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EXPERT OPINION ON VACCINE AND/OR DIAGNOSTIC BANKS FOR MAJOR ANIMAL DISEASES

STRATEGIC PLANNING OPTIONS FOR EMERGENCY SITUATIONS OR MAJOR CRISES

1. ACKNOWLEDGEMENTS

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2. ACRONYMS AND ABREVIATIONS

AHS	African Horse Sickness
AHSV	African Horse Sickness virus
AI	Avian Influenza
ASF	African Swine Fever
ASFV	African Swine Fever virus
BT	Bluetongue
BTV	Bluetongue virus
CRL	Community Reference Laboratory
CSF	Classical Swine Fever
CSFV	Classical Swine Fever virus
CVMP	Committee for Medicinal Products for Veterinary Use
DG SANCO	Directorate General Health and Consumers
DIVA	Differentiating infected from vaccinated animals
EC	European Commission
EFSA	European Food Safety Authority
ELISA	Enzyme-linked Immunosorbent Assay
EMA	European Medicines Agency
EU	European Union
EUFMD	The European Commission for the Control of foot-and-mouth disease
FMD	Foot-and-mouth disease
FMDV	Foot-and-mouth disease virus
GMP	Good Manufacturing Practice
HPAI	Highly Pathogenic Avian Influenza
LPAI	Low Pathogenic Avian Influenza
MS	Member State
NRL	National Reference Laboratory
OIE	World Organisation for Animal Health
PCR	Polymerase chain reaction
Ph. Eur.	Pharmacopoeia Europaea
rRT-PCR	Real Time Polymerase Chain Reaction:

3. KEY MESSAGES

1. Vaccination is a fundamental tool in a strategy to control and eradicate major emerging diseases.

2. Emergency vaccination has to be considered as one tool in a whole range of measures as a part of a complex strategy to control and eradicate major animal diseases.

3. Emergency vaccination for most relevant infectious diseases should in general be seen in a new light, directly linked to the availability of effective diagnostic tools substantiating that vaccinated animals, or meat and other products obtained from vaccinated animals, are free from pathogens and can be traded safely.

4. Emergency vaccination has to be understood as *vaccinate-to-live*, meaning that vaccinated animals are kept to the end of a normal production cycle, and that their meat and other products can be marketed.

5. Diagnostic banks for particular infectious diseases are necessary to supplement vaccine banks to enable a holistic strategy of disease control and eradication.

6. The establishment and maintenance of vaccine and diagnostic banks must be part of a strategic plan prepared during 'peace time', ready for an emergency.

7. The issue of vaccine and diagnostic banks can only be treated in the context of a control and eradication strategy specific to each major animal disease (e.g. FMD, CSF, AI) and various outbreak scenarios.

8. For most of the relevant infectious diseases, existing legislation regarding emerging vaccination should be amended so that vaccination becomes a realistic option in the event of a crisis.

9. Trade issues regarding vaccinated animals or fresh meat and meat products obtained from vaccinated animals should be resolved.

10. Relevant legislation regarding veterinary medicinal products is not well suited to approve the use of vaccines in emergency situations.

11. The current review of legislation dealing with veterinary medicinal products is an ideal opportunity to introduce a mechanism to approve vaccines for emergency use at European level.

12. Proposals to be considered could include alternatives to vaccine banks, such as vaccine master seed stocks and 'mock up' authorisations for particular vaccines.

13. Vaccination and testing should replace unnecessary culling.

4. SUMMARY

4.1. Background

Vaccination is a fundamental tool in a strategy to control and eradicate major emerging diseases. Vaccine and diagnostic banks improve the feasibility of emergency vaccination by guaranteeing supplies if there is no immediate alternative and/or by bridging the period until a Member State can purchase its own vaccines and diagnostic tests.

The European Commission launched an external evaluation to review its animal health policy in 2005. Based on the results of this evaluation, strategic aims and objectives for animal health were set out in the Commission Communication on the new EU Animal Health Strategy[1] where *'Prevention is better than cure'* and its Action Plan[2] respectively, which cover the period 2007–2013. The action plan is structured around four main pillars or areas of activity:

1. Prioritisation of EU intervention;
2. The EU Animal Health framework;
3. Prevention surveillance and preparedness;
4. Science, Innovation and Research.

Identifying problems before they emerge while being ready to manage major animal disease outbreaks and crises is one of the expected outcomes of the Animal Health Strategy. This is an essential component of Pillar 3 of the new strategy.

Under Pillar 3 in particular, as point 24 of the programming document[3] for the Action Plan, a task force was created to assist the Commission in the development of this policy paper on EU vaccine/antigen banks for major animal diseases such as foot-and-mouth disease (FMD), classical swine fever (CSF), avian influenza (AI) and others. Such banks should be available in emergency situations or major crises.

It is widely agreed that in many cases, the best means of combating animal diseases once they occur is in accordance with the principle that 'vaccination is better than unnecessary culling'. This has also been confirmed by opinions from the other European institutions and stakeholders during the drafting of the Strategy and its Action Plan.

Emergency vaccination has to be seen in a new light, directly linked to the availability of effective diagnostic tools substantiating that vaccinated animals or the meat and meat products obtained from vaccinated animals are free from pathogens and can be traded safely.

That is why it is necessary to discuss the availability and quality of diagnostic tests when discussing vaccines. Furthermore, the issue of EU vaccine banks can only be treated in the context of a specific vaccination strategy for each major disease (e.g. FMD, CSF, AI). The establishment and maintenance of vaccine and diagnostic banks must be part of a strategic plan prepared during 'peace time', ready for an emergency.

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1 COM (2007)539 final, 19.09.2007.

2 COM (2008)545 final, 10.09.2008.

3 http://ec.europa.eu/food/animal/diseases/strategy/pillars/action_en.htm.

The document focuses mainly on diseases which have historically had a major impact in the European Union or which are considered to be major risks in future; and on vaccines and/or diagnostic tests which can or should be applied in an emergency situation. The exercise of categorising animal diseases, as provided for in the Animal Health Strategy, is of capital importance for setting priorities for intervention in the field of animal health. In addition, DISCONTOOLS[4] may be a good support tool for identifying further relevant issues (e.g. diseases for which veterinary medicines need to be developed or other means of control for certain diseases).

However, all elements concerning vaccination need further reflection in a number of fora in the coming years. Hence the present paper does not go into the area of measures that should be taken, nor does it suggest policy options to ensure the free circulation of products derived from vaccinated animals. Although this puts a brake on the use of vaccination as a tool in combating the spread of contagious animal diseases, other tools and policy options will be addressed in a broader context. Such options might include, *inter alia*, a ban on consumer labelling of products derived from vaccinated animals, effective public communication strategies regarding the harmlessness of products derived from vaccinated animals, and the conclusion of conventions on the free circulation of products derived from vaccinated animals between governments, farmers' organisations, consumer organisations and retail and trade operators. These issues remain outside the scope of this paper.

4.2. Scope of this paper

The scope of this paper is:

- Identification of the infectious diseases for which vaccine or antigen banks should be available in the EU in the near future;
- Conditions under which vaccination against certain infectious diseases is recommended;
- Recommendations for vaccination strategies under emergency situations;
- The use of vaccines as part of DIVA-strategies[5];
- Estimates of size and costs of envisaged vaccine stocks;
- Identifying the need for diagnostic banks (e.g. for particular ELISA or PCR tests);
- Recommendations for improving EU legislation on use of vaccines in emergency or endemic situations.

4.3. Criteria for vaccine banks

Council Directive 82/894/EEC of 21 December 1982 on the notification of animal diseases in the Community lists in Part A of Annex I the diseases of terrestrial animals which are subject to notification. In total, 22 infectious diseases are listed, all of which are traditionally considered to have a major impact on animal health, but also on trade or human health (zoonotic character). They are therefore used as an initial pool of diseases for this exercise.

[4] Disease Control Tools, www.discontools.eu.
[5] DIVA: Differentiating infected from vaccinated animals.

However, not all of these diseases have been judged as justifying emergency vaccine and diagnostic banks. The diseases identified so far for this strategy are: FMD, AI, CSF, AHS, BT and ASF.

The criteria identified for having a vaccine and/or diagnostic bank are:

- That vaccination against the particular disease is a possible and effective control tool to protect animals from infections and prevent the spread of the disease;
- That vaccination can be part of a holistic strategy, including other methods to control and eradicate the disease;
- That vaccination is a realistic and cost-effective strategy compared to others based on non-vaccination;
- That trade issues are resolved regarding the vaccinated animals or the fresh meat and meat products obtained from vaccinated animals;
- That adequate vaccines or antigens and the means to vaccinate large numbers of animals are available, or could be made available for a programme to control the infection at short notice.
- That adequate diagnostic tools and/or surveillance systems are in place to substantiate that vaccinated animals are free from the infectious agent against which the vaccine has been used;
- That there is a legal basis for emergency vaccination;
- That there is a legal basis for the vaccines/antigens bank, including testing procedures for antigens/vaccine and the emergency release of vaccines;
- That there are surveillance systems in place to ensure the closest possible vaccine match where pathogens tend to antigenically diversify (early warning); and administrative and legal procedures to allow rapid adaptation of vaccine variants to the specific epidemiological situation.

4.4. Conditions for emergency vaccination

Emergency vaccination, supported where necessary by a vaccine and diagnostic bank, is needed in an emergency situation when an infectious agent is introduced under circumstances with potential for rapid spread and significant damage, such that a policy of non-vaccination risks failure or would require massive resources and/or culling of animals. Conditions under which emergency vaccination is recommended include, for example:

- Where infection has occurred in or threatens an area with a relatively high-density population of vulnerable animals;
- Following multifocal introduction of infection, or where infection has not been rapidly detected and controlled, leading to multifocal spread;
- Where there is a high risk of uncontrollable spread of infection, for example by the airborne route;

- Where there is inadequate capacity or resources for control by non-vaccination, or where such measures are considered economically, socially or ethically unacceptable; or not practically feasible for any other reason;

- Where there is a significant risk of a (potential) zoonotic agent spreading from animals to humans or vice versa.

4.5. Alternatives to vaccine banks

The probability and impact of certain exotic diseases affecting the EU's livestock populations and spreading may be too low to justify setting up a ready or near-ready stock of vaccines or antigens, though this would be technically feasible. A less costly, but still beneficial alternative might be to provide funds to establish specific vaccine master seed-stocks, from which vaccines could be produced and deployed more quickly than if starting from scratch.

However, a number of regulatory and legal issues would have to be addressed to ensure that vaccine master seed-stocks could become a useful additional tool to combat such incursions. Without substantial amendment of existing legislation on veterinary medicinal products, it is difficult to envisage a system of common master seed-stocks which could be used as a source for the rapid manufacture of authorised veterinary vaccines for emergency use. For example, master seed-stocks would have to be tested according the *Pharmacopoeia Europaea* (Ph. Eur.) requirements to ensure they were free of potential contaminants.

An interesting regulatory concept that has been introduced for human influenza vaccines is that of a 'mock up' authorisation. The applicant company chooses a strain that is a good candidate for a future outbreak, provides the formulation data, the selected dose and very limited clinical data. Once this has been reviewed by regulators, the company receives a 'mock up' licence authorising use of the product only in case of an epidemic of that specific strain. If an epidemic occurs with a different strain, the licence can be activated following the rolling review concept, where regulators assess data with the new strain as they become available.

4.6. Recommendations for particular vaccine and diagnostic banks

4.6.1. Foot-and-mouth disease (FMD)

- There is a continuing need for an EU FMD vaccine bank, containing stored antigens in sufficient quantity to provide up to 5 million doses per strain, depending upon the level of risk.

- The selection of vaccine strains to be represented should be based on risk assessment informed by up-to-date knowledge of the global distribution of FMDV serotypes and strains and of the likelihood of their spreading to the EU.

- Industry could be contracted to prepare vaccine seed-stocks to cover strains of lower perceived risk, but regulatory procedures add considerably to the time needed before such seed-stocks could be turned into final product.

- Efforts should be made to harmonise procedures and reach agreement on sharing of vaccine antigens or formulated vaccines between vaccine banks of Member States,

including the EU Bank. This could give access to greater quantities of vaccine, for more diverse strains.

- The Commission should continue to support research efforts intended to produce more potent and more thermostable FMD vaccines with more rapid onset of protection, improved DIVA properties and broader strain coverage.

- A bank of pen-side FMDV-detection systems should be established, providing rapid access to 500 lateral flow devices and 5 portable nucleic acid detection systems.

- A commercial supply of DIVA serological test kits should be established, to enable at least 2.5 million animals to be tested at short notice.

- There is a need to review the supply of serotype-specific serological test kits to see if diagnostic reagent banks are also warranted for these assays. This could be undertaken by Commission services with technical assistance from the European Union Reference Laboratory for FMD and the EU FMD Research Group.

4.6.2. Avian Influenza (AI)

- The only scenario where emergency vaccination might be applied is in an area at risk of HPAI because of a neighbouring infection. It is difficult to foresee the possible application of metaphilactic vaccination in an area where a HPAI virus is already actively circulating, due to the rapid spread of infection and the time needed to induce an adequate level of immunological protection in a large poultry population at high risk of AI.

- Recommendations on which AI vaccines are most likely to be required for emergency vaccination may be provided by the OIE/FAO Network of AI Reference Laboratories. National and Regional AI vaccine banks, including those of individual EU Member States and of the EU, should be advised on vaccine strain selection by the National and the European Union Reference Laboratory respectively.

- It is recommended to have stocks of vaccine available for two H5 viruses and two H7 viruses from the Eurasian ‘lineage’, possessing different neuraminidases (N) subtypes. The establishment of a stock that includes a bivalent (H5 and H7) inactivated vaccine should be considered to improve efficiency. The option of stocking H9 vaccine should also be evaluated, as it is considered a potential candidate for a human pandemic.

- A minimum number of doses should be established, based on the number needed to sustain an emergency vaccination programme for at least three months in areas with the highest poultry population densities. Therefore a minimum of 7-8 million doses are needed.

- Taking into account the need to store vaccine strains of at least four virus subtypes to perform heterologous vaccination, the size of the AI vaccine bank should be around 30–40 million doses. This could be reduced if a bivalent (H5 and H7) inactivated vaccine were available.

- Under certain epidemiological circumstances, the vaccine bank should also be available to control LPAI outbreaks.

- The relatively short shelf life of inactivated AI vaccines (12-24 months) needs to be taken into account in planning for an AI vaccine bank. This problem may be overcome by applying the principle of an antigen bank, or by applying the rolling stock principle.

- AI viruses appear to be evolving antigenically. Constant monitoring of antigenic characteristics of circulating AI viruses by testing new virus isolates is recommended.
- The availability of a diagnostic bank with a suitable discriminatory test is not an essential pre-requisite to implement rapid emergency vaccination, if sentinel birds are part of the strategic programme. However, the European Union Reference Laboratory for AI should provide recommendations on how a suitable antibody ELISA could be made available in due time should emergency vaccination be necessary.

4.6.3. Classical swine fever (CSF)

- Emergency vaccination should be accepted as an important and valuable tool for the control of CSF in wild boar and domestic pigs. Strategic programmes for emergency vaccination should become part of the contingency plans of Member States with a high pig density.
- To allow efficient and timely emergency vaccination, an EU vaccine bank with a live attenuated vaccine (e.g. C-strain) is needed, enabling vaccination of at least 2 million pigs.

- Alternatively, the storage of the E2-subunit vaccine as an antigen should be tested and evaluated by the European Union Reference Laboratory for CSF or by other appropriate mechanisms. Providing the evaluation is positive an antigen bank should be implemented.

- A diagnostic bank for an E^{RNS}-marker ELISA is needed, together with the marker vaccine bank. Test kits for not less than 50 000 pigs should be available within seven days of a request. The availability of a diagnostic bank with a suitable PCR test is not an essential pre-requisite.

- Practically oriented screening schemes are needed to identify infected animals in a post-vaccination area.
- A properly designed and implemented emergency vaccination strategy together with a targeted search for chronically-infected animals in vaccinated herds during final screening would mean a lower risk for fresh meat than a conventional non-vaccination strategy.

- If one of the new prototype modified live marker vaccines is licensed, it should be included in the vaccine bank together with the appropriate diagnostic tests.

