ABSTRACT

In European Union Member States, most of the biomass of *Trichinella* parasites is circulating among wildlife and many human infections originate from the consumption of untested game meat. Annually, hundreds of millions of fattening pigs test negative for *Trichinella* in the European Union. The preliminary harmonised monitoring scheme proposed in this report relies on compartmentalisation to identify regions and categories of animals at lower risk of *Trichinella* infection in which reduced testing could be carried out. The scheme proposes the introduction of an additional monitoring region, a low risk region, that does not exist under current European Union Regulations. Member States or their regions are categorised into three region groups based on the degree of confidence that *Trichinella* can be considered absent in fattening pigs. Within these three regions certain animal populations are monitored with different intensity. Animal populations destined for human consumption and requiring continuous testing for *Trichinella* are: sows and boar, horses, hunted wild boar and other susceptible wildlife for human consumption. Reduced testing would apply to fattening pigs, from low risk or negligible regions. In the proposed scheme, monitoring of wildlife not intended for human consumption would be carried out in regions with negligible risk. The detection method of choice for all animal species is the artificial digest method but the necessity of its use in combination with quality controls is highlighted.
SUMMARY

In European Union Member States, most of the biomass of *Trichinella* parasites is circulating among wildlife (both carnivores and wild boar). Consequently, many human infections originate directly from the sylvatic cycle following the consumption of untested game meat consumed by hunters and their families. Leaving animal carcasses in the field after skinning, or removing and discarding the entrails, a practice employed by hunters and often referred to in literature, is a contributing factor of transmission to new animal hosts. Human perturbations of the sylvatic environment may also affect the epidemiological patterns of human and animal infection with *Trichinella*. Changes in agricultural practice including, for example: set-aside, use of wider field margins and conservation efforts to increase biodiversity in managed land and creation of national parks, may have been associated with increased numbers of animal species that can act as hosts for *Trichinella* such as the feral wild boar and the red fox and with an increase in the biomass of *Trichinella*.

*Trichinella* in domestic pigs still occurs in 13 Member States, most frequently in backyard and free-ranging pigs. There is also evidence that *Trichinella* is currently circulating in large-scale industrial pig farms in Romania. This cycle occurs where high-risk farming practices can be found, such as the intentional feeding of food waste, potentially containing pork scraps, or unintentional exposure to carcasses of dead swine or infected wildlife, usually by unsecured free-range pasturing. Annually, hundreds of millions of fattening pigs test negative for *Trichinella* in the European Union, including fattening pigs from holdings which have not yet been officially recognised as *Trichinella*-free. Backyard and true free-ranging pigs on the other hand are often not tested for *Trichinella*, as pigs slaughtered for private domestic consumption are not required by European Union legislation to undergo meat inspection. Such animals that do not reach the market (estimated at a few millions per year) can include infected animals and are often the source of human infection. This is despite the fact that several Member States require national legislation on testing or having voluntary testing schemes and consumer education. The presence of the parasite in domestic and/or wild animals alone does not necessarily lead to infection in humans. *Trichinella* spp. infections in humans are related to cultural food practices, such as the consumption of dishes containing raw or undercooked meat.

These epidemiological facts, the expenses incurred by the mandatory testing and the desire to accelerate the inspection process of pigs in large slaughterhouses, suggest that more attention and resources should be focused on the high-risk populations that still harbour *Trichinella* parasites and less resources should be spent on the hundreds of millions of fattening pigs from modern holdings, that represent a negligible risk to human health.

The monitoring scheme proposed in this document relies on ‘compartmentalisation’ to identify regions and categories of animals at lower risk of *Trichinella* in which reduced testing could be carried out without compromising public health.

This scheme, which is otherwise mostly in line with current European Community legislation proposes the introduction of an additional monitoring group, low risk region, that does not exist under current European Community Regulations. European Union Member States or regions are categorised into three groups ('regions') based on the confidence that *Trichinella* can be considered absent in fattening pigs above a specified design prevalence. Within these three regions certain animal populations are monitored with different intensity. Animal populations destined for human consumption and requiring continuous testing at slaughter are: sows and boar (from controlled and non-controlled housing), horses, hunted wild boar and other susceptible wildlife (susceptible carnivores and omnivores). Reduced testing would apply to fattening pigs, from low risk or negligible regions. In the proposed scheme, the monitoring of wildlife (susceptible carnivores and omnivores) not intended for human
consumption would have to be carried out in regions with negligible risk in all fattening pigs. The proposed surveillance schemes are explained in detail in Objective 4. ('Propose harmonised monitoring and reporting schemes') and examples for calculating surveillance sensitivities are provided in Appendix B.

This scheme is a framework, which would allow countries that have gathered sufficient evidence to fall into the low risk or negligible region groups and then continue to provide the evidence as proposed. The framework is also developed to facilitate Member States that cannot fall back on years of historic data and cannot confidently demonstrate their situation to be able to gather sufficient evidence relatively quickly to reduce the numbers of low risk pigs for testing. In our view, if this is carried out reliably, public health would not be compromised but would have large economic benefits and free resources that could be applied to controls of meat from animals slaughtered for own consumption or for direct supply to the consumers. We recommend that this be included in official controls to reduce the number of human cases acquired via this route.

The detection method of choice for all animal species is still the artificial digestion method, presently considered the gold standard, which cannot yet be replaced by alternative methods. However, it is important to highlight the need for these methods to be used in combination with quality controls (e.g. specific training and ring trials).
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BACKGROUND

In the Community Summary Report (CSR) on zoonoses (EFSA, 2006a), the information received from the Member States (MSs) is analysed and summarised specifically to identify trends in the occurrence of zoonotic agents and sources of human infections. As there are currently no harmonised rules or recommendations for reporting and monitoring Echinococcus spp., Trichinella spp., Cysticercus spp. and Sarcocystis spp. in the European Union (EU), the data obtained is often difficult to analyse and interpret.

EFSA’s Scientific Panels on Biological Hazards and on Animal Health and Welfare issued an opinion on the Review of the Community Summary Report on Zoonoses, Zoonotic Agents and Antimicrobial Resistance in the European Union in 2004 (EFSA, 2006b). In this opinion the panels concluded among other things: parasites (Toxoplasma gondii, Echinococcus spp., Trichinella spp. and Taenia spp./Cysticercus spp.) have been reported less frequently in humans, and have caused fewer outbreaks, than bacteria and viruses in the EU in 2004. However, in many instances the impact of these zoonotic agents (severe illness, disability, death and costs related to diagnostic procedures, hospitalisation and treatment) on vulnerable groups of the population, and often in immunocompromised persons, has probably been considerable.

The panels also stated that there is a need for a common strategy on data collection, monitoring and reporting as well as an improvement of harmonisation of definitions, in order to improve the usefulness of the data presented in the CSR.

TERMS OF REFERENCE

The objective of the call is to obtain proposals for projects, which will develop harmonised monitoring and reporting schemes for Trichinella spp., in animals and, when appropriate, in foodstuffs under the Directive 2003/99/EC (EC, 2003). The schemes shall be applicable in all EU MSs.

These schemes shall, in particular, specify:

- the animal species and/or foodstuffs, which should be monitored and the study populations (subgroups of the population) to be targeted. The animal species may cover farm animals, pet animals, zoo animals and wildlife;
- the stage when the sampling should take place (e.g. at farm, at slaughterhouse);
- sample size (the number of samples to be collected) and the procedure how to select the samples;
- the type of specimen to be taken and sampling techniques;
- the diagnostic and analytical methods to be used;
- the information to be collected at national level; and
- the information to be reported to the Commission and EFSA.

The rationale for the specifications chosen in the monitoring and reporting schemes must be given. When developing the schemes, it is advisable to take into account public health needs, the feasibility and cost-effectiveness of the schemes as well as different MS situations.

The schemes shall also include suggestions for the analyses of data at national and Community levels, and, in particular, indicate where the following of trends over the reporting years would be useful.
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INTRODUCTION

Directive 2003/99/EC (EC, 2003) of the European Parliament and of the Council of 17 November 2003 on the monitoring of zoonoses and zoonotic agents forms the basis for data on zoonoses being collected throughout the MSs and reported to the EU on an annual basis. These data are collected and examined by the European Food Safety Authority (EFSA), who, in collaboration with the European Centre for Disease Control (ECDC) and assisted by the Zoonoses Collaboration Centre (ZCC), produce an annual report, the Community Summary Report on Trends and Sources of Zoonoses, Zoonotic Agents, Antimicrobial Resistance and Foodborne Outbreaks in the European Union, which is then published in the EFSA Journal. The report is aimed at the detection of sources and trends within the EU MSs and to aide the long-term goal of protecting human health.

Trichinella is included in list A of Annex I, Directive 2003/99/EC (EC, 2003), which determines the agents that have to be monitored on a mandatory basis. Official testing is carried out according to Regulation (EC) No 2075/2005 (EC, 2005b) of 5 December 2005 laying down specific rules on official controls for Trichinella in meat, which covers most of the relevant animal species.

The objective of this report is to develop a harmonised scheme for monitoring and reporting Trichinella in defined animal populations in the EU. The results from the application of such a harmonised scheme should create data that would enable comparison of infection status between MSs and identification of trends at national and Community levels. Public health needs, the feasibility of schemes and the different MS situations will be taken into account.

The overall objective was broken down into several milestones. The first milestone was to review the current disease situation and national monitoring in the MSs. The rationale behind this was to identify public health needs in the MSs, and to create a basis for formulating the sampling plans. Each species of Trichinella was assessed with respect to its relevance to public health, impact on human health and epidemiology. A list of animals and foodstuffs was created for the relevant agents and their suitability within monitoring schemes was assessed. Analytical methods are one of the limiting factors in surveillance. Existing analytical methods were summarised and assessed regarding their feasibility in sampling schemes for use throughout the EU.

The milestones/objectives, approach, underlying rationale and results are described in detail hereafter.
OBJECTIVES

Objective 1. Identify current disease situation in Member States and current national level of monitoring and reporting information

1.1 Rationale

In the call for proposals it is specified that harmonised schemes should consider different situations in MSs and the schemes should be designed to be applicable to all EU MSs. Consideration should also be paid to testing schemes currently carried out in MSs. The table was designed to gather data needed to assess public health needs, the current testing situation and to define epidemiological parameters.

1.2 Approach

A spreadsheet for data and information collection was designed and circulated to the MSs using established contacts, national competent authorities and networks within the project team (network of national reference laboratories for parasites). The spreadsheet asked for information on confirmed human cases and the current disease situation in relevant animal populations, as well as for supporting information on sampling and testing carried out in the MSs, as a basis for formulating monitoring schemes. Where answers were not received a literature search was carried out in order to fill the gaps. A summary table of the responses was compiled and can be found in Appendix H. The current situation in the different MSs is summarised in the result section.

1.3 Results

A vast amount of literature on Trichinella has been published since its discovery in 1835 and a comprehensive summary of the worldwide distribution of Trichinella has been published recently (Pozio, 2007). These sources of information, which are constantly being updated, have been used to complete and/or complement data from questionnaires.

Current situation of Trichinella infections in humans in the EU

No autochthonous infections in humans have been documented in Austria, Belgium, Cyprus, the Czech Republic, Denmark, Finland, Greece, Ireland, Luxembourg, Malta, the Netherlands, Portugal, Slovenia, Sweden and the United Kingdom in the last 30 to 40 years or more. The lack of infection is mainly because of two reasons: national food habits and/or the low national prevalence of the infection in animals. In Bulgaria, Estonia, France, Germany, Hungary, Italy, Latvia, Lithuania, Poland, Romania, Slovakia and Spain, autochthonous trichinellosis in humans occurs, although the reported prevalence varies and this is attributed to the consumption of undercooked or untested raw meat originating from wild boar and/or from backyard and free-ranging pigs. In Romania, the prevalence of human trichinellosis is markedly higher due to the additional exposure to raw meat from infected pigs on “industrial” farms (Blaga et al, 2007). In addition, human infections also occur in some MSs because of the consumption of imported animals or animal products from other MSs or from third countries. Excluding Romania, where a large number of human infections still occur (350 cases reported to EFSA in 2006 and 432 in 2007), less than 300 cases are reported in EU MSs annually.
The variation in the prevalence of human cases can be attributed to several factors. In the first instance, for an infection to happen, the agent needs to be present in the animal, reaching the food chain. However, prevalence in the animal population cannot be directly linked to prevalence of human cases, as this also depends on the existence as well as the level of veterinary controls, aimed at the prevention of infected meat entering the human food chain. Finally, believed to be the most important factor, human behaviour i.e. the consumption of raw or undercooked infected meat, determines the occurrence in humans.

