to 3. The duration (time from mis-stun to death or re-stun) was judged to last between one and two minutes, hence a score of 3. It was estimated that the probability that the hazard occurs was 0.04 (i.e. 4% of fish suffer a mis-stun), with minimum and maximum estimates of 0.01 and 0.10, respectively. In summary:

- score for the intensity of the adverse effect (*I_adverse_effect*) = 3
- score for the duration of the adverse effect (*D_adverse_effect*) = 3 (between one and two minutes)
- the probability that the hazard occurs (*P_hazard*) = 0.04 (ranging from a minimum estimate of 0.01 to a maximum estimate of 0.10)

Thus the risk score for this example mis-stun is:

(3/3 * 3/4 *0.04) * 100 = (1 * 0.75 * 0.04) * 100 = 3

This score has a range that is determined by the minimum and maximum estimates of the probability that the hazard occurs (*P_hazard*), 0.01 and 0.10 respectively.

Minimum score = (3/3 * 3/4 *0.01) * 100 = 0.75

Maximum score = (3/3 * 3/4 *0.1) * 100 = 7.50

The magnitude equals intensity score/3 * duration score/4 * 100; and in this example is 75:

(3/3 * 3/4) *100 = 75

### Interpretation of the risk score

Due to the limited amount of quantitative data on many effects of hazards on fish stunning and killing, the risk assessment was mainly based on expert opinion. The methodology used does not give a precise numerical estimate of the risk attributed to certain hazards; however the output can be used to rank the problems and designate areas of concern, as well as, guidance for future research. The methodology does not take into account interactions between factors and assumes linearity in the scores. These assumptions cannot be tested. Secondly, the risk scoring is semi-quantitative. Thus the scores allow a ranking but the absolute figures are not on a linear scale (e.g. a risk score of 12 should not interpreted as being twice as important as a risk score of 6).

One key objective of this work is to compare different methods of stunning and slaughter within each species. This will be achieved by summing the risk scores for all the hazards arising for each method of stunning and slaughter. This figure will be used to rank and compare the methods. Risk scores are given for the commonly used methods (see Table 6). However, it should be noted that insufficient data were available to calculate the overall exposure to the hazard within the European population, i.e. how commonly are those methods actually used within the member states of the EU. For comparison purposes, this calculation is important as it quantifies more precisely the number of fish at risk for that particular method of slaughter. Moreover, a hazard with a small risk score but a high magnitude may still have serious welfare effects for a large number of fish. The converse is also true.

# APPENDIX B

| Hazard ID | | Description of adverse effect | Severity of the adverse effect *score* 1 mild, 2 moderate, 3 severe | Duration of the adverse effect minutes | Duration Score of the adverse effect *score* 1 = <1min, 2 = 1-10min, 3 = 10-30min, 4 = >30min | Uncertainty | Probability of the exposure to the hazard | | | Probability 3d that exposure to the hazard leads to the adverse effect | | | Risk score | | | Magnitude |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | **most likely** | min | max | **most likely** | min | max | **most likely** | min | max | |
| **1** | Dipnetting by hand - exposure to air | Distress, escape behaviour, flapping, relative changes in physiological stress | 1 | 10 sec | 1 | 1 | **1** | 1 | 1 | 1 | 1 | 1 | **8.33** | 8.33 | 8.33 | 8.33 |
| **2** | Transport - Poor water quality | No behavioural signs, only measurable by physiological stress indicators and water quality parameters changes, early onset of rigor mortis. | 1 | 30 | 3 | 3 | **0.5** | 0 | 1 | 1 | 1 | 1 | **12.50** | 0.00 | 25.00 | 25.00 |
| | | | | | | | | | | | | | | | | |
| **3** | Chilling - Temperature shock | Escape behaviour, disturbed osmoregulation, relative changes in physiological stress indicators, celular faillure, cold | 2 | 40 | 4 | 3 | **1** | 1 | 1 | 1 | 1 | 1 | **66.67** | 66.67 | 66.67 | 66.67 |
| **4** | Asphyxia on ice | osmoregulation, dehydration, relative changes in | 3 | 120 | 4 | 1 | **1** | 1 | 1 | 1 | 1 | 1 | **100.00** | 100.00 | 100.00 | 100.00 |
| **5** | Crushing by other fish and ice | Distress, pain. | 2 | 120 | 4 | 2 | **0.2** | 0 | 0.5 | | | | **13.33** | 0.00 | 33.33 | 66.67 |
| | | | | | | | | | | | | | | | | |
| **6** | Lay on table prior to gill cut - exposure to air | behaviour, flapping, relative changes in physiological stress | 1 | 0.5 | 2 | 1 | **1** | 1 | 1 | 1 | 1 | 1 | **16.67** | 16.67 | 16.67 | 16.67 |
| **7** | Exsanguination of conscious fish | Pain, escape behaviour, flapping, death due to asphyxia | 3 | 30 | 4 | 1 | **1** | 1 | 1 | 1 | 1 | 1 | **100.00** | 100.00 | 100.00 | 100.00 |
| **8** | Miscut | Pain, escape behaviour, increased | 3 | 40 | 4 | 1 | **0.005** | 0 | 0.01 | 1 | 1 | 1 | **0.50** | 0.00 | 1.00 | 100.00 |
| **9** | Failure to exsanguinate | splashing, some behaviours are | 2 | 90 | 4 | 2 | **0.005** | 0 | 0.01 | 1 | 1 | 1 | **0.33** | 0.00 | 0.67 | 66.67 |
| **10** | Evisceration or filleting while conscious | Pain, trauma, flapping, death due to asphyxia. | 3 | 60 | 4 | 1 | **0.005** | 0 | 0.01 | 1 | 1 | 1 | **0.50** | 0.00 | 1.00 | 100.00 |