4.6.4. African horse sickness (AHS)

- A vaccine bank with live attenuated vaccines against serotypes 2, 4 and 9 of AHSV should be established. However, other serotypes, e.g. serotype 5, should be considered for future planning of vaccine banks.

- If one of the new prototypes of recombinant vaccines is licensed, it should be included in the vaccine bank as a priority.

- The vaccine bank should contain a minimum of 150.000 doses for each of the proposed serotypes.
- In addition, developing a working seed bank for all nine serotypes of the virus is recommended, to cover the first steps for developing an inactivated vaccine.

- The availability of a diagnostic bank with a suitable test is not an essential pre-requisite.
- After establishing the initial stock, continuous monitoring of the epidemiological situation of AHS in the countries geographically close to the EU is recommended, to identify serotypes which might become risks for EU livestock, and so adjust the development and procurement of vaccines against those serotypes.

4.6.5. Blue tongue (BT)

- Establishing vaccine seed-stocks for BTV serotypes not currently present in the EU is recommended.

- The vaccine seed-stocks should enable the production of enough inactivated vaccine to provide up to 5 million doses for each of the proposed serotypes.
- Establishing continuous monitoring of the epidemiological situation of BT in countries geographically close to the EU is recommended to identify serotypes which might become risks for EU livestock, so as to adjust the development and procurement of vaccines against those serotypes.
- The availability of the BT diagnostic tests is not a problem; therefore a diagnostic bank is not needed.

4.6.6. African swine fever (ASF)

- No vaccines exist against ASF.

- A diagnostic bank should be established with antibody ELISA test kits for testing not less than 100 000 pigs.

4.7. General issues related to the vaccines industry

To establish a vaccine or antigen bank, industry needs to respond to a relevant tender. To market authorised vaccines in the EU, the size of the vaccine/antigen bank, the price per dose, the shelf life of the vaccine or antigen and renewal plans are important issues. For vaccines that are not yet authorised in the EU, the existence of a vaccine elsewhere in the world, the need to start a development programme, and standards relating to safety, quality and efficacy need to be considered.

A cost-efficient method to make available a vaccine bank rapidly is to deploy 'rolling stock' where a company has ongoing production and increases its reserve stock by the quantity of the tender. The advantage of this is that emergency vaccine is manufactured, tested and released in accordance with European legislation and can be distributed quickly.

Consideration needs to be given to the merits of creating an antigen bank rather than a vaccine bank. An antigen bank has many advantages over vaccine banks, and is especially suitable if the vaccine has a short shelf life, or in case of diseases with antigenic variance, such as FMD, AI, etc where the formulation of the vaccine can be decided once the field virus has been typed.

The only disease for which there is a regulatory framework within the EU for rapid release of vaccine from pre-tested antigen is FMD (Ph. Eur. Monograph 0063). To apply the principle of a vaccine or antigen bank for diseases other than FMD, appropriate regulatory aspects need to be addressed in EU legislation.

The vaccine industry needs a financial incentive to develop a vaccine against a new disease or new serotype. If there is no vaccine against an emerging disease and there is no existing market for such a vaccine, there is no such incentive. For such emerging diseases, public funding is recommended for vaccine development, along with a commitment for an antigen/ vaccine bank linked to this funding.

Relevant legislation regarding veterinary medicinal products is not well suited to approving the use of vaccines in emergency situations. The current review regarding legislation dealing with veterinary medicinal products is an ideal opportunity to introduce a mechanism for approving vaccines for emergency use at European level.

4.8. Overview of proposals for vaccine and diagnostic banks

DISEASE	Vaccine Bank		Diagnostic Bank	
	Type	**Size**	**Type**	**Size**
FMD	Selection of vaccine strains based on up-to-date risk assessments	2.5 – 5 mil doses per strain	• Serological tests kits • Lateral flow devises • Portable PCR systems	• for 2.5 mil animals • 500 • 5
AI	H5 and H7 strains (selection of vaccine strains based on recommendation of OIE/FAO Network of AI	7 – 8 mil doses per strain	Not needed	
CSF	• Live attenuated vaccine • E2 subunit vaccine	2 mil doses	• Not needed • E^{rns} ELISA	• for 50.000 animals
AHS	Against serotypes 2, 4 and 9	150.000 doses per serotype	Not needed	
BT	Vaccine seed-stocks for the BTV serotypes not currently present in the EU.	5 mil doses of final vaccine per serotype	Not needed	
ASF	No vaccine available		Antibody ELISA	for 100.000 animals

5. MAIN REPORT

5.1. Issues and background

Rigorous and rapid measures must be taken to interrupt the chain of infection during the emergency of major contagious disease outbreaks. Such measures may include the pre-emptive culling of animals suspected of being infected or contaminated due to their direct or indirect contacts with a confirmed outbreak. In the past this situation has led to the slaughter of a high number of animals in which the infection could not be confirmed post-mortem or on the basis of conventional diagnostic tests on samples taken at the time of slaughter.

Emergency vaccination has to be considered as one tool in a range of measures as part of a complex strategy. In the past, use of vaccines in emergencies was limited by the possibility that vaccinated animals might spread the virus further. Moreover, infected animals could not be easily and rapidly identified or distinguished from vaccinated but uninfected animals.

Emergency vaccination to control infectious animal diseases can be implemented either as a ***vaccination-to-live*** or ***vaccination-to-kill*** strategy. Protective vaccination or v***accination-to-live*** means that vaccinated animals are kept to the end of a normal production cycle and their meat eventually marketed. Suppressive vaccination or ***vaccination-to-kill*** means, that animals around an infected farm are vaccinated to reduce the spread of infection and to gain time, but that they will eventually be destroyed later.

Regarding the relevance and use of vaccine banks in the past, it was noted that although significant funds were spent on these, they were not always used during crises. The main reasons Member States have been reluctant to vaccinate, despite this being allowed by EU legislation, are: availability of appropriate vaccines; concerns over potential trade barriers by third countries; or misplaced concerns about consumer safety.

It is important that emergency vaccination be seen in a new light, directly linked to the availability of effective diagnostic tools substantiating that vaccinated animals, their meat or meat products obtained from them are free from pathogens and can be traded safely.

The slaughter and destruction of very large numbers of animals gives rise to considerable public concern, particularly for diseases that do not pose a risk for human health. It has also led to very high costs and losses for the Community budget, Member States, stakeholders and ultimately for consumers. For example, in the UK foot-and-mouth disease (FMD) outbreak in 2001, over 4 million animals were killed. This cost the public sector over 4.5 billion EURO, and the private sector over 7.5 billion EURO (NAO, 2002). The classical swine fever (CSF) epidemic in the Netherlands in 1997/1998 also had a major impact in economic losses and illustrates the ethical dimension of the disease. Direct losses amounted to 2.3 billion EURO. More than 12 million pigs had to be destroyed, of which fewer than 10% were directly affected by the disease. During the last decade, large-scale culling of pigs due to CSF was also conducted in Germany, Belgium, Italy, Spain, Austria, Czech Republic, Slovakia and Romania.

Emergency vaccination in a crisis situation is increasingly seen as complementary to other zoosanitary measures, and as a means to reduce reliance on culling alone. There have been significant advances in the development of diagnostic tests and modern vaccines, and new

techniques that enable detection of an infected herd reliably within a short time so that preventive culling can be minimised.

Real Time Polymerase Chain Reaction: rRT-PCR; marker Immunoassay: marker ELISA and vaccines as part of a genetic or conventional DIVA strategy (***D**ifferentiating **I**nfected from **V**accinated **A**nimals)* mean that a **change in control and eradication policy is now feasible**.

The European Commission launched an external evaluation to review its animal health policy in 2005. Based on the results, strategic aims and objectives for animal health were set out in the Commission Communication on the new EU Animal Health Strategy[6] where ***'Prevention is better than cure'*** and its Action Plan[7] respectively, which cover the period 2007–2013. The action plan is structured around four main pillars or areas of activity:

1. Prioritisation of EU intervention;
2. The EU Animal Health framework;
3. Prevention surveillance and preparedness; and
4. Science, Innovation and Research.

Identifying problems before they emerge, while being ready to manage major animal disease outbreaks and crises is one of the expected outcomes of the Animal Health Strategy and is an essential component to Pillar 3 of the new strategy.

Under Pillar 3, in particular as point 24 of the programming document[8] for the Action Plan, a task force was created to assist the Commission in developing a policy paper on EU vaccine/antigen banks.

The group was asked to deliver a paper on the necessity of vaccine banks for major animal diseases such as FMD, CSF, avian influenza (AI), to be made available in emergency situations or major crises. Furthermore, the task force was asked to give recommendations on the strategic use of vaccines, including their use in combination with diagnostic tests (e.g. DIVA-strategy) to prove that animals are free from pathogens, thus making normal slaughter or trade possible.

It is widely agreed that the best means of combating animal diseases once they occur is to proceed in accordance with the principle that 'vaccination is better than unnecessary culling'. This was also confirmed by opinions from other European institutions and stakeholders during the creation of the Strategy and its Action Plan.

It is also certain that animal health risks are growing due to increasing global mobility, growing international trade and climate change, hence the need for an adequate emergency vaccination strategy for both existing and emerging diseases. Part of that strategy could be the increased use of (both suppressive and protective) emergency vaccinations as rapid containment in disease eradication operations. To enable this, and to cut down the time needed to make available vaccinations when needed, EU vaccine banks need to be expanded. Rapid deployment and use of vaccines could help to reduce the number of healthy animals slaughtered and destroyed.

Animal health is closely linked to human health, and important in economic terms. Moreover, animal welfare is increasingly seen as common sense in the EU. Animals are sentient beings.

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6 COM (2007)539 final, 19.09.2007.

7 COM (2008)545 final, 10.09.2008.

8 http://ec.europa.eu/food/animal/diseases/strategy/pillars/action_en.htm.

Their protection and humane treatment is one of the challenges for a cultured and civilised 21st century Europe, as endorsed recently by the Treaty of Lisbon.

Unfortunately, markets both within and outside the EU may not accept products from (emergency) vaccinated animals, even though they are as safe for consumption as those derived from unvaccinated animals. Livestock farmers and other operators need sufficient guarantees that products from vaccinated animals are marketable without price reductions. This is a crucial issue which the EU must resolve to guarantee the free movement of goods while encouraging the use of vaccination.

However, this paper does not deal with issues concerning the free circulation of products derived from vaccinated animals. Although they put brakes on the use of vaccination as a tool in combating the spread of contagious animal diseases, other tools and policy options will be addressed in a broader context elsewhere, and remain outside the scope of this paper. They include, *inter alia*, a ban on consumer labelling of products derived from vaccinated animals, effective public communication strategies regarding the safety of products derived from vaccinated animals, and the conclusion of conventions on the free circulation of products derived from vaccinated animals between governments, farmers' organisations, consumer organisations and retail and trade operators.

Similarly, the relationship between increased use of vaccination and the long-term sustainability of the keeping and production of live animals and products of animal origin is also complicated. This too will be addressed in more detail at other fora. The contributors to this paper simply wish to acknowledge that there are many possible synergies between these areas and stress the need for further reflection.

5.2. Scope of policy paper

The scope of this policy paper is:

- Identification of infectious diseases for which vaccine or antigen banks should be available in the EU in the near future;
- Conditions under which vaccination against certain infectious diseases is recommended;
- Recommendations for vaccination strategies under emergency situations;
- The use of vaccines as part of DIVA-strategies[9];
- Estimates of size and costs of envisaged vaccine stocks;
- Identifying the need for diagnostic banks (e.g. for particular ELISA or PCR tests);
- Recommendations for improving EU legislation on use of vaccines in emergency or endemic situations.

[9] DIVA: Differentiating infected from vaccinated animals.

5.3. Criteria for having vaccine and antigen banks

Vaccination is a fundamental tool in a strategy to control and eradicate major emerging diseases. Vaccine and diagnostic banks improve the feasibility of emergency vaccination by guaranteeing supplies where there is no immediate alternative and/or by bridging the period until Member States can purchase their own vaccines and diagnostic tests. Setting up and maintaining vaccine and diagnostic banks must be part of a strategic plan prepared during 'peace time' to be ready in case of emergency. To be effective, emergency vaccination must:

- Be feasible and effective as a control tool against a particular disease to protect animals from infections and prevent their spread;
- Be part of a holistic strategy including other methods to control and eradicate the disease;
- Be a realistic and cost effective strategy compared to others based on non-vaccination;
- Be part of a context in which trade issues are resolved regarding vaccinated animals or fresh meat and meat products obtained from them;
- Be available at short notice: adequate vaccines or antigens and the means to vaccinate large numbers of animals must be available for launching a control programme to interrupt the chain of infection;
- Be coupled with adequate diagnostic tools and/or surveillance systems, in place to substantiate that vaccinated animals are free from the infectious agent against which the vaccine has been used;
- Be covered by legal bases for emergency vaccination;
- Be covered by a legal basis for the vaccines/antigens bank, including testing procedures for antigens/vaccine and emergency release of vaccines;
- Be implemented with surveillance systems to ensure the closest possible vaccine match where pathogens tend to antigenically diversify (early warning), with administrative and legal procedures to allow rapid adaptation of vaccine variants to the specific epidemiological situation.

In view of the above, the issue of vaccine banks can only be treated in the context of a vaccination strategy specific for each major animal disease outbreak (e.g. FMD, CSF, AI). Furthermore, vaccines should in future be used only in tandem with diagnostic tools and surveillance systems to substantiate that vaccinated animals are free from the infectious agent when control measures are lifted. That is why the availability and quality of diagnostic tests must be considered when vaccines are discussed.

5.4. Conditions for emergency vaccination

Emergency vaccination, supported where necessary by a vaccine and diagnostic bank, is needed in an emergency situation when an infectious agent is introduced with potential for rapid spread and significant damage, such that non-vaccination would risk failure or require massive resources and/or culling. Emergency vaccination is recommended:

- When infection has penetrated or threatens an area with a relatively high density of susceptible animals;

- Following multifocal infection, or where infection has not been rapidly detected and controlled, leading to multifocal spread;

- Where there is a high risk of uncontrollable spread of infection, for example by the airborne route;

- Where non-vaccination is not practically feasible, for reasons including inadequate capacity or resources, or is seen as economically, socially or ethically unacceptable;

- Where there is a significant risk of a (potential) zoonotic agent spreading from animals to humans or vice versa.

Vaccination to protect rare breeds, zoological animal collections and other animals representing valuable genetic resources might also be considered.

5.5. Identification of infectious diseases for which vaccine, antigen and diagnostic banks should be available in the EU in case of an emergency

Council Directive 82/894/EEC of 21 December 1982 on the notification of animal diseases in the Community lists in Part A of Annex I the diseases of terrestrial animals which are notifiable. In total, 22 infectious diseases are listed, all of which are traditionally considered to have a major impact on animal health but also on trade or human health (zoonotic character). They are therefore used as an initial pool of diseases for this exercise. However, not all of them justify establishing an emergency vaccine and diagnostic bank. Those which have been identified so far for such action are: FMD, AI, CSF, AHS, BT and ASF.

Table 2 in the Annex gives an overview of the infectious diseases for which vaccine, antigen and diagnostic banks should be available in the EU for an emergency.

The document focuses mainly on diseases which have had a significant impact historically in the European Union or which have such potential in future. Such prioritising and categorising is vital in the Animal Health Strategy. DISCONTOOLS[10] may be a good support tool for identifying other relevant diseases.

5.6. Alternatives to Vaccine banks

The probability and likely impact of certain exotic diseases affecting livestock in the EU may be too low to justify setting up ready or near-ready stocks of vaccines or antigens, though this would be technically feasible. Providing funds to establish specific vaccine master-seed stocks from which vaccines could be produced and deployed more quickly than if starting from scratch might be less costly, but still beneficial.

For instance, there are 24 bluetongue serotypes that do not cross-protect against one another. Recent events have shown that it is hard to predict which 'new' serotype may be the next to arrive in Europe. Maintaining a vaccine bank for them all would be extremely costly. Experience has shown that once new serotypes arrive, they spread for months and even years.

[10] Disease Control Tools, www.discontools.eu.