Current situation of *Trichinella* infections in domestic and wild animals in EU MSs

*Trichinella* occurs in wild animals throughout EU MSs with the exception of Malta, Cyprus and Great Britain, and prevalence varies depending on the geographical region, host and agent species. The occurrence of *Trichinella* in livestock is very dependent on husbandry practices i.e. the level of on-farm biosecurity measures and therefore a distinction between 'industrial farms', which generally operate under controlled housing conditions, and non-controlled housing conditions, such as backyard and free-range farms, is considered useful. The only country where *Trichinella* still occurs in 'industrialised' holdings is Romania. Single sporadic cases related to only single detected larvae found in industrialised pigs were reported from some EU MSs, but the parasites have never been identified by sequencing at species level. In Bulgaria, Estonia, Finland, France, Germany, Hungary, Italy, Latvia, Lithuania, Poland, Portugal, Romania, Slovakia, Spain and Sweden *Trichinella* has been reported from backyard or free-ranging pigs in the last 10 years, though in some of these countries occurrence is rare (see country information below).

No infected domestic pigs reared in Austria have been detected in the last 24 years. Both *T. spiralis* and *T. britovi* infections occur in wild boar and red foxes.

In Belgium, parasites have never been documented in domestic pigs. Between 12,000 and 15,000 wild boar are tested annually by digestion and in November 2004, larvae of *T. britovi* were detected in a pooled wild boar sample.

In Bulgaria, *T. spiralis* and *T. britovi* are widespread in wildlife and in free-roaming and backyard pigs.

No *Trichinella* infection has been documented in Cyprus and Malta and the habitat characteristics suggest that these two islands could be considered as not having *Trichinella*, but an accidental importation cannot be excluded as recently documented in two other Mediterranean islands (Sardinia and Corsica).

In the Czech Republic, no infected pigs have been detected in the last 50 years (Pozio, 2007). *T. britovi* was documented in red foxes and wild boar.

In Denmark, no infection with *Trichinella* has been documented in pigs since 1930. In 1996-1997, a prevalence of 0.1% was detected in red foxes from a small area of Jutland. In 2007, *T. pseudospiralis* was detected in two minks from the island of Bornholm.

In Estonia, there is a high prevalence of infection of *T. nativa* and *T. britovi* in wildlife; however, infection in domestic pigs is rare.

In Finland, the domestic cycle was endemic up to 2004 and the condemnation rate of pigs was around 0.0001%. A high prevalence of *T. spiralis*, *T. nativa*, *T. britovi*, and *T. pseudospiralis* was detected in wild animals.
In France and Germany, only the sylvatic cycle currently exists. Sporadic infections have been documented in free-ranging and backyard pigs, which are all believed to have originated from the sylvatic cycle.

In Greece, Trichinella infections were documented more than 20 years ago in wild animals and some backyard and free-ranging pigs, but a human outbreak occurred following consumption of a free-roaming pig in northern Greece in 2009.

In Hungary, the sylvatic cycle occurs among red foxes and wild boar. A T. spiralis focus in backyard pigs is active near the border with Romania.

In Ireland, T. spiralis infections occur only in red foxes. No infection has been documented in domestic pigs in the last 38 years.

In Italy, only the sylvatic cycle (T. britovi) occurs among wildlife and the parasite is seldom transmitted to backyard or free-ranging pigs.

In Latvia, Lithuania and Poland, domestic and sylvatic cycles are still present with a high prevalence of the infection in wildlife.

No findings of Trichinella were reported over the last years from Luxembourg.

In the Netherlands, the sylvatic cycle (T. spiralis, T. britovi and T. pseudospiralis) has been documented with a low prevalence in the red fox and wild boar populations. Trichinella spp. infection has not occurred in the Dutch pig population since 1979.

In Portugal, the sylvatic cycle (T. britovi) occurs in carnivores of very few regions; whereas, a single backyard pig was found infected in 1966.

Romania is the EU country with the highest prevalence of Trichinella infections in domestic pigs. The average prevalence of Trichinella in home-slaughtered pigs (typically backyard pigs) is 8.9% and 7.7% in abattoir-slaughtered pigs originating from large industrial production units (Blaga et al., 2009). These parasites are also widespread among wildlife.

In Slovakia, only the sylvatic cycle occurs in red foxes, whereas infections in free-ranging and backyard pigs seldom occur.

In Slovenia only the sylvatic cycle has been occasionally documented; no infection has been documented in domestic pigs in the last 50 years.

In Spain, T. spiralis and T. britovi are highly prevalent in wildlife; infections also occur quite frequently in backyard and free-ranging pigs.

In Sweden, only the sylvatic cycle occurs and infections in domestic pigs have only been reported up to 1995.

In the United Kingdom, Great Britain is believed not to have Trichinella since no infection has been documented in red foxes in the last 50 years or in domestic pigs since 1977; whereas in Northern Ireland, T. spiralis was detected in a red fox in 2007 and in one red fox in 2009 but has not been detected in a domestic pig since 1979.
Objective 2. Identify animal species and/or foodstuffs which could be affected and specify which should be monitored

2.1 Identify parasite species to be monitored

2.1.1 Rationale

In the Call for Proposals (CFP/EFSA/Zoonoses/2007/01); in the Report of the Task Force on Zoonoses Data Collection, in the Manual for Reporting on Zoonoses, Zoonotic Agents, Antimicrobial Resistance and Food-borne Outbreaks in the framework of Directive 2003/99/EC and on some other pathogenic microbial agents for information derived from the reporting year 2006 (EFSA, 2007); and in the Community Summary Reports on Trends and Sources of Zoonoses, Zoonotic Agents, Antimicrobial resistance and Foodborne outbreaks in the European Union in 2004 and 2005 (EFSA, 2005 and 2006a), *Trichinella* is either referred to as *Trichinella* spp. or it is not further specified. We considered it important to clarify first which species are relevant in the context of public health, i.e. which species are zoonotic and to address the consequent public health burden. The impact on human health needs to be considered for assessing the feasibility and public health benefit of sampling schemes especially in the light of the economic impact that those sampling schemes will pose to individual MSs. We consider that monitoring efforts should be commensurate with the public health risk that is posed and proportionate with the resources dedicated to the protection of public health from all food-borne zoonoses.

2.1.2 Approach

Literature (scientific publications, textbooks, official websites (OIE/WHO/ECDC)) on *Trichinella* was reviewed and the information/existing knowledge on zoonotic species summarised. The identified species were run through a number of criteria, listed below, and their zoonotic potential assessed. A summary of the results can be found in the spreadsheet 'Trichinella Zoonotic species RA' in Appendix E.

The species were considered under the following criteria:

Criterion 1: Zoonotic (Y/N).

For the purpose of this project, species, which had not been reported in literature as zoonotic, were not intended to be taken further through the qualitative risk assessment. However, some of the *Trichinella* species and/or genotypes have not yet been confirmed as being infectious to humans. These species were marked as 'Human cases not yet documented, but potentially considered zoonotic' and included in the risk assessment.

Criterion 2: Pathogenicity (+ - ++++)

The clinical symptoms of trichinellosis in humans generally are dose and species dependent and show the same characteristics for mild, moderate and severe infections and differ mostly in their strength/intensity. The classical symptoms are gastrointestinal signs (diarrhoea, nausea, vomiting), fever, myalgia and periorbital oedema. 'Complications' (myocarditis, encephalitis and thromboembolic diseases) are more likely, but not exclusively, expected to occur in severe cases and are the main cause of fatalities. Chronic disease is still under debate but the persistence of larvae in humans who did not receive treatment, or to whom treatment was given late, has been reported to cause symptoms such as impaired muscle strength, coordination, and conjunctivitis, for up to 10 years (Dupouy-Camet et al., 2002; Dupouy-Camet and Bruschi, 2007).
Criterion 3: Geographical criteria

Some Trichinella species occur in geographically confined areas, where they are adapted to certain climatic conditions and/or the availability of certain host species. Due to the 'human factor' (e.g. illegal import of exotic meat/insufficient official controls/inappropriate freezing treatment), the introduction of species currently not circulating in EU MSs is theoretically possible. Here it was assessed how easy the establishment of such species would be in the EU, depending on the likelihood of introduction, similarities to the known host species and comparable suitable climatic conditions.

Criterion 4: Economic impact and related disease burden

For assessing the economic impact of human clinical disease, the costs of diagnostic procedures, treatment costs and/or number of sick days, and long-term effects were considered. Again, this was carried out on a qualitative scale, to give a rough guideline and justification of monitoring schemes.

2.1.3 Results

Human infections caused by Trichinella T8, T9, T12 and T. zimbabwensis have not yet been reported. Nevertheless, all Trichinella species are considered zoonotic because they are phylogenetically very close and because of their broad host spectrum among mammals (Dupouy-Camet et al., 2002; Pozio et al., 2009a). It is suspected that human infections with these parasites have not been reported yet because they circulate among wildlife of remote areas with sparse human populations.

Humans are generally considered highly susceptible to infections with Trichinella spp., though it is important to differentiate between infection and clinical disease. Clinical symptoms range from asymptomatic to fatal and vary over time. The severity of symptoms is directly correlated with the number of infective larvae ingested and also influenced by the Trichinella species. If and why different species seem to cause differences in the severity of symptoms is not fully understood at this point. It is suspected that the number of new born larvae produced by the female may be the main contributing factor, though the published data on the reproductive capacity index of Trichinella spp. (RCI) are based on data derived from experimental animals rather than humans. It is thought that immunity and individual susceptibility in the human host also play an important role (Dupouy-Camet and Bruschi, 2007). Current studies on dose response models are being carried out to gain new insights into the transmission risk for humans and first results indicate that the infective dose may be lower than presumed (Takumi et al., 2009).

The mortality rate (<0.2%) in humans is based on a survey that was carried out by the International Commission on Trichinellosis (ICT) between 1995 and 1997. Fatalities are rare and mostly associated with complications as mentioned above (Dupouy-Camet et al., 2002).
Economic impact: diagnostic procedures for identifying trichinellosis in humans are generally not considered expensive, neither are treatments. Hospitalisation is generally limited to cases with complications and long-term effects rarely reported. However, severe cases do occur, they require intensive care and can lead to death. Uncommonly, sequels of neurological or cardiac complications could lead to some partial and definitive disabilities. Recommendations have been made that the use of DALYs (Disability-Adjusted Life Years), a quantitative measure, might lead to more comprehensive analyses and interpretation at EU level (Opinion of the SPBH and the SPAHW, 2006).

Currently circulating in Europe are: *T. spiralis*, *T. britovi*, *T. nativa* and *T. pseudospiralis*, which are the species considered relevant to be monitored. However, since *T. nativa* circulates only among carnivores living in cold regions, the importance of this species for humans is very limited and only relevant to people that consume game meat from bears and other carnivores from those regions, hence it is recommended that the monitoring of this particular species be limited to those particular areas. Other species have not yet been found in Europe and the potential for establishment is considered low because of a lack of suitable host species and/or climatic requirements. Meat of animal species imported from third countries could contain *Trichinella* species not present in Europe. These products fall under import regulations and are required to be tested before entering the EU (Regulation (EC) No 2075/2005 (EC, 2005b), Article 13).

However, when discussing the *Trichinella* species relevant to be monitored, it needs to be stressed that for this particular parasite at this point, this is only of theoretical relevance. Analytical methods (artificial digests) used for monitoring meat/muscle tissue will automatically detect any *Trichinella* larvae, regardless of the species. Species determination, as required according to Regulation (EC) No 2075/2005 (EC, 2005b), Article 6, will automatically identify any existing species, which should be reported as this provides important epidemiological information.

### 2.2 Identify relevant animal species and/or foodstuffs to be monitored

#### 2.2.1 Rationale

Parasite species are often reported in a wide variety of hosts, not all of which necessarily play a role in the transmission of the infection, have an impact on the human food chain or are suitable for surveillance in a public health context. The aim here was to identify which species would be suitable for surveillance in all MSs and consideration was given to existing surveillance carried out in MSs.

#### 2.2.2 Approach

A table was compiled with animal species in which the zoonotic agent has been reported. The animal species were then assessed as to their role in the epidemiological chain and the human food chain. A summary of the results can be found in the spreadsheet ’Relevant animals and foodstuffs to be monitored RA’ in Appendix F.
2.2.3 Results

In most EU MSs, with the exception of Romania, no *Trichinella* infections have been documented in pigs from indoor farms, where animals are reared according to modern standards. Most *Trichinella* infections in domestic animals occur in backyard and outdoor husbandry systems, in which livestock has direct or indirect (via vectors e.g. rodents) contact with wildlife and/or are caused by illegal human feeding practices (feeding of non- or improperly treated swill). Transmission risk from wildlife to farms depends mostly on farm practices, the contact between pigs and wildlife (direct and indirect) and the prevalence of *Trichinella* in wildlife. Therefore, the significance of wildlife testing for protection of public health varies from area to area. Backyard and free-ranging pigs and wild boar have a higher likelihood of coming into contact with wildlife and thus of being infested with *Trichinella*. Sows and boar in commercial systems, due to their greater longevity, are also more likely to be infected than fattening pigs. The testing of these groups at slaughter for human consumption can provide useful information, describing the circulation of these parasites and the parasite species circulating in Europe. Consequently, for our proposed monitoring schemes, we divided the livestock populations into different risk categories.

*Trichinella* spp. is circulating in wildlife in all MSs, with the exception of Cyprus, Malta, and Great Britain, though accidental introduction could occur in all of those areas. The role that wildlife plays in the epidemiology of *Trichinella* varies depending on the aetiological agent(s) circulating in MSs and on livestock rearing practices. Consequently, the role of wildlife in monitoring schemes varies within areas as to its importance in protecting human health. This needs to be addressed and justified according to its suitability for a defined area.

Sows and boar

Sows and boar generally have a longer life span compared to fattening pigs and generally hold a higher social status in the group, which means they will have preferential and greater access to food (Copado et al., 2004). Due to this cumulative effect (long life span and high social position in the pig group), sows and boar can act as a reservoir of *Trichinella* spp. and are considered a high-risk population that should always be tested (Pozio et al., 2009c).

Fattening pigs

The intensity with which fattening pigs should be monitored for *Trichinella* depends on the husbandry type and rearing practices i.e. controlled vs. non-controlled housing. On 1 January 2008, legislation (Regulation (EC) No 853/2004 (EC, 2004a) of the European Parliament and of the Council of 29 April 2004 laying down specific hygiene rules for food of animal origin) came into force requiring slaughterhouse operators to ‘request, receive, check and act upon’ food chain information (FCI) for all pigs sent to the slaughterhouse. Within this framework it is mandatory to provide information about production systems, and of particular importance is information about outdoor production and production under controlled housing. The term ‘controlled housing’ is further described in Regulation (EC) No 2075/2005 (EC, 2005b) and Regulation (EC) No 1244/2007 (EC, 2007) of 24 October 2007 regarding implementing measures for certain products of animal origin intended for human consumption and laying down specific rules on official controls for the inspection of meat, but leaves discussion points about risk assessments regarding outdoor access and classification of piglets/pigs that have been moved from outdoor to controlled housing systems.
Horses

Since outbreaks during the period 1975 to 2004 (Liciardi et al., 2009), linked to the consumption of Trichinella infected horsemeat and involving a large number of people, horses destined for human consumption have to be tested for the parasite according to Regulation (EC) No 2075/2005 (EC, 2005). Raw horsemeat is still consumed in some EU MSs. Because of their long life span and the difficulties involved in tracing the origin of horses reaching the food chain, it is recommended that under no circumstances horses reaching the human food chain should be exempt from Trichinella testing. Whereas the results of these tests are of limited epidemiological value, they are extremely important for protecting public health.

Wildlife and meat species other than domestic pigs and horses

Wildlife monitoring can contribute directly to the protection of human health but also provides useful information on the circulation and epidemiology of the parasite species. However, the driving force behind wildlife monitoring in an EC legislation framework is considered the protection of human health rather than scientific interest, on which the monitoring schemes proposed in this report are based.

Currently wildlife monitoring in MSs is carried out in the following contexts:

a) direct protection of human health when wildlife enters the food chain;
b) regions where the risk of Trichinella in domestic swine is officially recognised as negligible (according to Article 3, paragraph 2 (ii), Regulation (EC) No 2075/2005 (EC, 2005b));
c) for officially recognised Trichinella-free holdings that permit outdoor access to piglets during the first weeks of life before weaning (Regulation (EC) No 2075/2005 (EC, 2005b));
d) for officially recognised Trichinella-free holdings in MSs, where Trichinella has been detected in domestic swine in the last 10 years (Regulation (EC) No 2075/2005 (EC, 2005b)), if: susceptible wildlife and holdings in that area coexist; and
e) for research purposes.

The wildlife surveillance system applied in these settings are purpose-designed and are summarised below.

There is no need to monitor Trichinella in foodstuffs other than carcasses.
Table 1. Summary of wildlife sampling

<table>
<thead>
<tr>
<th>Context</th>
<th>Sampling schemes</th>
<th>Area</th>
<th>Monitored animals</th>
<th>Aim</th>
</tr>
</thead>
<tbody>
<tr>
<td>Meat inspection</td>
<td>Wildlife destined for human consumption, according to 2075/2005. Sampling as and when required</td>
<td>Not specified</td>
<td>All wildlife for human consumption</td>
<td>Protection of human health</td>
</tr>
<tr>
<td>Negligible region</td>
<td>Risk-based or equivalent</td>
<td>Not specified</td>
<td>Most suitable indicator animal</td>
<td>Demonstration of low prevalence (not defined in legislation)</td>
</tr>
<tr>
<td>Trichinella-free holdings that permit outdoor access to piglets</td>
<td>Annual, risk-based, epidemiologically related to the geographical location of holdings</td>
<td>Indicator animals, present in that area which can be followed on an annual basis (historic data)</td>
<td>Detection of Trichinella in indicator animals of &lt;0.5%</td>
<td></td>
</tr>
<tr>
<td>Trichinella-free holdings in MSs, where Trichinella has been detected in domestic swine in the last 10 years</td>
<td>If: susceptible wildlife and holdings in that area coexist</td>
<td>Risk-based, in areas where wildlife and holdings applying for Trich-free status co-exist</td>
<td>Most suitable indicator species</td>
<td>Not specified</td>
</tr>
<tr>
<td>Research</td>
<td>Individually designed to include species and area of interest to the research carried out.</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

2.3 Conclusion

None of the currently existing wildlife sampling schemes provides suitable data for monitoring and trend analysis at Community level as sampling involves different wildlife species, sampling protocols, timeframes and purposes. However, despite the lack of a harmonised wildlife sampling scheme in Europe, reporting of the situation in wildlife is important and results can be valuable for epidemiological investigations of food-borne outbreaks or for disease management. At this point there is not sufficient public health justification for recommending a harmonised wildlife sampling scheme throughout EU MSs. To be able to continue maximising the use of data that are being collected for different purposes in MSs, we recommend that these data be recorded centrally and reported to EFSA and to the Community Reference Laboratory for parasites with information on the animal species, the origin/collection points of the animals (GIS coordinates data or nearest towns), test results (positive and negative results, Trichinella species, infection level) and test sensitivities (see analytical tests). The data can be compiled in maps and can be updated over time, which would reflect the actual situation more accurately than existing approaches. Details on which samples to use and the tests can be found in ‘Objective 3: identify most suitable diagnostic and analytical methods to be used’.
**Farmed wild boar**

All farmed wild boar (destined for the human food chain/placed on the market) should be tested for *Trichinella* at slaughter. Similar to horses, farmed wild boar can have an extended lifespan and the origin of the animals may be similarly difficult to trace back. However, if farmed wild boar are reared under the same conditions as fattening pigs from non-controlled housing and meet the same criteria for traceability, consideration could be given to move them into this category. This would have only practical relevance in negligible regions.

**Feral wild boar, hunted**

All hunted wild boar submitted to a game handling establishment/placed on the market, directly supplied to a consumer or a restaurant or consumed in a private setting, should be tested for *Trichinella*. On average, free-roaming wild boar have a longer lifespan compared to fattening pigs and a wider home range, which allows them more contact with wildlife or other potential sources of infection over a longer period. In Europe, wild boar is a good indicator of the circulation of *T. spiralis* and *T. pseudospiralis*; it is less important as an indicator of the circulation of *T. britovi*; and is not significant for the epidemiological cycle of *T. nativa*.

**Foxes & racoon dogs**

The framework in which wildlife monitoring is important has been outlined in Table 1.

Whereas foxes are not important in the context of meat inspection, they are often used as indicator animals in monitoring programmes or for epidemiological studies. Generally foxes are present and abundant across EU MSs. They hold a high position in the animal food chain. Foxes are considered as a very good indicator species, especially for *T. britovi*. *T. spiralis* can also be detected in foxes, though in regions where wild boar is present; wild boar should be considered the preferred indicator for *T. spiralis*. However, the *Trichinella* species that foxes harbour seems to vary within MSs and the relevance to the domestic cycle does not seem to be comparable in each MS. Most (about 90%) *Trichinella* infected carnivores harboured *T. britovi* and most (about 80%) *Trichinella* infected domestic pigs harboured *T. spiralis* (Malakauskas et al., 2007; Pozio et al., 2009b; Széll et al., 2008).

Raccoon dogs are considered as suitable indicator hosts for *Trichinella* as are foxes due to their continuous spread throughout MSs. At present, consistent populations of this animal are present in Finland, Estonia, Latvia, Lithuania, Poland, and Germany. Furthermore, this animal species has also been detected in France, Italy, Switzerland, the Netherlands and Belgium and it is expected that the current area of distribution will expand further in the near future. Since this host species is more widespread in north-eastern countries than elsewhere in the EU the total percentage of the four different species is strongly influenced by its distribution. Today, of 71 *Trichinella* isolates from raccoon dogs in the EU, 14% were *T. spiralis*, 24% *T nativa*, 56% *T. britovi* and about 6% *T. pseudospiralis* ([www.iss.it/site/Trichinella/index.asp](http://www.iss.it/site/Trichinella/index.asp)).

**Other susceptible wildlife apart from foxes, racoon dogs and feral wild boar**

All wildlife susceptible to *Trichinella* and consumed by humans should be sampled and tested for direct protection of human health. Results from all studies carried out on any species should be reported as described above. Methods are specified in the following section.
Objective 3. Identify most suitable diagnostic and analytical methods to be used

3.1 Approach

Existing analytical methods, as cited in publications or official methods, were compiled and test validity (sensitivity, specificity), was listed as far as available. Also considered were the expenditure and complexity of test methods. The costs were estimated roughly, where possible, bearing in mind that they vary from country to country and depend on the daily throughput in a diagnostic facility. A summary of the results can be found in the spreadsheet 'Summary of analytical methods' in Appendix G.

3.2 Rationale

For most agents more than one detection method exists, applicable to different sample materials and producing results that often vary from method to method. These methods were compiled to identify the limitations of what can be achieved diagnostically, to compare the cost benefits of various methods and to assess practical aspects. Not every test can be used for every sample type. However, if two different methods produce the same result, e.g. measuring national prevalence to a certain level, the results of both methods could be directly compared. A cost estimate was also included as this is an important criterion when recommending analytical methods.