So a vaccination policy can be beneficial, even if it cannot be put into immediate effect. Cutting down the time needed to produce the right vaccine could be helpful in bringing the disease under control and mitigating losses. The same may be true for other diseases. Even for those that spread rapidly, such as FMD, ensuring that master-seed stocks are available to combat strains less likely to occur might be beneficial in case of prolonged outbreaks. However, a number of regulatory and legal issues need to be addressed to ensure that vaccine master-seed stocks can become a useful additional tool to combat disease incursions. Establishing master seed stocks of disease agents without initiatives in other areas will not facilitate the rapid production of emergency vaccines should the need arise.

Providing industry with a source of accessible master seeds for new vaccines is a useful step, but only one of many in developing and manufacturing vaccines for emergency use.

Master-seed stocks should be tested according the Ph.Eur requirements to ensure they are free of potential contaminants. Furthermore, master seeds need to be adapted to the manufacturer's specific production conditions. Seed viruses may need to be further passaged to adapt them for growth in the cells and/or media used in production. Yields of antigen need to be established under the manufacturer's proposed conditions. Specifications for in-process tests and final quality controls need to be established and validated.

The formulation of the final vaccine needs to be defined, including any adjuvant used to increase its immunogenicity. The stability of the product needs to be established by real-time studies, usually for up to three months beyond the proposed shelf-life.

Specific safety and efficacy studies should be performed with a representative batch of the product under laboratory and field conditions in the target species to establish the indications and claims for the product.

Without substantial amendment of existing legislation on veterinary medicinal products, it is difficult to envisage a system of common master-seed stocks which could be used as a source for the rapid manufacture of authorised veterinary vaccines for emergency use.

An alternative would be to use targeted EU funds to develop authorised vaccines for emergency use in collaboration with manufacturers. This could yield vaccines that industry would otherwise have little incentive to develop. It would also ensure that appropriate quality, safety and efficacy tests are conducted. Master-seed stocks held by specific manufacturers could then be a reserve for the production of reserve antigens or vaccine under emergency conditions.

From the regulatory perspective, there is currently no scope for the concept of master seed authorisation alone. Even in cases where regulatory requirements have been minimised to facilitate the timely use of authorised vaccines against epizootic diseases such as AI and BT in the EU, data on manufacturing methods, minimum antigen composition, production and control of active ingredients and certain safety and efficacy laboratory studies are still required. The demonstration of a correlation between antigen content and efficacy is considered critical for such authorisation.

So the use of authorised master seeds alone is not an option in the current regulatory framework. There would have to be a number of changes to the relevant legislation to enable such an approach.

An interesting regulatory concept that has been introduced for human influenza vaccines is that of a 'mock up' authorisation. The applicant company chooses a strain that is a good candidate for a future outbreak, provides the formulation data, the selected dose and very limited clinical data. Regulators review this, and the company receives a 'mock up' licence. This allows the use of the product only for an epidemic of the specific strain. If an epidemic occurs with a different strain, the licence can be activated following the rolling review concept. This enables regulators to assess data with the new strain as they become available. When the package of information is ready, the authorisation process is very brief (3-5 days) and is considered a variation. The level of clinical data requested depends on the risk/benefit of vaccination in any given outbreak. Moreover, for human seasonal influenza, there is a highly developed network of laboratories that provides seed strains for the preparation of seasonal flu vaccines. There is no similar network for veterinary vaccines.

6. FOOT-AND-MOUTH DISEASE (FMD)

6.1. Severity/likelihood

Foot-and-mouth disease (FMD) is caused by an RNA virus, FMD virus (FMDV), within the family *Picornaviridae*. FMDV has a wide host range, including domesticated and wild ruminants and pigs. The severity of infection varies greatly according to the species, breed and age of the host. Dairy cattle and pigs are worst affected, while small ruminants may show mild or unapparent infection. Fatalities are rare, but can follow myocarditis in young animals. FMD is not a significant zoonosis. The disease is highly contagious, and may be spread by a variety of direct and indirect means, including trade in animals and animal products. This, along with the occurrence of multiple serotypes and subtypes that cross-protect incompletely or not at all, makes control of FMD very difficult. FMDV is unevenly distributed worldwide. Developed countries have found it intolerable to modern intensive farming practices and have eradicated the virus, while poor countries lack the resources and infrastructure to do this. The virus remains endemic in large parts of Asia and Africa and in some South American countries. Meanwhile, FMD-free countries restrict trade with infected countries to try to prevent introduction of the virus. EU Member States have eradicated FMD, have official FMD-free without vaccination status from the OIE, and rely in the first instance on measures to prevent infection. Therefore, there is no ongoing vaccination to maintain immunity. However, the threat of introduction remains, and a number of outbreaks have occurred in the last 30 years (Valarcher et al., 2007). By far the largest and most costly was the 2001 epidemic that centred on UK with spread to Ireland, France and the Netherlands.

6.2. FMD vaccines

Conventional, inactivated FMDV vaccines are produced by growing virulent virus in very large quantities in cell culture, then separating the virus-rich supernatant from the remaining cellular debris. The virus supernatant is inactivated twice with aziridines (usually binary ethyleneimine) and the inactivated virus suspension can be further purified to separate virus from the viral non-structural proteins (NSP) that are necessary for virus replication but are not part of the virus icosahedral structure. At this point, the inactivated virus particles (also known as virus antigen) can be mixed with an adjuvant and excipients, and formulated into final vaccine, or they can be concentrated and stored deep-frozen above liquid nitrogen as an antigen bank. The frozen antigens can be thawed, and formulated into vaccine for emergency or routine use. Deep-frozen antigen is stable for at least five years, while formulated vaccine has a shelf life of up to two years if kept refrigerated (Lombard and Fuessel, 2007).

Conventional inactivated vaccines have been used for many years to control FMD and were instrumental in eradicating the disease from Europe. The OIE Diagnostic Manual provides guidance on minimum production standards and quality control procedures. Several billions of doses are used annually throughout the world, but only a limited number of producers (three in Europe) can meet the stringent quality requirements of EU Member States[11]. The three European producers can meet most of the vaccine strain requirements of EU Member States.

However, the emergence of new strains of the virus, along with changes in viral distribution, give rise to new threats and altered or novel vaccine strain requirements. Under current regulatory requirements, it may take up to six months to prepare and validate new stocks of vaccine antigen from existing master-seed stocks, and at least a year if there is a requirement to develop a completely new vaccine strain. Furthermore, existing vaccine seed strains do not provide optimal coverage against the full spectrum of known strains of FMDV. For example, in parts of Africa, there is no market, hence no incentive to prepare vaccines tailored to local strains of FMDV. So no equivalent vaccine seed virus strains are available for inclusion in European vaccine banks.

The protection induced by FMD vaccines takes at least four days to develop, is of relatively short duration, and is only maintained by administering booster doses at regular intervals of six to 12 months. Primary vaccination courses for animals usually consist of two doses, separated by a month to achieve maximum protection. Due to antigenic variation between and within FMDV serotypes, vaccine-induced protection is type-specific and sometimes relatively strain-specific. The potency and cross-protectiveness of vaccines can be increased up to a certain level by increasing the antigen content in each dose. For emergency situations, vaccines may be formulated at 6 rather than 3 PD_{50} so that protection can be conferred rapidly after administration of a single dose.

Research to develop new, improved FMD vaccines is underway in Europe and elsewhere. In the U.S., adenovirus vectored vaccines may become commercially available within the next five years. Their potential advantages are speed of onset of protection, with a reduced risk for FMDV escape during production or from incomplete inactivation. Another promising line of research is the development of recombinant empty capsids, which may have enhanced stability and can be produced without the need to handle live FMDV. Other avenues of research are also being explored.

When vaccination is used during an outbreak, vaccinated animals are likely to be exposed to infection. The degree of vaccine-induced protection will depend on many factors, including the potency of the vaccine, the degree of antigenic match between the vaccine and the challenge virus, the interval between vaccination and challenge, and the weight of challenge. Consequently, vaccinated animals may not always be protected from disease. Even if protected, they may become subclinically infected and shed virus. Serological and other tests can be used to help detect vaccinated and infected animals that show minimal disease. The serological tests rely on the fact that replicating virus, but not immunisation with purified vaccines, elicits a measurable antibody response to the viral NSPs. An advantage of NSP

[11] To ensure the consistency of each batch of vaccine, and to guarantee its quality, safety and efficacy, EU manufacturers are required to produce vaccines in accordance with the rigorous principles of Good Manufacturing Practice (GMP) and validate each stage of production as well as the in-process and final product tests used to control the vaccine.

serology tests is that they are not serotype specific, so a single test can detect antibodies induced by all serotypes of FMDV. The OIE Manual provides details of recommended FMD DIVA tests. The OIE Terrestrial Animal Health Code provides guidance on FMD surveillance, including approaches to substantiate FMD-free status, encompassing use of NSP serology after vaccination.

6.3. FMD vaccine banks

FMD vaccine antigen banks are held by a number of different EU Member States, as well as by certain other countries elsewhere. Some countries have collaborated to establish regional vaccine banks, notably the North American vaccine bank and the European Community vaccine bank. Some of these banks have been activated to support emergency vaccination outside the EU as a means of controlling FMDV incursions (Balkans, 1996, North Africa 1999, Turkey 2000, Japan 2000, Turkey 2003, Turkey 2006, Iraq 2009). However, vaccination using vaccine from antigen banks has not been used in EU Member States. This can be attributed partly to (old) trade rules that involved a prolonged delay in regaining FMD-free trading status after vaccination, and partly to a lack of DIVA testing strategies. Trade rules are now more favourable for emergency vaccination, and DIVA test strategies better developed.

A European Coordinated Action project funded under FP6 established a grouping of FMD vaccine bank managers with a view to exchanging information and best practice with one another and with industry. This could lead to improved and harmonised vaccine standards and ultimately to the sharing of vaccines, leading to savings and increasing the quantity and strain diversity of vaccines available.

Recommendations on which FMD vaccines are most likely to be required for emergency vaccination are provided by the FMD World Reference Laboratory and the OIE/FAO Network of FMD Reference Laboratories[12]. These also provide field isolates for the development of new vaccines. National and Regional FMD vaccine banks, including those of individual EU Member States and of the EU, are advised on vaccine strain selection by national experts and by the European Community Reference Laboratory respectively, and through consultation with industry concerning the most up-to-date strains available.

The European Commission for the Control of FMD (EUFMD) conducts regular surveys of vaccine antigen stocks held by EU Member States. In recent years, results have been treated as confidential[13], due to concerns that release of information could be helpful to potential bioterrorists, who might plan to deliberately introduce a FMDV for which no vaccine was available.

The Standing Technical Committee of the EUFMD (the EUFMD Research Group) has produced a position paper on the amount of vaccine of a given strain needed in the European Vaccine Bank. The recommendations were that the EU should have at least 2.5 million doses

[12] http://www.wrlfmd.org/ref_labs/fmd_ref_lab_reports.htm.

[13] Recommendation in report of the 36th Session of the European Commission for the Control of FMD, Rome, 27-29 April 2005: 'In order to safeguard the confidentiality of antigen and vaccine bank data the results of the survey in respect of information on the virus strains and the number of dose equivalents of material stored, both in total and by individual countries, should not be openly published but should be held in safe keeping by the EUFMD Secretariat and only divulged at the written request of officially authorised, individual national authorities or official EU or FAO representatives.'.

available for each antigen strain held in the EU reserve bank. For strains circulating in neighbouring countries, at least 5 million doses should be available (Dekker et al., 2007).

6.4. Diagnostic considerations

As for other viral diseases, a range of methods are available to confirm the presence of FMDV or anti-FMDV antibodies. Where tests are required very rapidly and/or have to be scaled up quickly to handle very high sample throughputs, there may be a case for establishing diagnostic kit banks as part of emergency response plans.

Confirmation of outbreaks can be done rapidly in the laboratory by use of automated real-time RT-PCR systems able to handle relatively high sample throughputs. This approach does not depend on a small number of specialist commercial suppliers and does not require a diagnostic bank. However, pen-side virus confirmation is much quicker, but depends on the supply of test systems from commercial suppliers. To guarantee this supply, a contingency reserve should be established. This could consist of 500 lateral flow devices for FMDV antigen detection and five portable nucleic acid detection systems. The benefits of these devices have been reported elsewhere (Ferris et al., 2008; King et al., 2008) and the EUFMD research group has developed a position paper on the subject.

Serological testing may be required in a FMD emergency to confirm suspect cases, to screen for undisclosed infection, to substantiate FMD-free status after the outbreak and to validate that vaccination has been carried out effectively. The last of these is not a requirement of the current EU FMD Directive, but might be needed in future. For other purposes, large numbers of test kits may be required at fairly short notice. The most flexible testing strategy involves the use of NSP serology, since this can detect infection by any of the FMDV serotypes. It can also substantiate freedom from infection, with or without vaccination.

The validation, use and interpretation of NSP serology as an adjunct to demonstrating FMD freedom after emergency vaccination has been studied extensively within an FP6 Research Project (FMD_Improcon project of the EU 6th Framework Programme, SSPE-CT-2003-503603). There have been many publications as a result of this work. As a follow-up, three workshops were held on the subject in 2007, bringing together representatives of official veterinary services, epidemiologists and virologists from across Europe.

There have been concerns as to whether NSP testing is sufficiently sensitive and specific to be of decisive benefit in demonstrating FMD-free status after emergency vaccination. This is particularly the case where NSP testing is used to demonstrate not only freedom from viral circulation, but also risks regarding carrier animals. A problem is that vaccination reduces virus replication in infected animals, and this in turn damps down the serological NSP response. Furthermore, in the face of vaccination, the number of infected animals may also be reduced.

Considerable knowledge is now available on the performance of different NSP tests. Gaps in knowledge about how many animals may become infected after emergency vaccination and how significant a risk these actually pose for onward virus transmission are partly addressed by results obtained from serosurveys in endemic countries where intensive vaccination has been practised and by modelling studies.

Overall, we can conclude that NSP serosurveys can provide a measure of reassurance that virus infection above a certain threshold is not present in vaccinated populations, but cannot completely rule out a very low level of infection.

Such surveys need to be used in conjunction with other measures, such as providing evidence of the effectiveness of vaccination, and of the quality of clinical surveillance. Surveys should be considered as an additional safeguard in case these other measures are not fully effective, rather than as the primary method of risk mitigation. The current provisions of the EU FMD Directive need to be reviewed to ensure they are in line with the latest scientific knowledge regarding tests for substantiating freedom from FMDV infection.

The EUFMD Research Group produced a position paper after the 2001 epidemic on the need for diagnostic reagent banks to deal with future serological test requirements in the event of FMD epidemics. This concentrated on overcoming difficulties due to a lack of commercially-available test kits for serotype-specific serology that can be used in the absence of vaccination. The paper recommended that a working group of SANCO, EUFMD and WRL should prepare a tender for a European reagent bank, which could also serve as a guideline for national reagent banks. Similar issues could arise with respect to contingencies for conducting post-vaccination surveys to demonstrate freedom from virus infection, as well as surveys to demonstrate adequate vaccine-induced immunity.

Following vaccination-to-live, the current EU Directive requires that all vaccinated animals should be blood-sampled and subjected to DIVA testing to help substantiate their freedom from infection. This means diagnostic testing is needed as well as vaccine. However, it is likely that NSP tests would also be used for conducting serosurveys following control of FMD by non-vaccination strategies. Therefore, a diagnostic NSP bank would be useful for servicing both emergency vaccination and non-vaccination strategies.

Although there are several commercial suppliers of NSP serology tests, one test is currently considered superior to the others. Ongoing sales in 'peacetime' are low, and it would take time to scale up production. Furthermore, supply on a market-forces basis is never guaranteed. Therefore, at least two EU Member States have established diagnostic NSP kit reserves. In one case, an arrangement has been made to supply annually 25 ready-to-use kits (enough to test 11 000 samples) with a one-year shelf-life. In addition, a three-year contract has been agreed for stockpiling reagents for 1 million tests. Maintenance of these includes storage at ultra-low temperature and annual testing and replacement if necessary. A supply of ready-to-use kits is guaranteed within four weeks of any request and attracts a charge in addition to that of holding reserves in readiness. The annual recurring cost for holding such reserves, assuming no drawdown, is approximately 100 000 Euros.