3.3 Results

For official controls of *Trichinella* in meat, the magnetic stirrer method for pooled sample digestion is considered the gold standard. Officially accepted variations on this method are being described in Regulation (EC) No 2075/2005 (EC, 2005b), Annex III. These methods are most suitable for the digestion of domestic swine, horse and wild boar meat. An overview of these methods taken from Webster et al., 2006, can be found in Table 2. Other animal species intended for human consumption and details of the examination procedures are also covered within this regulation (Regulation (EC) No 2075/2005 (EC, 2005b), Annex III).

‘Other wildlife’ (e.g. foxes, raccoon dogs, mustelids), are often tested to assess the prevalence of *Trichinella* in reservoir/indicator animals. It is generally recommended to amend the digest method depending on the animal species to be tested, to optimise its performance. A number of studies on this subject have been carried out (Gamble et al., 2000; Kapel, 2000; Kapel et al., 2005; Nöckler et al., 2007). Table 3 summarises the predilection sites in various animal species with regard to the *Trichinella* species. However, heterogeneity in the distribution of larvae in contaminated muscle tissues does exist and may affect the reliability of monitoring. An overview on recommendations on the length of digestion depending on the animal species and muscle type can be found in Table 4.

Generally, variations in the digestion methods can be considered acceptable, as long as they have been validated for the tested animal species and in the laboratory in which the tests are performed and it has been demonstrated that the performance is meeting the required criteria. For meat inspection the required method sensitivity thought sufficient to prevent human clinical trichinellosis is at least one to three larvae per gram of tissue. For epidemiological studies in wildlife the test sensitivity can be increased by adjusting the sample size.
One method to ensure these standards are met is to carry out specific training and then ring trials, as stipulated in Regulation (EC) No 2075/2005 (EC, 2005b) and recommendations made by the International Commission on Trichinellosis (Gamble et al., 2000). Further details on ring trial details are listed below.

In 2005, the cost estimates for classical *Trichinella* inspection by digestion, ranges from Euro 0.12 to Euros 2.5 per pig. In large industrialised slaughterhouses in Denmark (10,000 pigs per day), the cost estimate for inspection by pooled sample digestion is Euro 0.15 per pig (Kapel, 2005). In France, the cost increases up to Euros 10.00 per horse. The cost increases dramatically when wild animals are tested due to the large amount of meat, which should be tested per animal and the longer digestion time.

Table 2: Summary of methods used for meat inspection for *Trichinella* taken from Webster et al., 2006.

<table>
<thead>
<tr>
<th>Table 1</th>
<th>Methods used for meat inspection for <em>Trichinella</em> in pork, horsemeat and wild boar in EU (according to current Directive 77/96/EEC)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Method number according to Directive 77/96/EEC - Annex I</td>
<td>Method</td>
</tr>
<tr>
<td>I</td>
<td>Trichinoscopy compressorium</td>
</tr>
<tr>
<td>II</td>
<td>Digestion (no mechanical intervention)</td>
</tr>
<tr>
<td>III</td>
<td>Digestion (twice hourly manual shaking)</td>
</tr>
<tr>
<td>IV</td>
<td>Stomacher (constant mechanical treatment) and sedimentation</td>
</tr>
<tr>
<td>V</td>
<td>Stomacher (constant mechanical treatment) and filtration</td>
</tr>
<tr>
<td>VI</td>
<td>Magnetic edirrer (coagulant mechanical treatment)</td>
</tr>
<tr>
<td>VII</td>
<td>Trichomatic 35 blender</td>
</tr>
</tbody>
</table>

1: Directive 77/96/EEC was repealed by Directive 2004/41/EC.
2: MSs may allow trichinoscopic examination for domestic swine and wild boar in exceptional cases until 31 December 2009. Further specifications regarding this exception can be found in Regulation (EC) No 2075/2005 (EC, 2005b), Article 16.
Table 3: Ranking of predilection sites of encapsulating and non-encapsulating *Trichinella* spp. in muscle tissues of experimentally infected animals. Ranking: 1 = highest mean infestation; - = no muscles sampled. Table taken from Kapel et al., 2005.

<table>
<thead>
<tr>
<th>Pig/wild boar</th>
<th>Horse</th>
<th>Fox</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Ts, Tna, Tb, Tne</td>
<td>Tpse</td>
</tr>
<tr>
<td>Tongue base</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Diaphragm</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Masseter</td>
<td>6</td>
<td>3</td>
</tr>
<tr>
<td>Tongue tip</td>
<td>3</td>
<td>5</td>
</tr>
<tr>
<td>Neck</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>Abdomen</td>
<td>7</td>
<td>8</td>
</tr>
<tr>
<td>Tenderloin</td>
<td>10</td>
<td>7</td>
</tr>
<tr>
<td>Throat</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>Shoulder</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Intercostals</td>
<td>9</td>
<td>10</td>
</tr>
<tr>
<td>Upper jaw</td>
<td>11</td>
<td>12</td>
</tr>
<tr>
<td>Upper forelimb</td>
<td>13</td>
<td>9</td>
</tr>
<tr>
<td>Lower forelimb</td>
<td>14</td>
<td>13</td>
</tr>
<tr>
<td>Upper hind limb</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Lower hind limb</td>
<td>12</td>
<td>11</td>
</tr>
<tr>
<td>Rump</td>
<td>8</td>
<td>14</td>
</tr>
<tr>
<td>Filet</td>
<td>15</td>
<td>15</td>
</tr>
</tbody>
</table>

T6 = *Trichinella* genotype 6
Tb = *T. britovi*
Tm = *T. murrelli*
Tna = *T. nativa*
Tne = *T. nelsoni*
Tpse = *T. pseudospiralis*
Ts = *T. spiralis*
Table 4: Digestion time (hours) of muscle tissue from different hosts. Complete digestion of 20g minced (3 mm) tissue sample in 500 ml HCl/pepsin by magnetic stirrer technique at 45 °C (Kapel et al., 2005)

<table>
<thead>
<tr>
<th>Pig/wild boar</th>
<th>Fox</th>
<th>Horse</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tenderloin</td>
<td>1/2</td>
<td>-</td>
</tr>
<tr>
<td>Filet</td>
<td>1/2</td>
<td>½</td>
</tr>
<tr>
<td>Diaphragm</td>
<td>1/2</td>
<td>½</td>
</tr>
<tr>
<td>Rump</td>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>Upper hindlimb</td>
<td>1</td>
<td>1-1/2</td>
</tr>
<tr>
<td>Intercostals</td>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>Neck</td>
<td>1</td>
<td>1-1/2</td>
</tr>
<tr>
<td>Masseter</td>
<td>1</td>
<td>1-1/2</td>
</tr>
<tr>
<td>Shoulder</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Abdomen</td>
<td>1</td>
<td>-</td>
</tr>
<tr>
<td>Upper forelimb</td>
<td>1</td>
<td>1-1/2</td>
</tr>
<tr>
<td>Lower hindlimb</td>
<td>1-1/2</td>
<td>1-1/2</td>
</tr>
<tr>
<td>Lower forelimb</td>
<td>1-1/2</td>
<td>2</td>
</tr>
<tr>
<td>Tongue base</td>
<td>1-1/2</td>
<td>2</td>
</tr>
<tr>
<td>Tongue tip</td>
<td>1-1/2</td>
<td>-</td>
</tr>
</tbody>
</table>
Test and laboratory performance evaluation:

Laboratories performing digestion methods for monitoring should be evaluated by the National Reference Laboratories (NRL). Additionally the NRLs should be part of the CRL proficiency testing. To evaluate the competence and skills of participants and the sensitivity of the digestion method in each participating laboratory, ring trials are being carried out in which meat samples are spiked with a certain number of larvae. These procedures are described in detail in Vallée et al., 2007 and Marucci et al., 2009. Test sensitivity in this case can be expressed as the percentage of muscle larvae recovered from each proficiency sample (= recovery rate), and here a required minimum recovery rate of 40% is discussed as acceptable for the moment, though the long-term goal should probably be an increase to about 75%, which is required by the Canadian accreditation programme for testing pork and horsemeat (Forbes et al., 2005). Likewise under discussion and in need of a harmonised approach, is how to derive the 'diagnostic sensitivity' and 'surveillance sensitivity', taking into account not only the recovery rate but also the sample size from an individual animal and the infection level in individual animals as well and the likely prevalence in a population, which is especially important for wildlife testing.

Indirect test to detect antibodies:

Serological tests (ELISA) intended for the detection of specific anti-Trichinella-antibodies are suitable for monitoring in domestic pigs but these assays are not recommended as a substitute for meat inspection of individual carcasses (Gamble et al., 2004). Due to seroconversion at the beginning of infection (two to three weeks for high, three to five weeks for moderate and more than five weeks for low infection dose) a “diagnostic window” with false-negative results may occur at which animals with infective muscle larvae cannot be detected. However, once seroconversion occurs, the sensitivity of serology (ELISA) is higher compared to larval findings by means of artificial digestion where sensitivity is limited to one to three larvae per gram which is usually used for examination in finisher pigs (Nöckler et al., 2000; Gamble et al., 2004).

Serological assays like ELISA tests can easily be automated and examination may include either blood serum or meat juice. Several in-house and commercial ELISA tests are preferably used for serological monitoring in pigs and wild boar. In a US programme for certification in the pork industry, pigs are randomly tested at slaughter by ELISA to verify that animals from certified farms are free of Trichinella infection (Pyburn et al., 2005). Experimental and field studies have demonstrated that the serological response after a Trichinella infection in horses is less consistent than observed in pigs (Gamble et al., 2004).

Most of these ELISA assays are based on excretory/secretory antigen and several studies revealed an overall sensitivity and specificity ranging from 93.1% to 99.2% and from 90.6% to 99.4%, respectively (Gottstein et al., 2009). In a recent study, the detection of a serological response was analysed in animals with different worm burdens. It was shown that the probability of detecting a serological positive animal was low in animals with low worm burdens indicating the difficulty using serology in monitoring programmes (Teunis et al., 2008). New insights using other validation methods might be used in the future. Harmonised protocols for the preparation of antigens as well as appropriate reference sera for the standardisation of ELISA and the calibration of the cut-off are not available yet. Additionally, there is a need for the development of appropriate sampling schemes if serological assays are used for monitoring programmes in the future.
Standardisation:

During the last meeting of CEN/TC275/WG6 (CEN/Technical Committee 275 Food analysis - Horizontal methods/Working Group 6 - Microbial contamination), held in Helsinki in May 2008, the WG 6 members approved a resolution (R. 172, TAG 7: Parasites) proposed by the CRL for parasites, to start working on the standardisation of a serological method for the detection of anti-\textit{Trichinella} antibodies in swine serum, together with the standardisation of the artificial digestion method. Article 11 of Regulation (EC) No 2075/2005 (EC, 2005b), includes the use of a serological method for monitoring purposes "once a suitable test is validated by the CRL". Such a test was validated and accredited by the CRLP in 2006, and further characterised by a collaborative study and can be accessed online at: [http://www.iss.it/binary/crlp/cont/First\%20Ring\%20Trial\%20ELISA\%20Trichinella.1192528039.pdf](http://www.iss.it/binary/crlp/cont/First%20Ring%20Trial%20ELISA%20Trichinella.1192528039.pdf).

This is awaiting the next step, which will be a call for experts on this issue. This is a state-of-the-art standardisation process.
Objective 4. Propose harmonised monitoring and reporting schemes

Within EU MSs, monitoring for Trichinella is currently being carried out according to Regulation (EC) No 2075/2005 (EC, 2005b). This Regulation stipulates that all pigs for human consumption must be tested but also permits various exceptions for pigs and other species if certain conditions are met. This is the basis for MSs applying different sampling schemes in different populations, according to various needs. Various testing methods are also specified in the regulations. Whereas this approach is useful from an epidemiological point of view, it makes it difficult for analysing data and comparing results at Community level. One of the notable issues is that the definition of negligible risk according to Regulation (EC) No 2075/2005 (EC, 2005b) is not well described.