6.5. Strategy on how the VB/DB should be used in case of emergency

EU strategy for emergency vaccination against FMD was set out in a 1999 SCAHAW paper. Policy has been reviewed since the 2001 epidemic. Methods for controlling FMD incursions are set out in the 2003 EU Directive, including the option to use emergency vaccination along with approaches needed to demonstrate FMD-free status afterwards. Member States have national contingency plans for dealing with FMD incursions, including provisions for emergency vaccination.

Critical questions in considering whether to proceed with emergency vaccination against FMDV are:

- Is vaccination likely to be a useful adjunct to prevent uncontrollable spread of the virus and likely to improve outcomes in terms of overall cost, duration of outbreaks, and numbers of animals culled?

- Is sufficient suitably matched vaccine available?

- Can logistics for applying vaccine be put in place?

- Can a plan be developed specifying what species should be vaccinated, in which locations, and in what order?

- Is there a satisfactory plan to demonstrate freedom from infection after vaccination, so that trade disruption can be minimised?

To help weigh up the cost-benefit of vaccination, a number of decision support tools have been developed, including decision trees and mathematical models. The latter attempt to predict outcomes for different control policies. Due to inherent uncertainty in input data, they are more suited for policy development rather than tactical use. At least one EU Member State has conducted a contingency planning exercise to identify regions where the characteristics of animal husbandry and density would be likely to trigger a requirement for emergency vaccination in the event of an FMDV incursion. This is extremely helpful in planning to meet this need.

6.6. Legal basis for vaccination/vaccine banks

Council decision 2009/470 of 18 June 2009 on expenditure in the veterinary field provides for Community aid to be granted to set up stocks of biological products, including vaccines, to control FMD. The establishment of Community reserves of FMD vaccine is regulated by Council decision 91/666/EEC. Commission Decision 2005/780/EC of 8 November 2005 on the purchase and storage of FMD virus antigens says the Commission shall bear the full cost which shall not exceed a maximum of EUR 2.5 million.

The use of vaccination in case of FMD is regulated by Council Directive 2003/85/EC of 29 September 2003 on Community measures for the control of FMD. Emergency vaccination can be conducted under certain conditions either as a protective vaccination or a suppressive vaccination. So far, only suppressive vaccination has been used. (The Netherlands, 2001). The use of emergency vaccination as a ***vaccination-to-live*** tool has not been used due to fear of possible trade disadvantages.

6.7. Recommendations for EU FMD vaccine and diagnostic banks

Considering the above, it is recommended that:

For vaccines

- There is a continuing need for an EU FMD vaccine bank, containing stored antigens in sufficient quantity to provide up to 5 million doses per strain, depending on the level of risk.

- The selection of vaccine strains to be represented should be based on risk assessment informed by up-to-date knowledge of the global distribution of FMDV serotypes and strains and of the likelihood of their spreading to the EU.

- Industry could be contracted to prepare vaccine seed-stocks to cover strains of lower perceived risk, but regulatory procedures add considerably to the time needed before such seed-stocks could be turned into final product.

- Efforts should be made to harmonise procedures and reach agreement on sharing of vaccine antigens or formulated vaccines among vaccine banks of Member States, including the EU Bank. This could give access to greater quantities of vaccine, for more diverse strains.

- The Commission should continue to support research intended to produce more potent and more thermostable FMD vaccines with more rapid onset of protection, improved DIVA properties and broader strain coverage.

For diagnostics

- A bank of pen-side FMDV-detection systems should be established, providing rapid access to 500 lateral flow devices and five portable nucleic acid detection systems.

- A commercial supply of DIVA serological test kits should be established, to enable at least 2.5 million animals to be tested at short notice.

- There is a need to review the supply of serotype-specific serological test kits to see if diagnostic reagent banks are also warranted for these assays. This could be undertaken by Commission services with technical assistance from the European Union Reference Laboratory for FMD and the EU FMD Research Group.

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7. AVIAN INFLUENZA (AI)

7.1. Introduction

Influenza viruses are segmented, negative strand RNA viruses that are placed in the family Orthomyxoviridae in three genera: *Influenzavirus A, B* and *C*. Only influenza A viruses have been reported to cause natural infections of birds. Type A influenza viruses are further divided into subtypes based on the antigenic characteristics of the surface glycoproteins haemagglutinin (H) and neuraminidase (N). At present, 16 H subtypes (H1–H16) and nine neuraminidase subtypes (N1–N9) have been identified. Each virus has one H and one N antigen, apparently in any combination. All subtypes and most possible combinations have been isolated from avian species.

Influenza A viruses infecting poultry can be divided into two distinct groups on the basis of the severity of the disease they cause. The very virulent viruses cause highly pathogenic (HP) avian influenza (AI), a systemic infection, in which mortality in some susceptible species may be as high as 100 %. These viruses have been restricted to strains belonging to the H5 and H7 subtypes, exhibiting a multi-basic cleavage site at the precursor of the haemagglutinin molecule. HPAI is a serious, highly contagious infection with high mortality in certain domestic birds (e.g. chickens and turkeys). Its clinical behaviour in domestic waterfowl and in wild birds is variable; it may or may not cause clinical signs and mortality. AI viruses may spread from birds to other animal species and to humans. Low pathogenic (LP) viruses generally cause a mild disease unless exacerbated by other factors (bacterial infection, etc.). LPAI of H5 and H7 subtypes are also subject to EU control legislation due to their potential to mutate to HPAI viruses.

7.2. Vaccination for AI control

To decrease the number of animals culled for the control of epidemic diseases and in response to ethical concerns including animal welfare, the EU Commission, as reported in the New Animal Health Strategy (2007-2013), has indicated the possible use of emergency vaccination as a measure to improve the control of major animal diseases, such as AI. Historically, vaccination against AI infections caused by the virus subtypes H5 and H7 has been used on several occasions to control the disease (Peyer et al., 2009). In the EU, the Commission authorised the implementation of AI vaccination programmes in poultry in Italy, the

Netherlands, France, Germany and Portugal. Preventive vaccination has also been carried out in zoo birds in 17 EU Member States in the last few years, in response to the threat of introduction of Asian H5N1 HPAI virus via wild birds.

7.3. AI vaccines

There are five general types of AI vaccines (i.e. inactivated, live, subunit, recombinant vectors expressing AI genes, and DNA vaccines), each of which has advantages and disadvantages. Although various types of AI vaccines have been tested in experimental conditions, relatively few have been licensed in industrialised countries. Traditionally, inactivated vaccines have been based on antigens produced from LPAI virus isolates. A new approach is being developed for the creation of inactivated vaccines for AI, based on the application of reverse genetics techniques (Hofmann, 2002).

There are currently four vaccines with EU-wide authorisation from the European Medicine Agency (EMEA). All are inactivated whole virus vaccines adjuvanted with mineral oil (Table 1). They have been authorised under 'exceptional circumstances' (in accordance with the minimum requirements indicated in the CVMP guidelines EMEA/CVMP/IWP/222624/2006), with conditions to carry out further studies to meet the full requirements of Directive 2001/82/EC as amended. They are subject to annual review (source: The EFSA Journal 2008, 715, 1-161). Several non-licensed inactivated AI vaccines have been used in the EU in emergency situations under the provisions of Art. 8 of Council Directive 2001/82/EC (amended by the Directive 2004/28/EC).

Conventionally, vaccines that have been used against HPAI or LPAI have been prepared from infective allantoic fluid inactivated by beta-propiolactone or formalin and emulsified with mineral oil.

Recombinant vaccines for AI viruses have been produced by inserting the gene coding for the influenza virus haemagglutinin (H5 or H7 for instance) into a live virus vector and using this recombinant virus to immunise poultry against AI. However, such vaccines have not yet been used in the EU.

The production of AI vaccines licensed in the EU is in conformity with the European Regulations (a.o. Ph. Eur.) and licensing conditions. The basic principles for producing vaccines, particularly inactivated ones, are reported in Chapter 2.3.4 of the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals.

International organisations recommend that vaccines used for AI control must be of high quality and should respect international standards and guidelines. The standards of OIE and the minimum requirements indicated by EMEA for vaccines to be used in birds against HPAI viruses (ref. Committee for Medicinal Products for Veterinary Use (CVMP) guideline EMEA/CVMP/IWP/222624/2006) should be respected.

As an example, according to EMEA guidelines, the efficacy of AI inactivated vaccines should be demonstrated in laboratory conditions, using a challenge model designed to define the onset and the duration of immunity for each of the indicated target species. The major goals must be a high degree of protection against mortality and clinical signs of disease and a significant reduction of excretion and transmission of the challenge virus.

Another important element for OIE and EMEA evaluation of AI vaccine potency is the assessment of the immune response. For a good quality vaccine, the onset of immunity should be as rapid as possible to allow its use in emergency conditions. The duration of immunity induced by the vaccine should cover the economic life of the target species.

7.4. AI vaccine bank

The efficacy of an emergency vaccination program is mainly related to its capacity to limit the initial spread of infection during the high-risk period of an epidemic and to the rapid implementation of effective field operations. This is generally related to:

- correct identification of the vaccination area, and the poultry species and production type (layers versus broilers) to be targeted;

- prompt deployment of adequate vaccines;

- rapid enforcement of appropriate complementary monitoring and control measures.

Prompt implementation of an AI vaccination plan is based on:

- vaccine availability — one of the major constraints. If stock of a suitable inactivated vaccine is not available, supply time can be four to five months from the start of production. This implies that the decision-making process must be fast-tracked (Table 2) and vaccine must be available for immediate use;

- onset of immunological response — an estimated seven to 10 days (minimum) are necessary for onset, and around two weeks are needed for immunological protection. Transmission experiments carried out in chickens demonstrated that two weeks after vaccination, virus transmission may be completely halted (van der Goot *et al*., 2005). In the field, different results are obtained in different poultry species. In turkeys, a good level of immunological protection is generally obtained after two vaccine applications with an interval of at least four weeks (4-6 weeks after primary vaccination) (Busani *et al*., 2009);

- vaccination coverage — application of inactivated vaccines is a major logistics issue. In Italy in 2002-2003, it took about 30 days to cover 70% of the targeted poultry population. (A poultry flock was considered as ‘protected’ 15 days after completion of the first vaccination). In that particular case, in one month, about 250 farms (out of a total of 360 targeted poultry holdings) and a total of 7.5 million birds were vaccinated once. This renders current inactivated commercial vaccines less well suited for this task. The outcome would have been better if effective vector vaccines, such as the Newcastle disease virus recombinant vaccine, had been available. Such vaccines would allow rapid application via sprays or drinking water.

AI viruses appear to be evolving antigenically. A constant monitoring system of the antigenic characteristics of circulating AI viruses by testing new virus isolates could be obtained through surveillance. Veterinary authorities could use information provided via surveillance to guide decision-making when establishing vaccine banks for use in avian species (Beato et al., 2009).

It would be necessary to have stocks of vaccine available for two H5 viruses and two H7 viruses from the Eurasian ‘lineage’, possessing different neuraminidase subtypes, preferably

rare ones such as N5 or N8. Setting up a stock that includes a bivalent (H5 and H7) inactivated vaccine could also be considered. The option of a stock of H9 and H1 vaccines could also be evaluated, since these viruses might be potential candidates for a human pandemic.

Recommendations on which AI vaccines are most likely to be required for emergency vaccination must be provided by the OIE/FAO Network of AI Reference Laboratories. National and Regional AI vaccine banks, including those of individual EU Member States and of the EU, should be advised on vaccine strain selection by the National and the European Community Reference Laboratory respectively.

The relatively short shelf life of inactivated AI vaccines (12-24 months) puts limits on developing a vaccine bank. This problem may be overcome by applying the principle of an antigen bank or, in the case of a vaccine bank, by applying the rolling stock principle.

The formulation of the antigen bank could be activated as per Table 4 — for example phase 1/level1.

7.5. Example of decision making steps for AI emergency vaccination

Sanitary measures applied during an AI epidemic - Phases -		
Phase 1 - Implementation of restriction measures at farm level - Establishment of protection and surveillance zones	**Phase 2** Enlargement of restriction zones (ban of restocking in large areas)	**Phase 3** Implementation of the vaccination plan

Phase	Level	Criteria		Actions
		Local/regional area	Multiregional/ national area	
0	**0**	No AI virus isolation in birds (industrial/rural poultry, wild birds)	No AI virus isolation in birds (industrial/rural poultry, wild birds)	• National monitoring plan (poultry/wild birds)
0	**1**	Isolation of H5 or H7 AI virus in wild birds	No information on virus isolation in birds (industrial/rural poultry, wild birds)	• **At local/regional area:** Intensification of surveillance in rural and free-range farms
0	**2**	See above	Isolation of H5 or H7 AI virus in wild birds	• **At multiregional/national area:** Intensification of epidemiological surveillance of rural and free-range farms
1	**0**	Isolation of H5 or H7 AI virus in the in rural sector — primary outbreak		• Outbreak notification — enforcement of eradication measures • Activation of the measures in **Phase 1** • Implementation of active surveillance in poultry reared in high risk areas (DPPA)
1	**1**	Confirmed spread to other rural farms — secondary outbreaks		• Outbreak notification — enforcement of eradication measures • Activation of the measures in **Phase 2** • implementation of active surveillance in poultry reared in high risk areas (DPPA) • activation of the formulation of the antigen bank • Asses the PVP
2	**0**	Spread from rural to industrial farms (primary outbreak)		• Outbreak notification — enforcement of eradication measures • Activation of the measures in **Phase 3** (emergency vaccination)

7.6. Diagnostic considerations

Consideration must be given to setting up licensed vaccine banks that enable a 'DIVA' (Differentiating Infected from Vaccinated Animals) approach. Sentinel birds can be used for 'DIVA' strategies. Sentinel birds are non-vaccinated birds that are kept in vaccinated flocks and routinely inspected and tested for AI infection. Alternatively, antigen-detecting tests can be used. These tests can either be based on the immunogenic detection of circulating antigen (rapid detection on test strips) or the more sensitive detection of AIV nucleic acid by RT-PCR. The latter method was put in place by several reference laboratories, and could be used as a pen-side test.

Currently, the only approach in the EU that can be applied to differentiate infected from vaccinated birds is the use of a heterologous vaccine (vaccine virus with the same H type as the field strain but a different N type: heterologous neuraminidase). With such a vaccine, the immune response to the homologous H type ensures protection, while antibodies against the neuramidase of the field virus can be used as a marker through the application of a suitable companion discriminatory test. The advantage of this method is that a vaccine bank of inactivated oil emulsion heterologous vaccines could be established.

The current immunofluorescence assay to detect antibody to 'wild type' virus has been shown to be relatively robust, specific and sensitive, but alternative systems that use automation may enhance throughput and reduce costs. A competitive ELISA recently came onto the market, developed specifically to detect antibodies to the N1 Neuraminidase. The non-structural protein 1 (NS1) of influenza virus is only produced during active replication of the virus, so detection of antibodies to this protein could be used as a 'marker' of infection, since there is no active viral replication with conventional vaccines. Therefore, both homologous and heterologous vaccination would theoretically allow the use of the anti-NS1 antibody test to be used as a DIVA tool.

The EU Diagnostic Manual for AI (Commission Decision 2006/437/CE) provides different strategies to be used to differentiate infected from vaccinated animals. The OIE Terrestrial Animal Health Code and the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals also provide guidance on AI surveillance.

7.7. Size of vaccine bank

The 2007 EFSA opinion on vaccination against avian influenza of H5 and H7 subtypes in domestic poultry and captive birds underlines that in the case of a potential outbreak of AI, vaccination of birds may reduce transmission among domestic poultry flocks, avoiding culling and any negative impact on animal welfare. EFSA recommended the implementation of good AI vaccination practices using safe and effective EU-authorised vaccines when required by the epidemiological situation, but added that their use should be defined in advance of any potential direct AI threat.

LPAI control in densely populated areas (DPPA): So far, vaccination to control LPAI infections has been limited to such areas in northern Italy (Busani et al., 2009). In this scenario, vaccine banks must be considered only for countries with a high risk of LPAI introduction and spread.

HPAI control: It is difficult to recommend vaccination in an area where a HPAI virus is actively circulating, due to the rapid spread of the infection and the time needed to induce an adequate level of immunological protection in a large poultry population at high risk. The only scenario where emergency vaccination might be justified is in an area at risk of HPAI introduction because of nearby infection (e.g. virus circulation in the Gelderse Valley, and application of AI emergency vaccination in Limburg, in the Netherlands).