4.1 Recommendation

In Regulation (EC) No 2075/2005 (EC, 2005b) no criteria are given to define negligible risk. It would be very helpful to define these and to prepare a format for the MSs specifying which criteria have to be fulfilled when applying for negligible risk, e.g. scenario tree versus Bayesian versus portfolio approach. It is recommended that EFSA commission a working group of mathematical modellers, epidemiologists, parasitologists and statisticians to review the methods available to decide what the criteria should be for negligible risk/disease freedom. In particular the problem of detecting disease at a very low prevalence, especially in small populations, needs to be addressed. It would also be essential to take into account economic and public health aspects to agree a valid level at which the risk to humans is considered to be minimal/negligible.

In the meantime, we propose a simplified framework, described in the following chapters, which would allow MSs with little or no historic data to reach a status, based on gathered evidence over a relatively short period of time, resulting in a reduced testing of low risk pig populations. The evidence and information gathered over successive years of the proposed scheme would allow broad scale trends in the status of MSs to be followed for targeted livestock populations.

In this framework we also address the negligible status for all fattening pigs in combination with wildlife testing. The requirements used in our framework (test sensitivities/prevalences) are in line with Regulation (EC) No 2075/2005 (EC, 2005b) as far as possible and with current practices and precedence cases, where regulations do not provide sufficient guidelines. We do advise though that these parameters should eventually be clarified as proposed in the recommendations above.

4.2 Definitions

Controlled Housing

For the purposes of this document, “controlled housing conditions and integrated production systems” is defined according to the FCI and along the lines of Appendix to Annex VIb in Regulation (EC) No 1244/2007 (EC, 2007).

Non-Controlled housing

Pigs from holdings that do not fall into the category ‘controlled housing’.

4.3 Sampling

The sampling scheme employed by a region and the animal populations that should be monitored are dependent on the status of the region with respect to Trichinella. Currently, testing in MSs is carried...
out according to Regulation (EC) No 2075/2005 (EC, 2005b). As mentioned before, within these regulations millions of ‘low-risk’ pigs are tested annually, whereas often the higher risk populations are left out. One reason that was identified was that in some MSs pigs that are slaughtered for ‘private consumption’ or direct supply to the consumer i.e. within a circle of family and friends, are not officially required to be tested although voluntary schemes may be in place. Whereas it is acknowledged that testing may be more difficult and more costly for individual animals in remote regions, from a public health point of view this cannot be considered acceptable and we strongly recommend that these exceptions to testing must be addressed. Below is a simplified sampling scheme, developed to facilitate MSs that cannot fall back on years of historic data and cannot demonstrate a high certainty about their situation, to be able to gather sufficient evidence relatively quickly to reduce the number of ‘low risk’ pigs for testing. In our view, if this is carried out reliably, public health needs would not be compromised, but would have large economic benefits and free resources that could then be focused on the populations posing a higher risk and therefore protect consumer health more efficiently. The proposed sampling scheme is a preliminary framework that in parts deviates from current EC Regulations, which would allow countries that have gathered sufficient evidence to fall into one of these categories and then continue to provide the evidence as proposed. Regions can be in one of three status categories. Table 6 (Appendix E) outlines the categories and testing that would be required in each.

The following can be applied to an entire MS, at national level or, where appropriate, to a defined region within a MS (e.g. Northern Ireland in UK).

In all areas where there are exceptions to testing all animals, farmers should also be given advice on biosecurity to minimise the risk of introduction of Trichinella through farm practices. A contingency plan should also be in place in case of a confirmed positive animal in a population assumed to have negligible risk. Details on contingency plans can be found in Regulation (EC) No 2075/2005 (EC, 2005b), and should identify:

- the origin of the infected pig;
- the presence of other infected pigs; and
- the source of infection (wildlife?).

It is important that contingency plans lead to corrective measures and increased biosecurity.

4.3.1 Region 1: ‘Endemic”

Definition

Trichinella present and circulating in livestock and wildlife unless proven otherwise.

Proposed sampling scheme

In countries where Trichinella is circulating, all Trichinella-susceptible animals destined for human consumption should be tested by artificial digestion, according to the EC Regulation (EC) No 2075/2005 (EC, 2005b).

Rationale

If all animals for consumption are monitored, there is considered to be little extra benefit in the monitoring of other wildlife. In this situation other wildlife does not need to be monitored (see below for certification of holdings) but wildlife should still be tested if it is destined for human consumption. The information from meat inspection can be used for surveillance and control.
In this category individual holdings may apply for *Trichinella*-free certification in accordance with current EC Regulation (EC) No 2075/2005 (EC, 2005b). In this case, risk based wildlife sampling would be required to be carried out in the area of the holding. The results of this sampling should be reported to EFSA with positive and negative animals identified. The sampling strategy needs to be harmonised between MSs.

The animal populations in which trends should be followed are summarised in Table 6 (Appendix E).

### 4.3.2 Region 2: Low-risk regions

This region does not fall in line with current EC regulations and is a newly proposed category. However, it has no significance for testing currently carried out in EU MSs and is proposed for discussion and future consideration.

'Low-risk' refers to the risk of *Trichinella* in fattening pigs from controlled housing. Regions in this group must provide evidence that the surveillance sensitivity is 95% or greater (see Appendix B) in fattening pigs from controlled housing.

All sows and boar, fattening pigs from outdoor and non-controlled indoor housing and wildlife for human consumption should still be tested as stipulated under EC Regulation (EC) No 2075/2005 (EC, 2005b).

Once an area has been granted low risk status, a statistically significant sample of fattening pigs from controlled housing should be sampled. Sampling should be stratified by slaughterhouse throughput to ensure a representative sample of the population. See Appendix C.

### 4.3.3 Region 3a: Regions with negligible risk in fattening pigs from controlled housing

Regions in which there is negligible risk that *Trichinella* is present in the defined livestock population will submit evidence that there is negligible risk that *Trichinella* is present in the controlled fattening pig population, given the design prevalence of less than 1/million. One method of providing this evidence is to show that the sensitivity of the surveillance in pigs from controlled housing is high e.g. 99% (see Appendix C).

### 4.3.4 Region 3b: Regions with negligible risk in all fattening pigs

The region will submit evidence that the likely occurrence of *Trichinella* in the controlled-housing pig population is less than 1/million and furthermore the criteria are fulfilled as set out in Regulation (EC) No 2075/2005 (EC, 2005b). One method of providing this evidence is to show that the sensitivity of the surveillance in pigs is high e.g. 99% (see Appendix C).

In either status, fattening pigs from farms that comply with the definition of controlled housing do not require testing. Sows and boar from these farms will all need to be tested. Furthermore, wildlife meant for human consumption needs to be tested in accordance with current Community legislation.

Depending on whether the area is applying for:

3a) negligible risk in fattening pigs in controlled housing; or

3b) negligible risk in the total fattening pig population, only:

3a) all pigs from non-controlled housing will be tested;
3b) a sample of pigs from non-controlled housing can be tested. The possibility of further classification within this group (pigs born, reared and finished under non-controlled conditions versus pigs born and reared under non-controlled housing but finished under controlled conditions) has been proposed for further consideration.

Depending on whether the area is applying for:

3a) negligible risk in fattening pigs in controlled housing; or
3b) negligible risk in the total fattening pig population, only:

3a) no testing of wildlife is required in this category as all outdoor pigs are still tested and this is sufficient. 3b) Wildlife should be tested to confirm an occurrence of equal to or less than 0.1% in the sampled population over a minimum of 10 years. This will involve a targeted sample of a minimum of 300 animals (fox or racoon dogs) per year or 3,000 over 10 years. An alternative for regions with small wildlife populations could be the detection of a prevalence of 0.1% with 95% confidence, based on local estimated populations of target animals (e.g. foxes, wild boar, racoon dogs).

All other wildlife for human consumption (including farmed wild boar) should be tested.

Risk-based sampling of wildlife

It is suggested that these areas be identified through the geographical analysis of the domestic pig and wildlife populations or on previous occurrences of *Trichinella* in wildlife or pigs. The area to be covered by the targeted surveillance will be defined by the MS and sampling should be carried out to enable the detection of a prevalence of *Trichinella* of 0.1% or greater over a period of 10 years (or shorter if larger numbers of foxes are available; e.g. 3,000 foxes tested in five years).

If a confirmed positive case is found in the population (either fattening pigs from controlled housing in region 3a or pigs and wildlife in region 3b), which was believed to have negligible risk, regions should have contingency plans in place. This may involve an investigation into the source of the positive case and whether further cases exist in the population. During the investigations, the status of the region will not change until the investigation is complete. If the investigation reveals more positive animals, the status might be lost; if no further positives are found then evidence is accumulated and the status is kept.

4.3.5 Methodology to determine low and negligible risk status

Under current EC legislation (Regulation 2075/2005 (EC, 2005b)), a category of holdings can be recognised as free from *Trichinella* where surveillance has been carried out, providing at least 95% confidence that where the prevalence of *Trichinella* exceeds 0.0001%, any infestations will be detected. However, no guidance is given as to how this should be demonstrated. A number of different statistical approaches to demonstrating disease freedom have been proposed in recent literature (Martin et al., 2007; Böhning & Greiner, 2006; Ebel et al., 2008; Branscum et al., 2006). These methods differ not only in the statistical methodology but also in data requirements. The scenario tree modelling method developed by Martin et al., 2007 has been adapted for use by Alban et al., 2008 in order to demonstrate that Denmark had a negligible risk of *Trichinella* in fattening pigs from controlled housing. Clearly, a single, robust approach, suitable for *Trichinella* is required but if a single statistical or modelling approach is to be recommended more work is needed to validate the approaches e.g. comparison of outputs using the same data and sensitivity analyses.
In the present work it was felt that the method adapted from Martin et al. (2007) based on the probability of the detection of disease given the test sensitivity and design prevalence, provides an adequate approach for the needs of this work. This method (Appendix B) is simple and transparent, yet provides adequate support for demonstrating that there is negligible risk of Trichinella in the defined population. None of these methods, however, can overcome the basic limitations of sampling theory, which mean that in large populations with a very low prevalence the number of animals that must be tested in order to detect disease is very large. This causes difficulties for countries with small pig populations and sufficient flexibility should be allowed, e.g. through the use of historical data over a number of years, to ensure these countries are not penalised.

4.4 Recommendation

As previously mentioned, it is recommended that EFSA put together a working group of mathematical modellers, epidemiologists, parasitologists, and statisticians to review the methods available to demonstrate negligible risk/disease freedom. In particular the problem of detecting disease at very low prevalence especially in small populations needs to be addressed.

4.5 Evidence needed for a region to move from region 1 (‘endemic’) to region 2 (‘low risk’)

Note: the proposed regions are not meant as stages that all countries necessarily have to run through. If sufficient historic data exist that allow sufficient evidence, countries can be moved into the suitable region and continue to provide evidence as described.

The area or MS must show that the surveillance system sensitivity in fattening pigs, with a design prevalence of 0.0001 (1/million) is greater than 95%. The method of calculating the sensitivity of the surveillance system is given in Appendix B.

4.6 Evidence needed for a region to move from region 2 (‘low risk’) to region 3 (‘negligible risk c’)

1) Negligible risk in fattening pigs from controlled housing

The area or MS must show that there is a greater than ≥99% probability that the target population (fattening pigs from controlled housing) is free from infection at the design prevalence (1/million).

2) Negligible risk all fattening pigs

The area or MS must show that there is greater than 99% probability that the pig population is free from infection at the design prevalence (1/million).