The minimum number of doses should be established based on the number needed to sustain the emergency vaccination programme for at least three months in areas with the highest poultry population densities.

Based on the Italian experience, to promptly implement and sustain an AI emergency vaccination plan in a densely populated area, a minimum of 7-8 million doses are needed. Taking into account the need to store vaccine strains of at least four virus subtypes (heterologous vaccination), the size of the AI vaccine bank should be around 30–40 million doses. This could be reduced if it is feasible to include in the stock a bivalent (H5 and H7) inactivated vaccine.

7.8. Estimated costs of the vaccine bank

The average cost of a dose of an inactivated AI vaccine could be estimated at around 0.026-0.030 €.

Thus, the total cost of the AI vaccine bank could be estimated as between 780 000 € to 1.2 million €.

A potential solution might be modelled on that chosen by Norway, where two different banks were established:

1. Vaccine bank with a final product: 1 million doses (to be used in emergency situations)

2. Antigen bank: 8 millions doses by subtype at the active ingredient stage, to be formulated and released within 4-5 weeks.

A vaccine bank has the advantage of having a fully tested and released product on stock for an emergency situation. If the size of the vaccine bank is in suitable relation to the size of the manufacturer's regular supply to the market, the principle of rolling stock may be possible.

If a vaccine bank is not possible, an antigen bank may be the alternative, provided there is a regulatory basis for: setting up such a bank; testing of the antigen; and the emergency release of vaccine from the bank.

7.9. Diagnostic bank

Chapter IX of Commission Decision 2006/437/EC, approving a diagnostic manual for avian influenza, describes the monitoring systems that could be applied to guarantee that a DIVA vaccination approach is adopted, as provided for in Council Directive 2005/94/EC. Two methods have been identified for monitoring the presence of AI in vaccinated poultry flocks:
- use of sentinel birds as an alternative or a supplementary method,
- application of a companion discriminatory test.

Based on these provisions, the prompt availability of a suitable discriminatory test should not be considered as an essential pre-requisite for rapid implementation of an emergency vaccination program, as sentinel birds could be used. Nonetheless, since there are some problems managing sentinel birds, particularly their identification within large flocks, a means of making a discriminatory test available within a few weeks should be defined when implementing emergency vaccination.

Regarding the application of a heterologous neuramidase vaccine, monoclonal antibodies against the nine neuramidase subtypes of AI viruses have been produced in the framework of the EU research project FLUAID. An ELISA for N1 with regard to H5N1 diagnosis has been validated by the laboratories of the EPIZONE Network of Excellence. Recommendations on how a suitable ELISA teat could be made available in due time in case of emergency vaccination should be provided by the EU Network of AI Reference Laboratories.

7.10. Legal basis for vaccination/vaccine banks

Council decision 2009/470 of 18 June 2009 on expenditure in the veterinary field provides for Community aid to be granted to set up stocks of biological products, including vaccines, for the control of AI. The possibility for the Commission and Member States to establish reserves of vaccine against AI to be used in poultry or other captive birds in case of emergency is provided for in Council Directive 2005/94/EC.

7.11. Recommendations for AI vaccine and diagnostic banks

- The only scenario where emergency vaccination might be applied is in an area at risk of HPAI because of a neighbouring infection. It is difficult to envisage the possible application of metaphilactic vaccination in an area where an HPAI virus is already actively circulating, due to the rapid spread of infection and the time needed to induce an adequate level of immunological protection in a large poultry population at high risk of AI.
- Recommendations regarding which AI vaccines are most likely to be required for emergency vaccination may be provided by the OIE/FAO Network of AI Reference Laboratories. National and Regional AI vaccine banks, including those of individual EU Member States and of the EU, should be advised on vaccine strain selection by the National and the European Union Reference Laboratory respectively.
- It is recommended to have stocks of vaccine available for two H5 viruses and two H7 viruses from the Eurasian ‘lineage’, possessing different neuraminidases (N) subtypes. Including a bivalent (H5 and H7) inactivated vaccine should be considered to improve efficiency. The option of stocking H9 vaccine should also be evaluated, as it could be a potential candidate for a human pandemic.
- A minimum number of doses should be established, based on the number needed to sustain an emergency vaccination programme for at least three months in areas with the highest poultry population densities. Therefore a minimum of 7-8 million doses are needed.
- Taking into account the need to store vaccine strains of at least four virus subtypes to perform heterologous vaccination, the size of the AI vaccine bank should be around

30–40 million doses. This could be reduced if a bivalent (H5 and H7) inactivated vaccine were available.

- Under certain epidemiological circumstances, the vaccine bank should also be available to control LPAI outbreaks.

- The relatively short shelf life of inactivated AI vaccines (12-24 months) needs to be taken into account in planning for an AI vaccine bank. This problem may be overcome by applying the principle of an antigen bank, or by applying the rolling stock principle.

- AI viruses appear to be evolving antigenically. Constant monitoring of antigenic characteristics of circulating AI viruses by testing new virus isolates is recommended.

- The availability of a diagnostic bank with a suitable discriminatory test is not an essential pre-requisite to implement rapid emergency vaccination, if sentinel birds are part of the strategic programme. However, the European Union Reference Laboratory for AI should provide recommendations on how a suitable antibody ELISA could be made available in due time should emergency vaccination be necessary.

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8. CLASSICAL SWINE FEVER (CSF)

8.1. Introduction

Classical swine fever (CSF) is induced by infection with classical swine fever virus (CSFV), a *Pestivirus* of the family *Flaviviridae*. CSF is a disease that has caused major socio-economic damage in the EU during recent decades. Although there has been considerable progress in eradicating and preventing the disease, there is still the threat of an epidemic. The main reasons are that the CSF virus is still present in feral pigs in some Member States, and that the virus is endemic in the Balkan region, including Bulgaria and Romania. Control measures are in place for these areas, but new outbreaks in domestic pigs cannot be ruled out.

8.2. CSFV Vaccination

There are, in general, only two relevant types of CSFV-vaccines on the market: live attenuated (modified live vaccines = MLV) and E2 subunit (marker or DIVA) vaccines (E2subV). While the MLV are licensed or authorised by national authorities, E2SubV was registered by the EMEA. Nevertheless, it is important to mention that no marker vaccines are available at the moment.

Classical live vaccines are used both in wild boar and domestic pigs worldwide, and are based on different attenuated virus strains. The most widely used vaccine strain is the so-called 'Chinese (C)-strain'. However, there is some confusion about the origin of the C-strains since there are several of them. Most, if not all, C-strains have been attenuated by serial passages in rabbits (Aynaud, 1988). Other more common vaccine strains are the Japanese GPE-negative strain, the Thiverval strain, and the Mexican PAV strains (EC, 2003; Blome et al., 2006). C-strain-based vaccines are also used for oral immunisation of wild boar with vaccine-carrying baits (Kaden et al., 2001a, 2001b, 2001c). In Germany and other Member States C-strain baits have been used over the last decade.

During the development of marker vaccines, it became clear that the E2-glycoprotein in a purified form could induce protective immunity. This finding was the basis for the development of an E2 subunit vaccine (E2subV) that contains as an antigen only the E2 glycoprotein of CSFV. The recombinant E2 glycoprotein is produced in cultures of insect cells infected with the baculovirus vector (Hulst et al., 1993). Pigs vaccinated with a sub-unit marker vaccine only develop antibodies against the E2 glycoprotein, whereas pigs that have been naturally infected or vaccinated with a conventional vaccine develop antibodies against different viral proteins (e.g. E2, ERNS, NS3). Consequently, it is possible to distinguish between an infected and an E2 vaccinated pig (DIVA) by means of an ELISA test that detects antibodies only against the ERNS glycoproteins upon infection (Moormann et al., 2000). Two differential diagnostic ERNS antibody ELISA tests (ERNS-antibody ELISAs) are commercially available (SCAHAW, 2003, Blome et al., 2006).

In general, most MLV (e.g. C-strain vaccines) are reported as highly efficacious after a single oral or parenteral vaccine application. The onset of protection starts a few days after vaccination. In contrast, E2 subunit vaccines are described as most efficacious after a booster injection and the onset of immunity takes several weeks. Also, vertical and horizontal spread of challenge virus was described in E2 subunit vaccinated pigs upon challenge (SCAHAW, 2003; Blome et al., 2006). It was shown that after oral application, MLV are highly efficacious both in domestic pigs and wild boar (Kaden and Lange, 2001; Kaden et al., 2001a; Kaden et al., 2000a).

Under current EU legislation, the use of CSF vaccines is prohibited. However, emergency vaccination is allowed, but stringent trade restrictions are imposed for live animals, fresh meat and meat products over a prolonged period. So far, vaccination in domestic pigs in the EU has only been conducted in Romania from 2007 to 2009. Nevertheless, oral vaccination of wild boar has been conducted in several Member States during the last decade.

A vaccine bank is therefore a prerequisite for emergency vaccination against CSFV. No other 'ready-to-use' CSFV vaccines are available in Europe at present.

Potential future vaccines are reviewed by Dong et al. (2006), Beer et al.(2007), in a report from a previous EC working group (SCAHAW, 2003) as well as in an OIE publication (Blome et al., 2005). In summary, all studies concluded that chimeric pestivirus constructs are the most promising second-generation candidates for a modified live CSF DIVA vaccine with the potential to combine the efficacy of MLV with the marker properties of E2subV (Dong et al., 2006, Beer et al., 2007). However, registered products will not be available within the next three years.

8.3. CSF vaccines (EFSA, Scientific Report on CSF, 2009)

Pig Type	Vaccine type	DIVA[5]	Strain	Comercial name	Producer	Registred or authorized
wild boar	MLV[6] oral	No	C - strain[7]	RIEMSER Schweinepest - oral vakzine	Riemser Arzneimittel AG	Bulgaria, France, Germany, Latvia, Luxembourg, Romania, Slovakia,
				SUICINPEST	Istituto Zooprofilattico Sperimentale Perugia	Italy
			Thiverval strain IP-77	PESTIVAC M	SNI Pasteur SA Bucharest	Romania
domestic pigs	MLV parenteral	No	C - strain	RIEMSER[8] Schweinepestvakzine	RiemserArzneimittel AG	Germany
				SUICINPEST	IZS Perugia	Italy
				PESTIFFA	Merial, France	Belgium, Netherlands, Spain
				CZV cepa china	CZ Veterinaria S.A.Spain	Spain
				PORKIRIN	Laboratorios Ovejero S.A.	
			Thiverval	COGLAPEST	CEVA-Phylaxia Co. Ltd.	Hungary, France
			Thiverval strain IP-77	PESTIVAC	SNI Pasteur SA Bucharest	Romania
			Thiverval strain RP/ 93	ROMPESTIVAC	Romvac Company S.A.	Romania
	E2 subunit	Yes	n.a. [9]	PORCILIS PESTI	Schering Plough-Intervet	EU level

[4] data received by EFSA questionnaire
[5] DIVA differentiation infected from vaccinated animals
[6] MLV: modified live virus vaccine
[7] C-strain: Chinese strain
[8] current vaccine in the EU vaccine bank
[9] n.a. origin of strain is not available

Links to the most recent information on commercial products:

- IDEXX: http://www.idexx.com/production/swine/swine3.jsp

- Prionics: http://www.prionics.com/en/diseases-solutions/classical-swine-fever

8.4. Diagnostics/DIVA

The technical annexes of EU legislation (Commission Decision 2002/106/EC) as well as the *OIE Manual of Standards for Diagnostic Tests and Vaccines* provide useful details on the laboratory procedures for CSF diagnosis. Recent reviews give additional information on most of the tests (Blome et al., 2006; Greiser-Wilke et al., 2007).

RT-PCR has been found to be the most sensitive method for detection of CSFV (Dewulf et al., 2004; Handel et al., 2004; Depner et al., 2006a; Depner et al., 2007a, Le Dimna et al.,

2008). In general, it can be said that from an RT-PCR negative result, it can be concluded with very high confidence that the tested animal or tissue sample is not infectious to other pigs at time of sampling. Depending on the vaccine, the sample to be tested, and the definition of DIVA, real-time RT-PCR can also be used as a DIVA test ('genetic' DIVA, Beer et al., 2007). A real-time RT-PCR can also be used in combination with a vaccine that does not contain any genome (i.e. E2-subunit vaccines), or has deletions or substitutions on the primer sites (i.e. deletion mutants or chimeric vaccines). A real-time RT-PCR positive result would be proof for an infection with field virus (Koenig et al., 2007a).

Newly-developed C-strain specific real-time PCRs (Leifer et al, 2009) can be used to test vaccinated animals for the presence of MLV, but a positive result in a CSF-specific RT-PCR, followed by a positive result in the C-strain specific RT-PCR, means that infections with wild type virus can still not be ruled out. Therefore PCRs that are specific for wild type virus (Li et al., 2007, Zhao et al., 2008) that can be used to detect or rule out wild type virus infections are more important, independent of the vaccination status of the animal. This new form of DIVA vaccination is an option for the future.

In CSFV-infected pigs, antibodies are usually detectable in serum samples from one to three weeks after infection. In pigs that have recovered from the disease, protective neutralising antibodies can be detected for several years, or even throughout their whole lifetime. For antibody detection in pigs, E2-specific ELISA systems are the tests of choice.

For serological DIVA diagnostics, Erns-specific ELISA systems are the only available tests for animals vaccinated with E2-subunit vaccines. The Erns-ELISAs were developed as companion tests for the E2-subunit vaccine (Van Rijn et al., 1999). Two commercially available Erns-ELISAs, by Bommeli (now IDEXX) and Cedi-Diagnostics (now Prionics), were evaluated in a large EU-trial in the late 1990s (Floegel-Niesmann, 2001). At that time, the ELISA from Cedi-Diagnostics lacked sensitivity, while the one from Bommeli was deemed not to be specific enough. A new evaluation by the EU community reference lab in 2003, together with 15 national reference labs from the EU, concluded that an improved version of the Bommeli test was suitable as a DIVA test in combination with the E2-subunit vaccine (Anonymous, 2003).

The sensitivity of the Erns-ELISA from IDEXX is in general somewhat lower than that of E2-ELISAs. Furthermore, it is not CSF-specific, but also detects antibodies against other pestiviruses. For a population where non-CSF pestivirus infections occur, the test is therefore less useful. In these cases, the Erns-test from Prionics could be used, as it is CSF-specific, but it lacks sensitivity (Floegel-Niesmann, 2001).

8.5. Strategic approach

The measures to control and eradicate CSF are laid down in Community legislation (Council Directive 2001/89/EC and Commission Decision 2002/106/EC). The main measures consist of eradication measures, based on stamping out if CSF is suspected and confirmed on pig holdings. Emergency vaccination with 'conventional' live attenuated vaccine or marker vaccine can be used as an additional tool to eradicate the disease.

At the moment, only modified live (MLV) vaccines are available in the European Union. The E2-subunit vaccine is for the time being not available (EMEA decision 2009). Therefore, all vaccination strategies for domestic pigs in the near future have to focus on non-DIVA MLV vaccines. However, new approaches such as antigen storage might also allow the storage of

E2-subunit vaccines in a vaccine bank. The Netherlands has an arrangement for an antigen bank for the E2-subunit vaccine.

Sufficient CSF vaccine dosages should be available in the EU to perform vaccination as an additional tool to control and eradicate CSF in an emergency situation in an area with a high pig density. In a recent EFSA scientific report concerning risk associated with fresh meat originating from vaccinated pigs, it has been shown that emergency vaccination could lower the risk of virus spread via meat and meat products.

Vaccination combined with real-time RT-PCR testing is a most promising strategic approach for the future. Therefore, the use of CSF vaccines can be recommended under certain conditions in an outbreak situation (high pig density, several outbreaks but restricted to a single region, etc.). A vaccine bank should enable a short reaction time, which is crucial for an efficient emergency vaccination programme.

At present, an EU CSF vaccine bank with 1 million doses of MLV C-strain vaccine is available. This was used in Romania for the eradication of CSF in back-yard holdings.

8.6. Feral pigs/wild boar

For vaccinating feral pigs (wild boar) oral vaccine (bait) products with MLV are needed. However, there is no need for a vaccine bank for wild boar, as sufficient commercial vaccines are available on the market. MLV vaccines from Riemser Arzneimittel AG and Merial have been proved safe and immunogenic.