Regions with small pig populations

Some regions have small pig populations, which cannot meet the required sample sizes to show low or negligible risk. It is suggested that as long as all pigs are tested in all sectors they should use historical data for more than 10 years. However, they will be required to continue to sample all pigs unless the criteria for reduced sampling is met. Other options include merging with a larger region in order to increase the overall regional pig population.
4.7 Sample sizes

4.7.1 Rationale for sample sizes

Once low risk (region 2) status or negligible risk status (region 3) in the pig population has been granted, the sample sizes required for fattening pigs from controlled housing and pigs from non-controlled housing (where samples of the population are tested) are outlined in Appendix D. The sample size should give confidence that the population remains free from infection at the specified level.

Where sampling of a proportion of the population is carried out, the sampling must be stratified and sampling carried out in proportion to the throughput of the slaughterhouse. As far as possible, sampling should also be spread throughout the year to avoid sampling the same farms or batches as far as possible. See Appendix D for an example.
Objective 5. Reporting

5.1 Data to be collected at national and/or EU level

The recommended information to be collected by MSs is described below and consists of two categories: a description of a surveillance programme and individual data for each sample. Due to the fact that the current EFSA web application can only collect aggregated data, reporting at EU level is restricted to overall results. However, future developments may allow the collection and analysis of individual level data in the EU and MSs should be encouraged to design databases to collect this information for national reporting. Some information e.g. production type of pigs, is collected as part of the FCI under the new hygiene regulations and should be available for all MSs.

5.1.2 EU level reporting: Description of surveillance programme

- MS name
- Status of MS; *Trichinella* present, low risk or ‘negligible risk’ (group 1, 2, 3).
- Date of start and end of surveillance (usually covering the year)
- Type of surveillance (all, risk-based, sample) in each population category (e.g. outdoor fattening, boar and sows)
- Number of positive animals in each sampled population category
- Number of animals tested in each sampled population category
- Percentage of animals tested that were positive in each sampled population category

5.1.3 National level collection and reporting: Individual sample information (for ALL wildlife and ALL positive pigs)

- Species and production type
- Date of analysis
- Analysis method used
- Species of parasite detected
- Geographical origin of the carcass if horses or wildlife

5.1.4 Population and related data

- Total population of the following in each MS (if known):
  - pigs (controlled and non-controlled housing for fattening pigs, breeding pigs plus the geographical location of pig farms - amalgamated to Nuts region)
  - description of how FCI (in particular whether finisher has been reared indoor since weaning) is exchanged between farm owner and slaughterhouse prior to slaughter
  - horses
  - farmed wild boar
  - foxes (population estimate)
  - racoon dogs (population estimate)
  - number of wild boar hunted (population estimate)
  - number of other wild animals slaughtered or hunted for human consumption

5.1.5 Reporting to EFSA

Reporting to EFSA will continue with the web-based form. Changes to the form will need to be made to reflect the additional data collected above.
Development of harmonised schemes for the monitoring and reporting of *Trichinella* in animals and foodstuffs in the European Union

Objective 6. Propose information to be analysed by the Commission and EFSA for detecting trends

6.1 Descriptive Analysis

- Tables showing the proportion of positive samples in each MS for each animals species and where applicable (e.g. pigs), production type monitored.
- Estimation of Community prevalence of *Trichinella* in each species and/or production type. Where MSs do not sample all animals, weighting to account for the proportion of animals sampled within the MS may be required to estimate prevalence.

6.2 Monitoring of trends over time

For the monitoring of trends in the incidence/prevalence of *Trichinella* in different animal species or types, the majority of countries will contribute very few positive results in livestock. Therefore it is recommended that reporting be carried out at species level and for pigs, for controlled housing and outdoor and sows and boar separately as this information is of interest. Where a stratified sample or all animals are tested, prevalence estimates can be obtained and compared between years. Due to the low levels it is likely that data from a number of years will need to be analysed in order to detect significant trends and it may take a number of years to detect any underlying changes in the populations. Methods such as logistic regression or other simple non-parametric tests such as chi-squared could be used to determine significance of differences between years in a MS. At Community level multilevel models could be applied to account for differences between MSs, using time as a covariate. It is recommended that the advice of a statistician be sought prior to any analysis as the appropriate methodology will depend on the quantity and quality of data collected and reported.

6.3 Spatial analysis

Geographical analysis should/can be carried out at regional level where surveillance is also carried out regionally or compartments are based on regions. Choropleth maps showing 1) the status of each region with relation to *Trichinella* (class 1, 2 or 3) and 2) the prevalence of *Trichinella* in each animal group within each region. Note that for most cases the prevalence will be very low. Cartograms may be useful to illustrate the distribution of pigs within the EU overlaid with prevalence figures.
REFERENCES


Development of harmonised schemes for the monitoring and reporting of Trichinella in animals and foodstuffs in the European Union


Kapel CMO, 2000. Host diversity and biological characteristics of the Trichinella genotypes and their effect on transmission, Veterinary Parasitology, 93, pp. 263-278.


Development of harmonised schemes for the monitoring and reporting of *Trichinella* in animals and foodstuffs in the European Union


APPENDICES

A. DEFINITION OF CONTROLLED HOUSING

Appendix to Annex VIib in Regulation (EC) No 1244/2007 (EC, 2007)

For the purposes of this Appendix, “controlled housing conditions and integrated production systems” means that the food business operator needs to comply with the criteria set out below:

(a) all feed has been obtained from a facility which produces feed in accordance with the requirements provided for in Articles 4 and 5 of Regulation (EC) No 183/2005 (EC, 2005a) of the European Parliament and of the Council of 12 January 2005 laying down requirements for feed hygiene (1); when roughage or crops are provided to the animals as feed, it shall be treated appropriately, and where possible, dried and/or pelleted;

(b) an all-in/all-out system is applied as far as possible. Where animals are introduced into the herd, they shall be kept in isolation as long as required by the veterinary services to prevent the introduction of diseases;

(c) none of the animals has access to outdoor facilities unless the food business operator can show by risk analysis to the satisfaction of the competent authority that the time period, facilities and circumstances of outdoor access do not pose a danger for the introduction of disease into the herd;

(d) detailed information is available concerning the animals from birth to slaughter and their management conditions as laid down in Section III of Annex II to Regulation (EC) No 853/2004 (EC, 2004a);

(e) if bedding is provided for the animals, the presence or introduction of disease is avoided by the appropriate treatment of the bedding material;

(f) holding staff comply with the general hygiene provisions as laid down in Annex I to Regulation (EC) No 852/2004 (EC, 2004b) of the European Parliament and of the Council of 29 April 2004 on the hygiene of foodstuffs;

(g) procedures are in place that control access to the premises where animals are kept;

(h) the holding does not provide facilities for tourists or for camping unless the food business operator can show by risk analysis to the satisfaction of the competent authority that the facilities are sufficiently separated from the animal rearing units that direct and indirect contact between humans and animals is not possible;

(i) animals do not have access to rubbish dumps or household waste;

(j) a pest management and control plan is in place;

(k) silage feeding is not used unless the food business operator can show by risk analysis to the satisfaction of the competent authority that the feed cannot transmit any hazards to the animals;

(l) effluent and sediment from sewage treatment plants are not released in areas accessible to animals or be used for fertilising pastures used to grow crops, which are used to feed animals, unless treated appropriately and to the satisfaction of the competent authority.
B. **CALCULATION OF SURVEILLANCE SENSITIVITY**

(see Also Martin et al., 2007)

If a surveillance process tests a representative group of \( N \) animals, all with negative results, and these animals are assumed to be independent of each other with regard to the probability of infection, the overall sensitivity of the surveillance system (SSe) is the probability that one or more positive pigs will be detected, given that the region is infected (Martin et al 2007a).

Surveillance Sensitivity = Pr(Identifying infection) = 1 - Pr(overlooking infection)

\[ = 1 - (1 - Se)^n \]

Where Se is the test sensitivity = 0.4 (Alban et al., 2008), \( n \) is the expected number of positive animals in the population given the design prevalence.

**Example:** If 15 million fattening pigs from controlled housing are tested in region X, and the design prevalence is 0.0001% (1/million), there are an estimated 15 positive pigs among those tested. Thus assuming the sensitivity of the detection method of 40%, the surveillance sensitivity is:

\[ = 1 - (1 - 0.4)^{15} = 0.999 \]

If the disease is present in the region then the probability that one or more positive pigs will be detected is 99.9%.

This method is dependent on the number of positive animals being identified in the sampled population. It does not take into account the risk that disease is introduced into the population during the monitoring period. Martin et al. (2008) and Alban et al. (2008) present models that extend the equations presented here to incorporate this.
C. EVIDENCE REQUIRED TO MOVE CHANGE GROUP

Example: Group 1 to Group 2

Region X wishes to apply to move from Group 1 to Group 2. For the previous 10 years all pigs have been tested for *Trichinella* as required under legislation and no positive pigs have been found for many years.

The pig population is approximately 15 million fattening pigs from controlled housing. The number of positive animals from this sector expected if all animals are tested under the design prevalence or 1/million is therefore 15 per year.

Assuming test sensitivity is 40% then the sensitivity of the fattening pig surveillance if all animals are tested in that year is:

\[ S_{se} = 1 - (1 - 0.4)^{15} = 0.999 \]

[Note if a country has a small pig population, or does not currently test all pigs, historical data can be used. The number of positive animals in the sample is multiplied according to the total number of animals tested e.g. in the above, for two years of sampling the total number of positive animals would be 30.]

The region is granted Group 2 status and changes the monitoring to a sample of fattening pigs from controlled housing.

**Scenario 1:** The region then wishes to apply for negligible risk status (Group 3) in its pig population as no positive pigs have been found. The region wants to stop monitoring in fattening pigs from controlled housing and to sample a proportion of pigs from non-controlled housing. While putting together the documentation a wildlife sampling programme is put into place and 2,000 foxes and raccoon dogs are sampled with one positive animal detected (prevalence <0.1%).

The region must show that the sensitivity of the surveillance system in the whole pig population is ≥99%

In addition to the 15 million fattening pigs the region has tested three million pigs from non-controlled housing and 500,000 sows and boar per year. This makes a total of 18.5 million animals so 19 positive animals are expected.

For one year of testing, the surveillance sensitivity is:

\[ S_{Se} = 1 - (1 - 0.4)^{19} = 0.999\% \]

[Note if the pig population is smaller, historical data can be used assuming no positives are found during that period.]

Together with this evidence and a monitoring programme in wildlife the region is granted Negligible Risk status for its pig population for as long as no positive pigs are identified.
Scenario 2. Region X detected a positive pig on an outdoor farm the previous year. The controlled and non-controlled sectors of the industry are distinct and separate and the region wishes to apply for negligible risk in its fattening pigs from controlled housing so it no longer has to test these while maintaining the testing of the outdoor finishers.

Using the calculation of surveillance sensitivity above, the sensitivity of the surveillance in fattening pigs has already been shown to be 99.9%. The region has therefore satisfied the criteria that there is negligible risk of *Trichinella* being present in controlled housing fattening pigs and it can stop monitoring this population. As only this sector has been analysed, they must continue to test all pigs from non-controlled housing, all sows and boar and wildlife for human consumption.

[Note: it is important that a region has to resubmit to move from group 2 to group 3 to prevent regions moving from group 1 to 3. It will be a quick process as it is likely the evidence is already available, especially if historical data is used]
D. SAMPLING OF PROPORTION OF POPULATION (PROPORTION OF CONTROLLED HOUSING OR NON-CONTROLLED HOUSING FATTENING PIGS).

Sampling frame

The sampling frame will include all slaughterhouses in the region that slaughter pigs of the population of interest (e.g. fattening pigs from controlled housing). Throughput data for each population at each slaughterhouse will be required to stratify the sampling (e.g. the number of pigs from controlled housing slaughtered per year according to the FCI).

Sample size

The total sample size provides the number of animals to be tested per year. The sampling unit is the individual carcass.

The sample size is determined by the region and should give at least 90% probability of detecting if present at the design prevalence of 1/million. Data from previous years may be used., e.g. Region X decides to test 500,000 outdoor pigs per year. Using a rolling 10 year window, this gives a total of five million pigs in five years and five positive animals expected. The sensitivity of the surveillance system is 92% according to the equation in Appendix B.