8.7. Domestic pigs

Since Member States have different priorities and strategies for the use of CSF vaccines in case of emergency, both types of vaccines (MLV and E2subV) have to be taken into consideration.

For the time being, vaccination strategies have to focus on highly efficacious non-DIVA MLV vaccines until the E2subV receives regulatory confirmation. The latter is expected to become available within a year or two. An antigen bank instead of a vaccine bank with the ready-to-use E2subV might be the best interim option. However, further validation data are needed before relying on an E2-antigen bank.

8.8. Size of CSFV vaccine bank

In model calculations, different types of outbreaks in husbandry farms were analysed for the Dutch situation. In the Netherlands, there is high animal density, with large production farms. In a larger outbreak, the number of farms infected and the number of animals to be vaccinated were calculated. The model is based on a strategy of using a marker vaccine in a 2 kilometre radius around the infected farm, in a worst-case scenario with late discovery and 11 to 20 secondary outbreaks at the point in time when the first infected farm is detected. In the table below, the number of animals which have to be vaccinated in worst-case spread (95% percentile) and medium-case spread (50% percentile) are presented:

Large Outbreak (11 – 20 farms)	50 %	95 %
Number of vaccinated sows	23 400	50 760
Number of vaccinated piglets	92 893	202 896
Number of vaccinated production pigs	11 610	39 762
Total number of vaccinated pigs	127 903	293 418

Consequently, a vaccine bank with a minimum of 300 000 doses would be needed for the Dutch situation.

For MLV, similar amounts of vaccine might be needed. In addition, larger areas might be vaccinated (3 km or 10 km), depending on the individual strategy of a Member State.

In conclusion, the CSF vaccine bank storing MLV CSF vaccine and E2subV should be maintained and extended. A minimum of 2 million doses is recommended.

The average cost of 1million doses of live attenuated CSF vaccine is estimated to be around 1 400 000 EUR, including purchase of the vaccine, four replacements (the maximum possible), and storage for three years (vaccine shelf life), should the final replacement take place shortly before the contract period ends.

8.9. CSFV diagnostic bank

Because of CSF prevalence in wild boar in several Member States, conventional commercial diagnostic tests in sufficient quantities are available (E2-ELISA, rRT-PCR). Therefore, a diagnostic bank for conventional tests seems unnecessary.

In contrast, the future use of marker (DIVA) vaccines is directly connected with the availability of suitable DIVA diagnostics. Therefore, a diagnostic bank should be taken into consideration if the use of DIVA vaccines is envisaged. In such a case, a diagnostic bank with 50 000 DIVA tests would be needed to enable the first screening of farms after immunisation.

8.10. Legal Basis for vaccination/vaccine banks

Commission Decision 2007/862/EC of 18 October 2007 on the renewal of the Community stocks of live attenuated vaccine against classical swine fever regulates the purchase of 1 million doses of live attenuated CSF vaccine for emergency vaccination of domestic pigs. The use of emergency vaccination in case of CSF is regulated by Council Directive 2001/89/EC of 23 October 2001 on Community measures for the control of classical swine fever.

In principle it is a *vaccination-to-kill* approach. Article 19 states that a Member State which intends to introduce vaccination in pig holdings shall submit to the Commission an emergency vaccination plan for approval. The plan must prescribe that all vaccinated pigs will be slaughtered or killed as quickly as possible and the fresh meat produced from these pigs will be processed in such a way to inactivate the CSF virus. Derogations may be authorised if a marker vaccine has been used. One Member State has carried out large-scale emergency vaccination with marker vaccine from the EU vaccine bank in industrial farms.

8.11. Conclusions

There is a continuous risk of CSFV outbreaks both in feral and domestic pigs. In addition, wild boar is a dangerous reservoir for CSFV. There are several regions in the EU with CSFV restrictions due to CSFV in wild boar. Finally, CSFV is a worldwide problem and the risk of CSFV being introduced from outside the EU remains significant.

At the moment, only live CSFV vaccines are available. However, E2 subunit vaccines should be also taken into consideration, and alternative ways of storage (e.g. an antigen bank) might be a solution for stability problems with this type of vaccine.

For the E2 subunit marker vaccine, an accompanying DIVA test has to be available shortly after an outbreak. Therefore, a diagnostic bank for an E^{RNS}-marker assay should be available to allow early testing.

8.12. Recommendations for CSF vaccine and diagnostic banks

- Emergency vaccination should be accepted as an important and valuable tool for the control of CSF in wild boar and domestic pigs. Strategic programmes for emergency vaccination should become part of contingency plans in Member States with a high pig density.
- To enable efficient, timely emergency vaccination, an EU vaccine bank with a live attenuated vaccine (e.g. C-strain) is needed, enabling vaccination of at least 2 million pigs.
- Alternatively, the storage of the E2-subunit vaccine as an antigen should be tested and evaluated by the European Union Reference Laboratory for CSF or by other appropriate mechanisms. Providing the evaluation is positive, an antigen bank should be implemented.
- A diagnostic bank for an E^{RNS}-marker ELISA is needed, together with the marker vaccine bank. Test kits for not less than 50 000 pigs should be available within seven days of a request. The availability of a diagnostic bank with a suitable PCR test is not an essential pre-requisite.
- Practically-oriented screening schemes are needed to identify infected animals in a post-vaccination area.
- A properly designed and implemented emergency vaccination strategy, together with a targeted search for chronically-infected animals in vaccinated herds during final screening, would reduce the risk of the virus remaining active in fresh meat more than a conventional non-vaccination strategy.
- If one of the new prototype modified live marker vaccines is licensed, it should be included in the vaccine bank together with the appropriate diagnostic tests.

9. AFRICAN HORSE SICKNESS (AHS)

9.1. Severity and likelihood

African Horse Sickness (AHS) is a non-contagious insect-borne viral disease affecting all species of Equidae. It is a notifiable disease due to its severity and rapid spread. AHS is caused by one or more of the nine different serotypes of an Orbivirus of the family Reoviridae.

The Orbivirus genus also includes bluetongue virus (BTV) and epizootic haemorrhagic disease virus (EHDV), which have similar morphological and biochemical properties, but distinctive pathological and antigenic properties as well as host ranges. Orbiviruses are unenveloped viruses approximately 70 nm in size. The genome consists of 10 double-stranded RNA segments, encoding seven structural proteins (VP1 to VP7), and four non-structural proteins (NS1, NS2, NS3, NS3A). Nine antigenically distinct serotypes of AHSV have been identified. Genome segment 7 encodes the inner capsid protein VP7, which is known to be highly conserved among the nine viral serotypes. It forms the basis for a number of antigen, antibody and nucleic acid based commonly used diagnostic tests.

The disease is characterised by a variety of clinical forms ranging from pulmonary, cardiac, fever and unspecific sickness. The mortality rate can be up to 70% in affected horses and 50% in mules. However, other equine species such as donkeys and zebras may never display clinical signs being infected. Zebras play an important role in the maintenance of the virus in Africa.
AHS is transmitted by at least two species of *Culicoides* midges. All serotypes of AHS virus occur in eastern and southern Africa. Examples of occasional outbreaks that have occurred outside sub-Saharan Africa are the pandemic in certain countries of the Middle East up to India and Turkey (1959-1961), the outbreaks in Spain (serotype 9, 1966) and the epidemic caused by serotype 4 in Spain (1987-1990), Portugal (1989) and Morocco (1989).

Laboratory diagnosis is possible based on the identification of infectious virus, virus nucleic acid, viral antigens or specific antibodies.

The risk of AHSV being introduced into the EU has risen over the last decade. The northward expansion of many other vector-borne diseases, for example the unexpected introduction of serotype 8 of Bluetongue virus (BTV-8) to northern Europe, has made this apparent. In addition, the epidemic of BTV-8 has shown that introduction can lead to a huge outbreak with all its impacts and consequences. AHSV is, like BTV, transmitted by a species of *Culicoides* midge. The risk of introduction is minimised by the current legislation on trade in equidae and their products. Nonetheless, there was an outbreak in Portugal and Spain in 1989. Further, the BTV-8 epidemic has shown that the current legislation can not completely prevent the introduction of these vector-borne diseases.

The EFSA report entitled ‘Epidemiological analysis of the 2006 bluetongue virus serotype 8 epidemic in north-western Europe’ (4 April 2008) warns Member States that they need to be aware of the AHS threat. It reports that the *Culicoides* fauna endemic to northern Europe can be vectors of BTV and other diseases. In other parts of the world, BTV vectors have been shown to transmit other viral pathogens of livestock -- African horse sickness virus, Akabane virus, epizootichaemorrhagic disease virus, and equine encephalosis. This suggests the same might happen in northern Europe if weather conditions favour introduction of the pathogen.

Veterinary authorities in the Member States should be aware of these threats so as to be able to respond quickly to possible incursions.

It can be concluded that:

- AHSV could spread very rapidly across national borders.
- An outbreak would have serious socio-economic consequences.
- An outbreak would have a major impact on international trade of animals and animal products.
- Neither individual owners of equidae nor the organised equidae industry have the instruments to control and eradicate a possible epidemic.

9.2. AHS vaccines

Modified attenuated vaccines against the nine serotypes of the AHSV are available outside the EU (*ARC-Onderstepoort Veterinary Institute*, South African Republic). A modified attenuated vaccine was used in Portugal and Spain during the epidemic of AHSV-4 between 1987 and 1988. However, the use of attenuated vaccines has some drawbacks, such as the possibility of reassortment of the vaccine virus with the field virus and the possibility of virulence reversion. Furthermore, a DIVA diagnostic test is not available. Due to these reasons, as well as possible commercial restrictions on the movement of equidae, the use of attenuated vaccines has been discouraged in the past.

During 1989-1990, a mass vaccination campaign was conducted in Spain with an inactivated vaccine against AHSV-4 to avoid the problems of live attenuated vaccines. A recombinant protein vaccine against AHSV-4, based on VP2, VP5 and VP7 structural proteins was also developed as an alternative to live attenuated vaccines, with promising results. At the same time, an indirect ELISA that uses the AHSV serotype 4 non-structural protein NS3 as an antigen was developed. The assay was used to differentiate between animals naturally infected, or vaccinated with the conventional live vaccine, from those vaccinated with an inactivated vaccine containing purified virions.

9.3. Vaccination strategy

The disease has had a huge impact in affected countries due to direct and indirect losses. Vaccination against AHSV is a very important tool for the control of the disease, so many countries are carrying out vaccination programmes. The aims are:

- To prevent clinical symptoms and mortality.
- To decrease viral circulation.
- To enable movement of animals from the restricted zone with appropriate sanitary guarantees.
- To achieve final eradication of the disease.

Animals have to be vaccinated against all serotypes which are present in a country, because there is no cross-immunity among the serotypes.

The fact that AHS is a vector-borne disease makes vaccination necessary, but it should be combined with other tools. These other tools, such as culling infected animals, transport bans,

hygiene measures and measures against the vector alone are not sufficient to eradicate AHS. The outbreaks in Spain and Portugal were eradicated by vaccination.

The horse industry in most parts of the EU is different from other animal industries. If an eradication strategy for a horse disease is not supported by the community, non-compliance may make it very difficult to succeed. Vaccination is a well-accepted control measure to eradicate a disease, particularly a horse disease. In addition to veterinary interests, there are also socio-emotional arguments for vaccinating against AHS.

Emergency vaccination is an obligation in case of an outbreak, according to the current EU legislation.

One of the proven vectors for AHS is *Culicoides imicola.* This occurs in the south of Europe, but is expanding its habitat, and recently reached the mainland of France. Again, the BT outbreak showed that other endemic species of *Culicoides* (there are many throughout the EU) could be competent vectors for AHSV-related virus.

The OIE code sets standards for movements of vaccinated or seropositive equidae and provides guidelines to be followed to maintain or recover AHS-free status following an outbreak where vaccination is used. AHS has a huge impact on trade, and vaccination does not diminish this.

Theoretically, and if available, use of DIVA vaccine for AHS could be of benefit to control outbreaks and to demonstrate the absence of AHSV circulation.

The Community legal framework for AHS surveillance and control is laid down in Council Directive 1992/35/EC.

9.4. Recommendations for an AHS vaccine and diagnostic bank

A very rapid response is important for a good vaccination strategy. A vaccine bank enables this. However, because there is no cross-protection among the AHSV serotypes, maintaining a vaccine bank against all nine would be very expensive. It is thus advisable to have an EU vaccine bank for those posing the highest risks in Europe. Taking into account the epidemiological situation of AHS in the Sub-Saharan region, the most dangerous serotypes are 2, 4 and 9.

Alternatively, a Working Seed Virus bank (WSV) could be developed for the nine serotypes, thus covering the first steps in developing a vaccine. The WSV could develop an inactivated vaccine, so that the ready-to-use vaccine would be available 16 weeks later in case of emergency (see flow chart of the AHS inactivated vaccine production process below). If an attenuated vaccine were to be produced, the time required to obtain the final product would depend on how long it took to achieve proper attenuation of the seed virus. These estimates do not take into account the time needed to obtain even minimum data necessary for authorisation of the vaccine.

Based on experience with other Community vaccine reserves, a total number of 150 000 doses of each of the three proposed attenuated serotypes would suffice for a first emergency response. Continuous monitoring of AHS in countries geographically close to the EU is recommended. Risk analysis should identify serotypes that may threaten the EU and trigger

development of vaccine against them. The cost of maintaining a bank for diagnostic tests is not justified.

9.5. Conclusions and Recommendations

- A vaccine bank with live attenuated vaccines against serotypes 2, 4 and 9 of AHSV should be established. Others, e.g. serotype 5, could be added subsequently.
- If one of the new prototypes of recombinant vaccines is licensed, it should be included in the vaccine bank as a priority.
- The vaccine bank should contain a minimum of 150 000 doses for each of the proposed serotypes.
- In addition, developing a working seed bank for all nine serotypes of the virus is recommended, to cover the first steps for developing an inactivated vaccine.
- The availability of a diagnostic bank with a suitable test is not an essential pre-requisite.
- After establishing the initial stock, continuous monitoring of the epidemiological situation of AHS in countries near the EU is recommended, to identify serotypes which might become risks for EU livestock, to enable development and procurement of vaccines against them.

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10. BLUETONGUE (BT)

Bluetongue is an infectious but non-contagious vector-borne disease caused by Bluetongue virus (BTV). BTV is an RNA virus belonging to the Orbivirus family that infects all domestic and wild ruminants. Bluetongue is an OIE listed disease, which requires compulsory notification within the European Union.

The BTV contains 24 different serotypes (a new isolate proposed as a 25th serotype was recently identified in Switzerland and known as Toggenburg virus). Eight have been detected in the last few years in the EU: serotypes 1, 2, 4, 6, 8, 9, 11 and 16. In nearby countries, serotypes 15 and 24 have been detected in Israel and Turkey. The virulence and mortality rate caused by the different serotypes, and strains within the serotypes, vary considerably.

New BTV serotypes tend to arrive due to imports of non-controlled viraemic animals from affected countries, or via infected vectors (biting midges) blown in by the wind. The role of wildlife in the spread of BTV is so far unknown.

Once the disease is established, BTV is transmitted when midges of the haematophagous genus *Culicoides* (Diptera: Ceratopogonidae) bite ruminant hosts. The main vectors in Europe are species of the *Culicoides imicola* complex, the classical Afro-Asiatic species, which prefer mild climates, or species of the *C. obsoletus* complex, responsible for the recent dramatic spread of BTV8 in northern Europe. Vertical transmission (transplacental) has been reported for BTV8 in calves, and is suspected in lambs born to infected mothers. Oral transmission has been also described, but appears to be rare.