Stratification

The total sample size for each region should be stratified across the slaughterhouses so that the number sampled in each slaughterhouse is proportional to the annual throughput. Furthermore, this should then be divided by 12 to give the number to be sampled per month.

This will ensure the whole population of target animals is eligible for sampling and maximise the chances of finding disease if present.

For example: slaughterhouse A slaughters approximately 70% of the total animals and is therefore allocated 70% of the 500,000 sample animals. Spread across the year this equates to just 29,167 animals to be sampled per month.

Table 5. Example of the calculation of sample sizes required for the stratification of sampling of slaughtered pigs by abattoir and month according to abattoir throughput

<table>
<thead>
<tr>
<th>Slaughterhouse</th>
<th>Throughput</th>
<th>Proportion of total national throughput</th>
<th>Total to sample annually</th>
<th>Number to sample each month</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>1,000,000</td>
<td>=1,000,000/1,500,000=0.7</td>
<td>=500,000*0.7</td>
<td>=350,000/12</td>
</tr>
<tr>
<td>B</td>
<td>400,000</td>
<td>=0.3</td>
<td>150,000</td>
<td>12,500</td>
</tr>
<tr>
<td>C</td>
<td>100,000</td>
<td>=0.1</td>
<td>35,000</td>
<td>4,167</td>
</tr>
</tbody>
</table>
### E. *Trichinella* - Zoonotic Species Risk Assessment

**Table 6:** Framework for simplified sampling scheme

<table>
<thead>
<tr>
<th>Population to be monitored</th>
<th>Monitoring Region 1</th>
<th>Monitoring Region 2</th>
<th>Monitoring Region 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fattening pigs from controlled housing</td>
<td>ALL (mandatory)</td>
<td>Proportionate sample to demonstrate surveillance sensitivity of ≥95% to detect infection of &lt;1 case/million in fattening pigs (≈ 6 million pigs)</td>
<td>No positive findings in region 2 for 2 years to demonstrate surveillance sensitivity in a) pigs from controlled housing or b) all pigs of ≥99%</td>
</tr>
<tr>
<td>Fattening pigs from non-controlled housing</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
<td>Proportionate sample to demonstrate surveillance sensitivity of ≥90% to detect infection of &lt;1/million. In combination with wildlife testing.</td>
</tr>
<tr>
<td>Sows and boar from controlled housing</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
</tr>
<tr>
<td>Sows and boar from non-controlled housing</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
</tr>
<tr>
<td>Horses</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
</tr>
<tr>
<td>Farmed wild boar</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
</tr>
<tr>
<td>Hunted wild boar</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
</tr>
<tr>
<td>Wildlife (other than wild boar) for human consumption</td>
<td>ALL (mandatory)</td>
<td>ALL (mandatory)</td>
<td>All (mandatory)</td>
</tr>
<tr>
<td>Other wildlife</td>
<td>Optional</td>
<td>Mandatory if move into Country class 3 (all pigs) anticipated, otherwise optional</td>
<td>If Class 3b (all pigs) then prevalence of &lt;0.1% must be demonstrated. Otherwise optional.</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>Optional Proportion to demonstrate a low level in wildlife &lt;0.1%</td>
</tr>
</tbody>
</table>

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## E. Trichinella - Zoonotic Species Risk Assessment

<table>
<thead>
<tr>
<th>Species</th>
<th>Host</th>
<th>Zoonotic (Y/N)</th>
<th>Human susceptibility (H/M/L)</th>
<th>Pathogenicity</th>
<th>Mortality Rate (%)</th>
<th>Long term effects</th>
<th>Geographical distribution</th>
<th>Presence in Europe (Y/N)</th>
<th>Likelihood of establishment in non endemic EU region (H/M/L)</th>
<th>Risk groups?</th>
<th>Monitoring in Europe recommended? (Y/N)</th>
<th>Comments</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trichinella spiralis (T1)</td>
<td>mammals including humans</td>
<td>Y</td>
<td>II</td>
<td>-++++</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown</td>
<td>North America, Europe and Asia; arctic and subarctic areas of Northern Hemisphere; southern India – 20°C; northern Alaska in January.</td>
<td>Y</td>
<td>N/A</td>
<td>People consuming raw or undercooked meat or meat products</td>
<td>Y</td>
<td>For the European distribution see Gottstein et al. (2009) or <a href="http://www.cdc.gov/trichinella/index.html">www.cdc.gov/trichinella/index.html</a></td>
<td>Gottstein et al., 2009; Pozio and Murrell, 2006*; Pozio et al., 2009.</td>
</tr>
<tr>
<td>Trichinella nativa (T2)</td>
<td>mammals including humans</td>
<td>Y</td>
<td>II</td>
<td>-++++</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown</td>
<td>Europe, Asia, Northern Australia, northern areas of African region, northern India – 20°C; northern Alaska in January.</td>
<td>Y</td>
<td>N/A</td>
<td>People consuming raw or undercooked meat or meat products</td>
<td>Y</td>
<td>In Europe, the circulation of this parasite is restricted to arctic and subarctic regions of Sweden, Finland, Estonia, this parasite does not infect pigs.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
<tr>
<td>Trichinella roberti (T3) [E. coli]</td>
<td>mammals including humans</td>
<td>Y</td>
<td>II</td>
<td>-+++</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown</td>
<td>Worldwide, Europe, in Europe has been reported in France, Italy, The Netherlands, Germany, Denmark, Sweden, Finland, Bulgaria, Hungary, Croatia, Slovak, Rep, and Lithuania.</td>
<td>N/A</td>
<td>Y</td>
<td>People consuming raw or undercooked meat or meat products.</td>
<td>Y</td>
<td>Is the Trichinella species present in most of EU countries but British islands and Ireland, Denmark, Malta and Cyprus, for the European distribution see Gottstein et al. (2009) or <a href="http://www.iss.it/site/trichinella/index.asp">www.iss.it/site/trichinella/index.asp</a> POZIO and Murrell, 2006. Pozio et al., 2008.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
<tr>
<td>Trichinella pseudoapolaris (T4)</td>
<td>mammals including humans and birds</td>
<td>Y</td>
<td>II</td>
<td>-++++</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown</td>
<td>Worldwide, in Europe, has been reported in France, Italy, The Netherlands, Germany, Denmark, Sweden, Finland, Bulgaria, Hungary, Croatia, Slovak, Rep and Lithuania.</td>
<td>N/A</td>
<td>Y</td>
<td>People consuming raw or undercooked meat or meat products</td>
<td>Y</td>
<td>Even if this parasite infect both mammals and birds, most of infections have been documented in mammals, especially in wild boar.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
<tr>
<td>Trichinella succinea (T5)</td>
<td>mammals including humans</td>
<td>Y</td>
<td>II</td>
<td>-+++</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown</td>
<td>Temperature areas of Neartic region; USA, Canada.</td>
<td>N</td>
<td>L</td>
<td>People consuming raw or undercooked meat or meat products, especially from imported Wild Boar from endemic regions. Humans imported from endemic regions have also been involved in human outbreaks.</td>
<td>2/3</td>
<td>This parasite does not infect humans. It has been imported in an infected form from USA which was the source of a human outbreak in France, in 1985.</td>
<td>Pozio and Murrell, 2006*; Anzalone R, Euro Surveill, 1998, 8:36-39.</td>
</tr>
<tr>
<td>Trichinella T4</td>
<td>mammals including humans</td>
<td>Y</td>
<td>II</td>
<td>-+++</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown</td>
<td>USA, Canada.</td>
<td>N</td>
<td>L</td>
<td>People consuming raw or undercooked meat or meat products, especially from imported Wild Boar from endemic regions. Humans imported from endemic regions have also been involved in human outbreaks.</td>
<td>2/3</td>
<td>This parasite does not infect humans. It has been detected only in carnivorous animals.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
<tr>
<td>Trichinella nativa (T7)</td>
<td>mammals including humans</td>
<td>Y</td>
<td>II</td>
<td>-+++</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown</td>
<td>Eastern Africa, Ethiopian region.</td>
<td>N</td>
<td>L</td>
<td>Humans</td>
<td>3/3</td>
<td>This parasite is circumscribed among carnivorous mammals and rarely infects wild pigs.</td>
<td>Pozio and Murrell, 2006*; Pozio et al., 2007.</td>
</tr>
<tr>
<td>Trichinella T8</td>
<td>mammals</td>
<td>(Y)*</td>
<td>unknown</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>South Africa, Namibia</td>
<td>N</td>
<td>L</td>
<td>Unknown</td>
<td>3/3</td>
<td>This parasite has been detected only in carnivorous mammals.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
<tr>
<td>Trichinella T9</td>
<td>mammals</td>
<td>(Y)*</td>
<td>unknown</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Japan</td>
<td>N</td>
<td>L</td>
<td>Humans</td>
<td>3/3</td>
<td>This parasite has been detected only in carnivorous mammals.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
<tr>
<td>Trichinella papuae (T11)</td>
<td>mammals including humans and reptiles</td>
<td>(Y)* (based on antibodies)</td>
<td>unknown</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Papua New Guinea and Thailand</td>
<td>N</td>
<td>L</td>
<td>Humans</td>
<td>3/3</td>
<td>This parasite has been detected only in carnivorous mammals.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
<tr>
<td>Trichinella taeniorhynchus (T12)</td>
<td>mammals including humans and reptiles</td>
<td>(Y)*</td>
<td>unknown</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Africa south of the Sahara; Ethiopia, Mozambique, Zimbabwe, South Africa</td>
<td>N</td>
<td>L</td>
<td>Humans</td>
<td>3/3</td>
<td>This parasite has been detected only in carnivorous mammals.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
<tr>
<td>Trichinella intermedius (T13)</td>
<td>mammals including humans and reptiles</td>
<td>(Y)*</td>
<td>unknown</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Unknown (lack of data or bias)</td>
<td>Argentina</td>
<td>N</td>
<td>L</td>
<td>Unknown</td>
<td>3/3</td>
<td>This parasite has been detected only in carnivorous mammals.</td>
<td>Pozio and Murrell, 2006*</td>
</tr>
</tbody>
</table>

*Y: Human cases not yet documented, but potentially considered zoonotic.  
*Y: Human cases not yet documented, but potentially considered zoonotic.  
*Y: Human cases not yet documented, but potentially considered zoonotic.  
*Y: Human cases not yet documented, but potentially considered zoonotic.  
*Y: Human cases not yet documented, but potentially considered zoonotic.  
*Y: Human cases not yet documented, but potentially considered zoonotic.  
*Y: Human cases not yet documented, but potentially considered zoonotic.

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F. **TRICHINELLA - RELEVANT ANIMALS AND FOODSTUFFS TO BE MONITORED**

<table>
<thead>
<tr>
<th>Animal species or foodstuff</th>
<th>Role in infection chain (DH/PH/SH/HL/DEH/RH)*</th>
<th>Part of human food chain/ diet (Y/N)</th>
<th>Known as source of human infection/ linked to outbreaks (Y/N)</th>
<th>Relevant to be monitored (Y/N)</th>
<th>Rationale for monitoring / application of result</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pigs</td>
<td>DH/PH</td>
<td>Y</td>
<td>Y</td>
<td>Y</td>
<td>Direct protection of human health</td>
<td>EC 2075/2005</td>
</tr>
<tr>
<td>Fattening pigs raised under controlled housing conditions and integrated production systems</td>
<td>DH/PH</td>
<td>Y</td>
<td>N</td>
<td>N</td>
<td>Direct protection of human health</td>
<td>Pozio, 1998</td>
</tr>
<tr>
<td>Fattening pigs NOT raised under controlled housing conditions and integrated production systems</td>
<td>DH/PH</td>
<td>Y</td>
<td>N</td>
<td>Y</td>
<td>Direct protection of human health</td>
<td>Pozio and Murrell, 2006</td>
</tr>
<tr>
<td>Breeding sows and boars</td>
<td>DH/PH</td>
<td>Y</td>
<td>Y</td>
<td>Y</td>
<td>Direct protection of human health</td>
<td>Martucci et al., 2008</td>
</tr>
<tr>
<td>Farmed wild boar</td>
<td>DEH/PH</td>
<td>Y</td>
<td>Y</td>
<td>Y</td>
<td>Indicator animal for determining prevalence in wildlife on Community level</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Horses</td>
<td>DEH</td>
<td>Y</td>
<td>Y</td>
<td>Y</td>
<td>Indicator animal for determining prevalence in wildlife on Community level</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Raccoon dogs</td>
<td>RH</td>
<td>N</td>
<td>N</td>
<td>Y</td>
<td>Indicator animal for determining prevalence in wildlife on Community level</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Foxes</td>
<td>RH</td>
<td>Not usually, but: human infections have been documented</td>
<td>Y</td>
<td>Y</td>
<td>Indicator animal for determining prevalence in wildlife on Community level</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Rodents (rats)</td>
<td>DH/PH</td>
<td>N</td>
<td>N</td>
<td>N</td>
<td>Not considered suitable for analysing trends on a Community level, as species do not occur throughout Europe and are under special protection (Bern convention). However, testing mandatory where meat consumed by humans. In regions, where the consistency of the population is high, these species could be good indicators.</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Bears</td>
<td>DH/PH</td>
<td>Only under exceptional circumstances.</td>
<td>Y</td>
<td>Y/N</td>
<td>Not considered suitable for analysing trends on a Community level, as species do not occur throughout Europe and are under special protection (Bern convention). However, testing mandatory where meat consumed by humans. In regions, where the consistency of the population is high, these species could be good indicators.</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Wolves</td>
<td>RH</td>
<td>N</td>
<td>N</td>
<td>Y/N</td>
<td>Indicator animal for determining prevalence in wildlife***</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Lynx</td>
<td>RH</td>
<td>N</td>
<td>N</td>
<td>Y/N</td>
<td>Indicator animal for determining prevalence in wildlife***</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Badger</td>
<td>RH</td>
<td>N</td>
<td>Y (Korea)</td>
<td>Y</td>
<td>Indicator animal for determining prevalence in wildlife***</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Strouts</td>
<td>DE/PH</td>
<td>N</td>
<td>N</td>
<td>N</td>
<td>Known cause of infections in humans, where dogs are consumed by humans. However, not relevant for Europe where dogs are Not part of human diet.</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Weasels</td>
<td>DH/PH</td>
<td>N</td>
<td>N</td>
<td>N</td>
<td>Known cause of infections in humans, where dogs are consumed by humans. However, not relevant for Europe where dogs are Not part of human diet.</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Mink</td>
<td>DH/PH</td>
<td>N</td>
<td>N</td>
<td>N</td>
<td>Known cause of infections in humans, where dogs are consumed by humans. However, not relevant for Europe where dogs are Not part of human diet.</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Stone marten</td>
<td>DE/PH</td>
<td>N</td>
<td>N</td>
<td>Y</td>
<td>Indicator animal for determining prevalence in wildlife***</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Marten</td>
<td>DE/PH</td>
<td>N</td>
<td>N</td>
<td>N</td>
<td>Known cause of infections in humans, where dogs are consumed by humans. However, not relevant for Europe where dogs are Not part of human diet.</td>
<td>Pozio et al., 2008</td>
</tr>
<tr>
<td>Dog</td>
<td>DH/PH</td>
<td>N</td>
<td>Y</td>
<td>N</td>
<td>Known cause of infections in humans, where dogs are consumed by humans. However, not relevant for Europe where dogs are Not part of human diet.</td>
<td>Pozio et al., 2008</td>
</tr>
</tbody>
</table>

*DH = definitive or final host in which an organism undergoes its sexual phase of reproduction. In the case of Trichinella, all hosts are DH as well as IH.*

(*IH = Intermediate Host. Animal in which the infectious agent undergoes some development, frequently with asexual reproduction).*

(*PH = Primary host. Animal that maintains an infection in its endemic area.*

(*SH = Secondary Host. Species that is additionally involved in the life-cycle of an agent, especially outside typical endemic areas.*

(*DEH = Dead-end host or incidental host. Host that usually does not transmit an infectious agent to other animals.*

(*RH = Reservoir Host. Host in which an infectious agent normally lives and multiplies, therefore a common source of infection (frequently a primary host).*)

***In some particular regions these animals can be good indicators, if the consistency of their population is high.

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**G. SUMMARY OF ANALYTICAL METHODS FOR TRICHINELLA**

<table>
<thead>
<tr>
<th>Diagnostic method/technique</th>
<th>Sensitivity</th>
<th>Specificity</th>
<th>Application (sample materials)</th>
<th>Application result</th>
<th>Throughput</th>
<th>Estimated costs per unit**</th>
<th>Technical requirements (instruments, etc)</th>
<th>Suitable for QA?</th>
<th>Comments</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>PCR analysis</td>
<td></td>
<td></td>
<td>Individuals/ herd level/ national prevalence etc.</td>
<td>E.g. number of animals tested per person and day.</td>
<td></td>
<td></td>
<td>blender, magnetic stirrer, separatory funnel, steve, funnel, beaker, stereo microscope</td>
<td>QA = Quality Assurance</td>
<td>No international Organization for Standardization (ISO) or the European Committee for Standardization (CEN) have standardised any of these methods. These methods should be validated and then standardized by a collaborative study involving a number of laboratories.</td>
<td></td>
</tr>
<tr>
<td>Magnetic stirrer method for pooled sample digestion</td>
<td>80-100%</td>
<td>100%</td>
<td>striated muscles of domestic swine, wild boar and horses.</td>
<td>Individuals/ herd level/ national prevalence / human health protection</td>
<td>500</td>
<td>0.15-2.5 €</td>
<td>Y</td>
<td>It is considered the gold standard method, this method has been validated in many laboratories, but it has been never standardized. European Commission Regulation 2075/2005, Annex I, Chapter I.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mechanically assisted pooled sample digestion method / sedimentation technique</td>
<td>?</td>
<td>100%</td>
<td>striated muscles of pigs, wild boar and horses.</td>
<td>Individuals/ herd level/ national prevalence / human health protection</td>
<td>500</td>
<td>0.15-2.5 €</td>
<td>Y</td>
<td>Only few laboratories use this method European Commission Regulation 2075/2005, Annex I, Chapter II (A)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Automatic digestion method for pooled samples of up to 35 g</td>
<td>80-90%</td>
<td>100%</td>
<td>striated muscles of pigs, wild boar and horses.</td>
<td>Individuals/ herd level/ national prevalence / human health protection</td>
<td>300</td>
<td>0.15-2.5 €</td>
<td>Y</td>
<td>European Commission Regulation 2075/2005, Annex I, Chapter II (B)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Magnetic stirrer method for pooled sample digestion</td>
<td>71% (recovery rate)</td>
<td>100%</td>
<td>Fenes, muscle tissue</td>
<td>Individual animals / National prevalence</td>
<td>60</td>
<td></td>
<td>Blender, magnetic stirrer, sieves, beaker, stereo microscope</td>
<td>Y</td>
<td>Based on method for pig but modified to adapt for different species. Zimmerman et al., 2008.</td>
<td></td>
</tr>
<tr>
<td>Trichinoscopy</td>
<td>0-100%</td>
<td>100%</td>
<td>Striated muscles, all animals. Does not reliably detect T. pseudospiralis or other non-encapsulated species.</td>
<td>individuals</td>
<td>15</td>
<td>3.0-10.0 €</td>
<td>compressorium, stereo-microscope</td>
<td>N</td>
<td>The sensitivity is related to the worm burden. This method is only accepted in exceptional cases until December 2009 (Article 16, EC 2075/2005). European Commission Regulation 2075/2005</td>
<td></td>
</tr>
</tbody>
</table>

*only for epidemiological surveillance on pig herds, not for diagnosis

**Will vary from country to country and depend on the throughput. Specify unit (animal/ pool of…). In this context it is meant to give a rough indication to allow comparison between methods, if possible.
### H. TRICHINELLA - SUMMARY OF COUNTRY RESPONSES

<table>
<thead>
<tr>
<th>MS</th>
<th>Contacted Institution</th>
<th>Response</th>
<th>Data on <em>Trichinella</em> available?</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Österreichische Agentur für Gesundheit und Ernährungssicherheit, Institut für veterinärmedizinische Untersuchungen Innsbruck</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>BE</td>
<td>Diergeneeskunde Department Prins Leopold instituut voor Tropische Geneeskunde</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>BG</td>
<td>Parasitic Zoonoses Laboratory National Diagnostic and Research Veterinary Institute</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>CY</td>
<td>State Veterinary Laboratory Veterinary Services</td>
<td>N</td>
<td>no</td>
</tr>
<tr>
<td>CZ</td>
<td>State Veterinary Institute, Department of Pathological Anatomy</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>DK</td>
<td>National Veterinary Institute, Technical University of Denmark, Section for Immunology and Parasitology</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>EE</td>
<td>Estonian Veterinary and Food Laboratory</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>FI</td>
<td>Oulu Research Unit Finnish Food Safety Authority Evira</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>FR</td>
<td>Food borne parasite NRL, UMR BIPAR INRA, AFSSA, ENVA, AFSSA LERPAZ</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>DE</td>
<td>Bundesinstitut Für Risikobewertung Bundesinstitut Für Risikobewertung</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>GR</td>
<td>Department of Parasitology Center of Athens Veterinary Institutions</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>HU</td>
<td>Central Veterinary Institute</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>IE</td>
<td>Central Meat Control Laboratory Veterinary Laboratory Department of Agriculture &amp; Food Laboratories</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>IT</td>
<td>National Reference Laboratory for Trichinella Istituto Superiore di Sanità</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>LV</td>
<td>Laboratory of Food and Environmental Investigations (LFEI)</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>LT</td>
<td>Food Microbiology Department, Laboratory department, National Food and Veterinary Risk Assessment Institute</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>LU</td>
<td>Diergeneeskunde Department Prins Leopold instituut voor Tropische Geneeskunde, Belgium</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>MT</td>
<td>Food Health and Diagnostics Laboratory, Veterinary Regulation, Fisheries Conservation and Control Division</td>
<td>N</td>
<td>no</td>
</tr>
<tr>
<td>NL</td>
<td>Microbiological Laboratory for Health Protection, National Institute of Public Health and the Environment</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>PL</td>
<td>Department of Parasitology of the National Veterinary Research Institute</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>PT</td>
<td>Laboratorio Nacional de Investigação Veterinaria</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>RO</td>
<td>Institute of Hygiene and Public Veterinary Health</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>SK</td>
<td>National Reference Laboratory for Parasites State Veterinary and Food Institute</td>
<td>N</td>
<td>via literature</td>
</tr>
<tr>
<td>SL</td>
<td>Laboratory of Parasitology, Veterinary Faculty, University of Ljubljana</td>
<td>N</td>
<td>via literature</td>
</tr>
<tr>
<td>ES</td>
<td>Centro Nacional de Alimentación. Agencia Española de Seguridad Alimentaria y Nutrición</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>SE</td>
<td>Dept. of Virology, Immunobiology and Parasitology, Section for Parasitology, National Veterinary Institute</td>
<td>Y</td>
<td>yes</td>
</tr>
<tr>
<td>UK</td>
<td>UK Food Standards Agency</td>
<td>Y</td>
<td>yes</td>
</tr>
</tbody>
</table>

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ABBREVIATIONS

CRL  Community Reference Laboratory
CSR  Community Summary Report
EC   European Commission
ECDC European Centre for Disease Prevention and Control
EFSA European Food Safety Authority
EU   European Union
FCI  Food Chain Information
IH   Intermediate host
MS   Member State
NRL  National Reference Laboratory
NUTS European Country Classification system
OIE  World Organisation for Animal Health
PCR  Polymerase chain reaction
PH   Primary host
QA   Quality Assurance
RA   Risk Assessment
RH   Reservoir host
SH   Secondary host
WHO  World Health Organisation
ZCC  Zoonoses Collaboration Centre