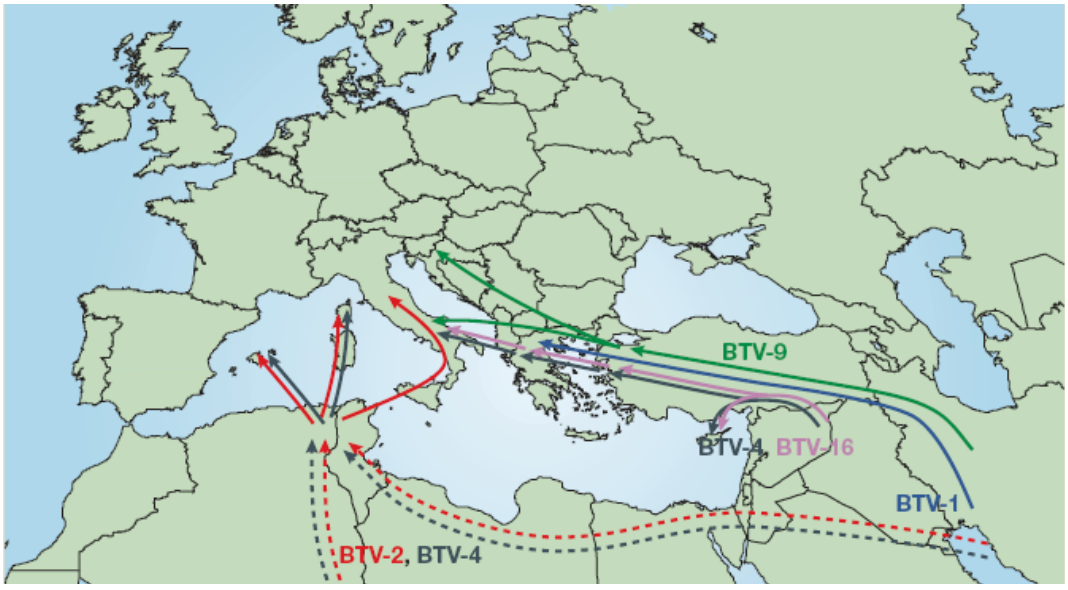
BTV used to be considered exotic in most of Europen countries, but recent incursions into southern and northern Europe have had a devastating impact on European farmers and the health and welfare of affected animals. The situation has changed with incursions of new serotypes, such as BTV8, in areas of the EU which were not considered risk zones and had never previously reported the disease. The same could happen with other serotypes.

Since transmission and spread are related to the presence of vectors, climate plays an important role. There is no virus transmission at temperatures below 10 °C, so some areas define 'vector free' periods when it is too cold for midge activity and hence virus spread. According to distribution of vector species, two different BT regions are seen:

Southern Europe: The main vector is *Culicoides imicola*, the classical Afro-Asiatic species, which prefers mild climates. Others are members of the *C. obsoletus* complex, *C. pulicaris* and other species of *Culicoides*, but they are rarer. Southern Europe has been affected since the late 1990s by a South — North or East — West drift of several BTV serotypes (1, 2, 4, 9 and 16). The main factor in this spread was thought to be windborne dispersal of infected midges. In 1999, **BTV9** was introduced into Greece and Bulgaria, and spread to the Balkans: Albania, Bosnia Herzegovina, Croatia, Kosovo, Macedonia Republic, Republic of Serbia and Montenegro. Subsequently, BTV9 spread to the Italian island of Sicily and mainland Italy. In 2003, **BTV4** was introduced into Sardinia, Corsica and Menorca, and in 2004, **BTV16** was found in Sardinia and spread to Corsica. In 2004, BTV4 was also found in Spain and Portugal, and in 2007 **BTV1** was found in Spain, Portugal and France.

Northern Europe: Vector species of the *C. obsoletus* complex are the main factor responsible for the recent spread of **BTV8** in northern Europe, though it is not known how it entered. Further spread has been attributed to animal movements into areas where competent vectors were highly abundant. Since 2008, outbreaks of bluetongue have occurred in Austria, Belgium, Czech Republic, Denmark, France, Germany, Greece, Hungary, Italy, Luxembourg, Netherlands, Portugal, Spain, Sweden, Switzerland, UK and even as far north as Norway. **BTV1** has also been spread by *C. obsoletus* complex in France and in northern Spain, territories where *C. imicola* has never been reported.

1 Routes of introduction of different BTV serotypes



Source: Purse, B.V. et al. (2005): Climate change and the recent emergence of bluetongue in Europe. *Nat Rev Microbiol* **3,** 171-81 (2).

In October and November 2008, the Netherlands and Germany reported laboratory findings of BTV6 in cattle located in neighbouring parts of their territories with very little, if any clinical signs of bluetongue disease. In early 2009, Belgium reported a similar situation regarding bluetongue virus type 11 (BTV11). Information on the genetic sequence available from the virus isolates indicates a high similarity with the South African modified live vaccines of BTV serotypes 6 and 11. The positive findings are most likely to be due to use of a modified live vaccine, which may have led to limited circulation in the local midge vector population.
Epidemiological assessment of the situation indicated that no virus had been isolated and no clinical signs of bluetongue disease were observed. Strengthened surveillance in Netherlands, Germany and Belgium has shown no evidence of further virus circulation in the 2009 vector season, nor was a virulent strain present.

10.1. Vaccination for BT control

The disease has caused significant economic losses due to direct effects in animals and indirect losses. Vaccination against BTV is considered to be the main means of control.
Vaccination can be used to:

- Prevent clinical signs and mortality due to the disease.
- Prevent viral circulation.
- Prevent further spread.
- Allow the safe movement of animals.
- Control and eradicate the disease (freedom from disease).

10.2. BT Control: Legislative Frame work

Council Directive 2000/75/EC (the Directive) lays down control rules and measures to combat BT within the European Community. These rules and measures include the establishment of *protection and surveillance zones* and a ban on animals of the susceptible

species leaving those zones. In accordance with the Directive, *vaccination* against bluetongue is only allowed within the protection zone.

Commission Regulation (EC) No 1266/2007 of 26 October 2007 lays down rules, among other aspects, on *exemptions from the exit ban* and on the harmonised *requirements for movements* of certain safe animals of susceptible species in relation to BT from restricted zones. Regulation (EC) 1266/2007 has been amended several times. The most important changes related to vaccination were in Regulation (EC) No 123/2009 of 10 February 2009, which allows *preventative vaccination in* 'lower risk areas'; parts of the restricted zone where the BT virus is not circulating. These areas are protected against uncontrolled movement of animals originating from zones in which there is virus circulation. At the same time, limited restrictions are applied to trade in vaccinated live ruminants from 'at lower risk areas' to disease-free areas.

10.3. BT vaccines

Much effort has gone into development of vaccines, and various formulae have been used in the EU in recent years, either attenuated or inactivated. BT vaccines need to be improved, as their efficacy is limited to specific serotypes and DIVA vaccine are not available. Nowadays, only inactivated and attenuated (modified live vaccines) are commercially available. Below we summarise the pros and cons of both, and the situation regarding other experimental vaccines.

Attenuated vaccines
Attenuated vaccines produced by Onderstepoort Biological Products (South Africa) have long been used to control BT in sheep in southern Africa, and more recently in Corsica, the Balearic Islands, and Italy during the BTV2 and BTV4 outbreaks of 2000-2005. These provide robust protection after one injection for at least a year, and are cheap to produce. However, they are not always safe and have some drawbacks: they can generate mild clinical signs after injection, abortions, transiently depressed milk production, and decreased semen quality. They can not be used in cattle, nor in pregnant females. As the vaccine virus can elicit a viraemia of over two weeks in vaccinated sheep, there is a possibility of reassortment of the vaccine virus with the field virus, and also the possibility of virulence reversion. Finally, attenuated vaccines are not DIVA vaccines. In short, the use of this vaccine has been discouraged. Nevertheless, modified live attenuated vaccines are also developed in Italy, and widely used there with excellent results.

Inactivated vaccines
Inactivated vaccines can generate safe, protective immunity if properly prepared. However, two injections may be necessary for strong, long-term immunity, particularly in cattle. Furthermore, the inactivated vaccines developed and produced recently are only available for a few serotypes (1, 2, 4 and 8). DIVA inactivated BT vaccines are theoretically possible, but have not yet been developed.

The European Food Safety Authority has recommended inactivated vaccines, and they have been used since 2005. Since 2008, a mass campaign using inactivated vaccines is ongoing in large parts of the EU.

Virus-like particles
BTV structural proteins can be produced as recombinant proteins encoded by Baculoviruses in insect cells, in which they auto-assemble as virus-like particles (VLP), presenting BTV

antigenicity without BTV genetic information. Multivalent BT vaccines could be produced in future, since VP2 from several viral strains can be included. Laboratory trials have shown that VLP are effective in protecting against homologous BTV challenge, and partially protecting against heterologous BTV challenge. Further studies are awaited to evaluate their structural long-term stability, their cost of production/purification, and their efficacy in the field. They are considered to be naturally safe and do not require inactivation, although a recent study pointed out that laboratory-produced VLP batches included large quantities of Baculoviruses.

Recombinant vaccines
Recombinant vectors could be developed as vaccines, if they are safe, inexpensive, DIVA, flexible for multi-serotype inclusions, and if they could provide rapid onset of immunity and long-term protective immunity in one shot. Some promising preliminary laboratory studies have been published.

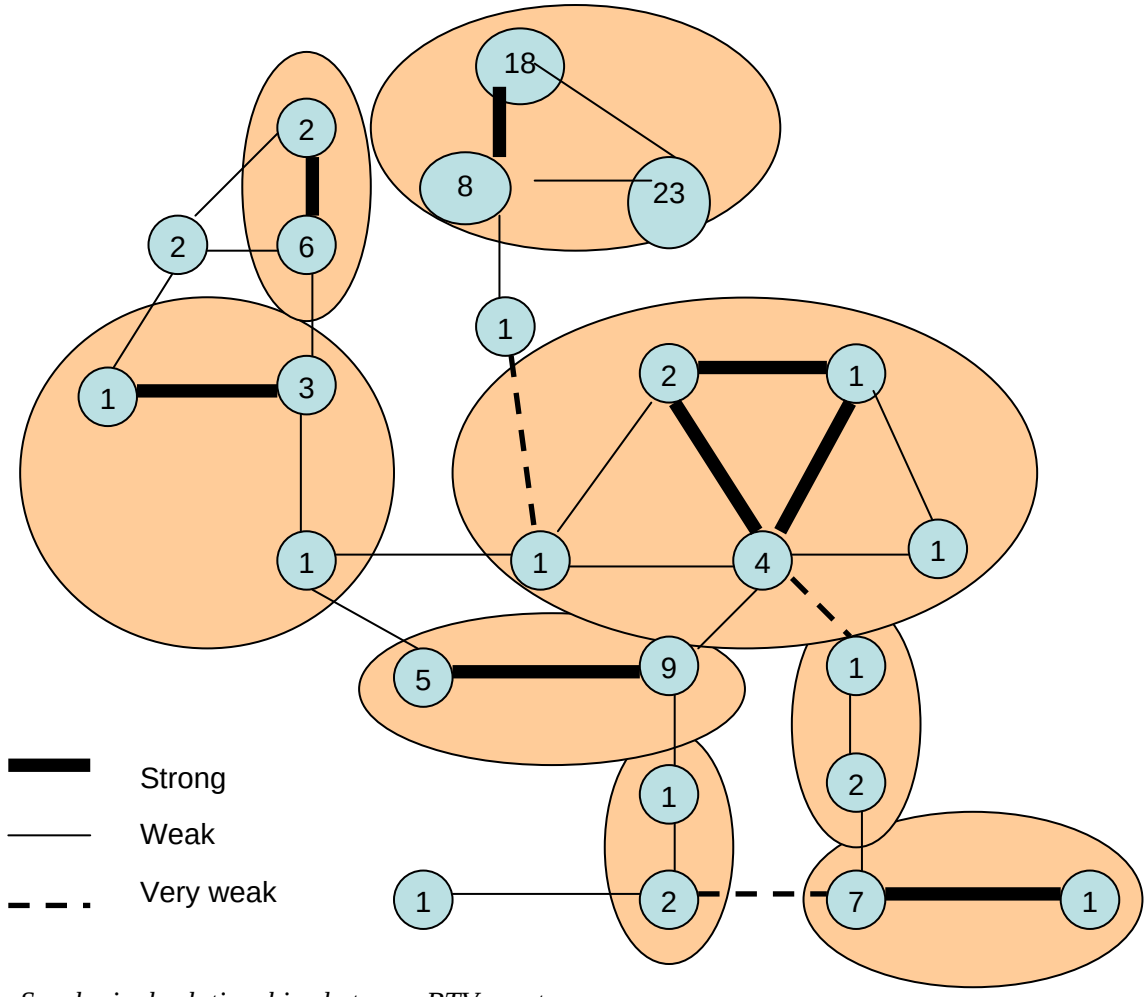
A recombinant vaccinia virus that expressed both VP2 and VP5 of Australian BTV serotype 1 induced variable titers of neutralising antibody in sheep, and afforded protection against homologous challenge, but this approach has not been pursued.

More recently, a recombinant canarypox virus-VP2/VP5 vaccine (two injections, 22 days apart) that induced highly effective protective immunity in sheep was described. This has a major advantage in that the existing VP7 competitive ELISA assay would distinguish vaccinated from naturally infected animals (DIVA), and it utilises an expression vector that is incorporated in several vaccines already in use in the EU and elsewhere. The vaccine is still being developed.

Finally, a replicative capripox encoding for VP2, VP7, NS1 and NS3 (one injection) was partially protective in sheep. Thus, recombinant vectors can provide protective immunity with DIVA properties, but their efficacy barely reaches that of inactivated vaccines, as several applications are required for efficient long-term protection.

10.4. Strategic approach for BT vaccine bank

At present, 24 different BTV serotypes have been described and there is limited cross-protection (see flow chart below). Establishing a vaccine bank for every serotype is considered impractical. It would be possible for the most relevant serotypes in the EU (1, 2, 4, 8, 9, 15, 16 and 24), but the sudden appearance of serotypes 8, 6 and 11 recently shows that any serotype could turn up at any time.



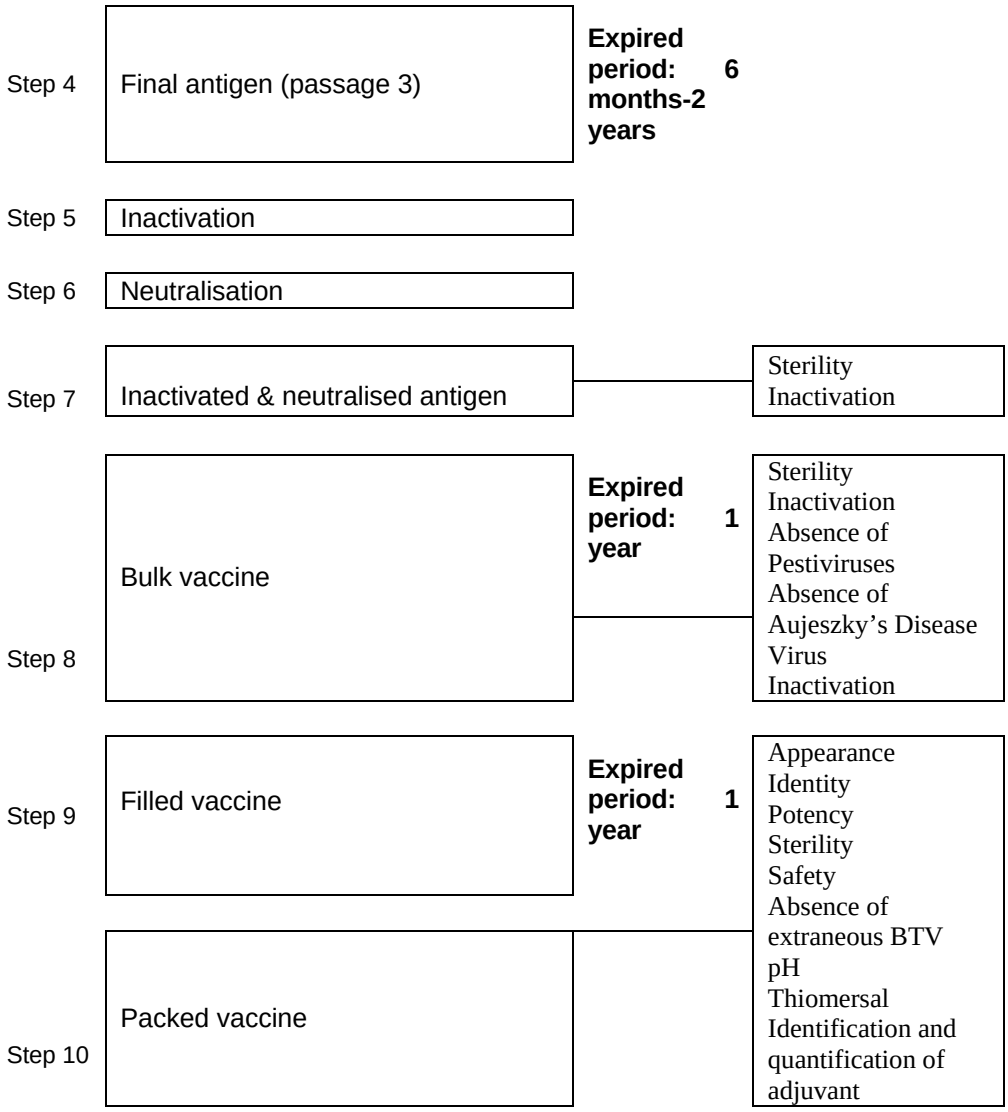
Serological relationships between BTV serotypes

Inactivated vaccines currently used in the EU due to their novelty or temporary license conditions have short shelf lives, usually no longer than one year. This limits the utility of vaccine banks, but they may become more practical as our knowledge of shelf lives grows.

Because vaccination is the most effective tool to control BT, to avoid economic losses due to movement restrictions, at least a Working Seed Virus (WSV) Bank that could be used in case of emergency would be needed.

With a WSV, the steps to make a packed inactivated vaccine would take around 5-6 months, including the testing and release of antigen and vaccine in accordance with existing European legislation. During the process, 12-14 weeks are needed to prepare, test and release the bulk antigen, and 12-14 weeks to produce, test, release and pack (see flow chart of the BT inactivated vaccine production process below):

Step 1	WSV	**Non expired date**
Step 2	Antigen passage 1 (pre-inoculum)	
Step 3	Antigen passage 2 (inoculum)	



Live attenuated vaccine can also be used in an emergency vaccination campaign. Repeated passages of the virulent virus field in cell cultures are needed to attenuate the virus. Between 20 and 60 passages are usually required (the average is 50). Two passages per week can be made, so it would take between 10 and 30 weeks to achieve proper attenuation of the virus. After that, several tests would be needed to demonstrate the safety and security of the obtained isolated virus, as well as any test required by the EMEA. The performance of all these tests could take years.

10.5. BT vaccine bank conclusions and recommendations

As fully-authorised vaccines become available and knowledge of true shelf life grows, vaccine banks become a practical solution. Meanwhile, establishing WSV Banks at least for non-endemic BTV serotypes in EU (3, 5, 6, 7, 10, 11, 12, 13, 14, 15, 17, 18, 19, 20, 21, 22, 23 and 24) is recommended. As previously indicated, it would take 5-6 months to produce ready-to-use vaccine. There are no live attenuated vaccines authorised for the EU as a whole; currently, only national authorisations exist.

Continuous monitoring of the epidemiological situation of countries geographically close to the EU is recommended to identify serotypes posing the greatest risk for EU livestock. This would advance development of vaccines against those serotypes.

10.6. BT diagnostic techniques

Bluetongue virus (BTV) belong to family Reoviridae, divided into 20 genus, differentiated by an immunological test which detects viral proteins, among them genus Orbiviridae. On the basis of a neutralisation test, 24 serotypes are distinguishable.

The only accepted technique for identification of the agent for international trade is Reverse-Transcription for Polymerase Chain Reaction. Nevertheless, there are other techniques to identify the agent, approved in the *OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals*, such as virus isolation or immunological methods.

Serological tests are also available and listed in the OIE Manual, but nowadays the Competitive Enzyme-linked immunosorbent assay is the preferred technique. Regarding Virus Isolation, some techniques are in common use, but Inoculation of Embryonated Chicken Eggs is usually favoured. Inoculation of sheep or cell culture may be more convenient in some cases.

To distinguish different serotypes, neutralisation tests are type-specific and able to differentiate among the 24 serotypes. Several laboratories produce tissue culture-based methods such as plaque reduction, plaque inhibition, microtitre neutralisation and a fluorescence inhibition test.

Finally, the Reverse-Transcription for Polymerase Chain Reaction allows the detection of the virus-specific nucleic acid from blood and other tissues of infected animals, with the inconvenience that it does not confirm the presence of infectious virus.

There are a large number of techniques available to diagnose BTV, most of them produced by several commercial laboratories. This means that enough tests or antigens are available within a short time period to manage the outbreak of a new serotype or any other situation that might require a large number of animals to be tested.

Several techniques are accepted for international trade. This raises the likelihood of enough tests being available if required, so that a control program to prevent the virus spreading can be launched relatively quickly. Capacity is guaranteed both by the number of techniques available, and by the fact that for each, there are several laboratories producing reagents, antigens and tests.

In conclusion, the availability of the diagnostic tests is good, so the cost of maintaining a bank for tests is not justified.

10.7. Conclusions and Recommendations

- Establishing vaccine seed-stocks for BTV serotypes not currently present in the EU is recommended.

- The vaccine seed-stocks should enable the production of enough inactivated vaccine to provide up to 5 million doses for each of the proposed serotypes.
- Establishing continuous monitoring of the epidemiological situation of BT in countries geographically close to the EU is recommended to identify serotypes which might become risks for EU livestock, so as to adjust the development and procurement of vaccines against them.
- Availability of BT diagnostic tests is not a problem, so a diagnostic bank is not needed.

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11. AFRICAN SWINE FEVER (ASF)

African Swine Fever (ASF) is a highly contagious infectious and complex disease caused by an icosahedral DNA virus of the family *Asfarviridae*. ASF virus infection produces a range of syndromes, varying from peracute, acute to chronic disease, and apparently healthy virus carriers. ASF acute and subacute forms may resemble a variety of other swine haemorrhagic diseases, and it can easily be confused with Classical Swine Fever and Erysipelas. Laboratory tests are required to distinguish between them and to establish a correct diagnosis.

Pigs are the only domestic animal species that is naturally infected by ASF virus. European wild boar is also susceptible to the disease, exhibiting clinical signs and mortality rates similar to those observed in domestic pigs. In contrast, African wild pigs such as warthogs (*Phacochoerus aethiopicus*), bush pigs (*Potamochoerus porcus*) and giant forest hogs (*Hylochoerus meinertzhageni*) are resistant to the disease and show few or no clinical signs. These species of wild pigs act as reservoir hosts of ASF virus in Africa. ASF is also transmitted by certain soft tick species (*Ornithorodoros* species). In areas where there are

competent vectors of the Ornithodoros tick genus, transmission via these vectors can promote virus persistence over a long period. In the absence of Ornithodoros ticks, ASF may persist in domestic pigs or wild boar, and is probably dependant on the existence of large, continuous populations of pigs whose high reproductive rate ensures constant availability of naïve pigs for infection and further spread.

Currently, ASF is endemic in many sub-Saharan African countries, covering east to west and also affecting central and southern countries. In Europe, it is still endemic in the island of Sardinia (Italy), and since 2007 has emerged in the Caucasus region (Georgia, Armenia, Azerbaijan, and Russia). If ASF is not controlled in Africa and the Caucasus, other trading countries cannot be fully protected against the disease. Today the potential distribution of the infection is transcontinental, and the authorities are aware ASF may emerge as a very dangerous animal health problem in the near future.

No treatment or effective vaccine is available against ASF, and disease control is based on a rapid laboratory diagnosis and the enforcement of strict sanitary measures.

11.1. Strategic approach

Measures to control and eradicate ASF are laid down in Community legislation (Council Directive 2002/60/EC and Commission decision 2003/422EC). No vaccines or drugs are available to prevent or treat ASF infection. All control and eradication measures applicable are based on classical disease control methods, including intensive surveillance, epidemiological investigation, tracing and stamping out of infected herds, and designation of infected zones. These measures are combined with strict quarantine and bio security measures and animal movement control.

Control and eradication of ASF is not easy. The various scenarios for ASF virus infection of domestic pigs and virus diffusion that may occur are complex. Therefore, there is no single recipe. Each scenario requires its own specific strategy.

ASF eradication programmes successfully used in the past in endemic areas of Europe (Portugal and Spain) demonstrated that vaccine was not essential. These programmes were based on the extensive use of serological tests. Specific antibodies could be detected in early stages of infection and persisted in pigs that recovered for months, even years. This allowed easy detection of infected carrier animals. For these reasons, serological techniques contribute significantly to eradication.

However, at present, there is the risk of new strains appearing, showing moderate-acute forms of the disease. If ASF is suspected, virus detection techniques such as PCR are the method of choice for diagnosis in surveillance programmes. Infected pigs show a viraemia that is commonly detected from 3 dpi for several weeks, or until pigs die of acute ASF, before antibodies are produced. In endemic areas of Europe, such as Sardinia, where chronic or unapparent forms could be present, control of movement of animals and extensive serological screening are also essential to detect and remove carrier pigs. It is also important to remember that low-virulence ASF strains may not cause signs or lesions that signal their presence. If ASF is suspected, pig movements should be restricted and diagnostic tests (which should include virus detection techniques) should be performed immediately.

11.2. Laboratory Diagnosis

Laboratory confirmation of a presumptive diagnosis of ASF will depend on detection of the virus and/or detection of antibodies, and it should be performed bearing in mind the epidemiological situations and different scenarios that may occur.

There is a wide variety of good, specific and sensitive tests to detect ASF-specific antibodies. However, there is only one commercial ELISA test available, which could be a constraint in an ASF emergency. The Community Reference Laboratory for ASF also offers an in-house OIE ELISA. Both ELISAs have been proved very sensitive and specific for the detection of ASF-specific antibodies.

Currently, sensitive, specific virus detection techniques based on conventional and real time PCR tests are also in use in most EU diagnostic laboratories. In addition, there is a commercial test for ASF Antigen detection (Ag-ELISA). This antigen ELISA shows low sensitivity for virus detection and is only recommended for acute forms of the disease. This Ag-ELISA technique is not recommended as a single virus detection technique, nor for diagnosis from individual samples in Member States.

There are detailed instructions for laboratory diagnostic procedures for ASF in the OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals, 2008 (Chapter 2.8.1).

Since no vaccine is available, the presence of ASFV antibodies is indicative of previous infection, and since antibodies are produced from the first/second week of infection and persist for long periods, they are a good marker for diagnosis.

11.3. Diagnostic considerations

In countries free of ASF, but where its presence is suspected, the laboratory diagnosis must be directed towards virus detection (by PCR) and isolation. ASF virus can be detected by PCR from a very early stage of infection in tissues, EDTA-blood and serum samples. Pigs that have recovered from acute or chronic infections usually exhibit a viraemia for several weeks. This makes the PCR test a very useful tool for detecting ASFV in pigs infected with acute, low or moderately virulent strains.

Recent findings have revealed that ASF virus is not as homogeneous as previously thought, but can exist in variants that widely differ genetically. Antigenic variation of different ASFV proteins has also been observed in these viral isolates. Trends in ASFV research are focused on the biological and molecular characterisation of currently circulating field isolates, belonging to a wide range of the genotypes and their comparison with classic virus isolates. The research seeks to understand their distinctive epidemiological behaviour, as well as ways of improving new diagnostic tools for field conditions in different regions and epidemiological situations.

ASF diagnosis through serological and virological screening tests performed during several recent outbreaks of ASF in East African countries showed a significant number of ASF seronegative domestic pigs, despite the ASF virus being found in a significant percentage. The epidemiological significance of this finding is now being studied in an EU project (ASFRISK, 2008). This also includes development of a front-line and pen-side test for use in outbreak surveillance programmes in countries with limited capacity for laboratory diagnosis. Nevertheless, serological screening tests are very useful for ASF chronic or unapparent forms produced by low virulent ASFV strains.

11.4. Vaccination

At present, there is no vaccine against ASF. Disease control is dependent on the awareness of practitioners and rapid diagnosis and enforcement of strict sanitary control measures.

ASF is a permanent threat for EU. Efforts to support the control of the disease in Eastern European countries should be increased.

Future research should include the feasibility of a vaccine. Some of the priorities in this field are: the role of virus and host genes in infection, the characterisation of ASF virus virulence factors, host response to infection and studies to elucidate pathways to block immune evasion.

11.5. Recommendations for an ASF diagnostic bank

A diagnostic bank should be available for emergencies. It should include antibody detection tests that are essential for diagnosis of the disease in case of chronic or unapparent forms and when virological tests are not available in the ASF NRL of the affected country. It should hold validated ELISAs with recombinant protein to carry out 100 000 analyses for antibody detection.

11.6. Conclusions

- There are no vaccines against ASF.
- A diagnostic bank should be established with antibody ELISA test kits for testing 100 000 pigs.

12. GENERAL ISSUES AND CONSIDERATION RELATED TO THE REGISTRATION OF VACCINES

The introduction of regulatory systems for human and veterinary medicines in the European Union has been driven by increasing recognition on the part of governments and consumers that medicines need to be safe, of high and consistent quality, and efficacious. Costs to meet these standards can usually be recouped for products with a substantial and predictable market**.** In contrast, for diseases which are exotic to the country or region concerned, there is little incentive for industry to invest in meeting the costs of authorising a vaccine, given uncertainties over use and financial return.

However, in the face of possible outbreaks of exotic diseases, the consequences of not vaccinating are increasingly unacceptable, given the risk of uncontrolled spread, and rejection of traditional slaughter policies. So veterinary authorities may seek to use an appropriate vaccine, whether or not it has met the usual regulatory requirements. The legal consequences of such action may, however, be significant and deter authorities from sourcing unauthorised medicines. The use of authorised vaccines provides an added level of assurance as to the safety of the vaccines themselves, and of products from animals treated with them. This is useful when seeking the support of retail organisations, stakeholder groups and independent food standards agencies.

There are two main routes to authorisation of veterinary vaccines within the EU. In the centralised procedure, the European Medicines Agency (EMA) receives applications to be

considered by the Committee for Medicinal Products for Veterinary Use (CVMP). Once the CVMP makes a positive recommendation, the European Commission issues a decision granting a marketing authorisation that is valid in all 27 EU Member States, as well as Norway, Iceland and Liechtenstein. Authorisation of vaccines to be used as part of Community campaigns through this procedure has many advantages. A single authorisation permits an identical product of proven quality to be available for use in all Member States. An alternative route is at national level, with extension to other Member States, via mutual recognition or through the decentralised procedure.

There are provisions in the existing regulatory framework to use vaccines without authorisation. Article 8 of Directive 2001/82/EC (as amended by Directive 2004/28/EC) permits Member States to use immunological medicinal products without authorisation 'in the event of serious epizootic disease'. Article 26 2001/82/EC for nationally authorised products, and Article 39 of regulation 726/2004 for centrally authorised products, provide for authorisations under exceptional circumstances for 'objective and verifiable reasons'. In such circumstances, some of the usual requirements for authorisation can be made into specific obligations to be carried out by the marketing authorisation holder as a condition of receiving authorisation. However, as these authorisations are reactive and usually follow disease outbreaks, they do not address the need to create an environment for authorising vaccines to anticipate disease outbreaks.

A recent development to address some of these difficulties is the revision of the technical requirements for authorisation of veterinary medicinal products within the EU (Directive 2009/9/EC). This has introduced the concept of the 'multistrain dossier', whereby a potentially large number of approved strains may be included within a single Marketing Authorisation (MA) and the final vaccines formulated to include one or more of the relevant strains. This initiative has been well received by industry and is well suited for development, approval and maintenance of vaccines against antigenically variable viruses (e.g. FMD, avian influenza and bluetongue). In addition, new strains may be added to the MA by means of a rapid regulatory procedure should new antigenic variants threaten the EU. However, these changes alone will not guarantee the availability of authorised vaccines for exotic diseases, nor will they ensure the addition of new vaccine strains for authorised products in the time frame needed by EU authorities.

Nevertheless, an additional requirement that vaccine banks should preferably only be stocked with authorised vaccines as either the final vaccine formulation or via authorised precursors would certainly act as a significant incentive to manufacturers to authorise their vaccines.

Another issue that should be explored during the current review of the legislation is the possibility of developing a fast-track procedure. This would involve a suitable Commission body considering information on products proposed for use in emergency situations and providing an opinion on their benefit:risk balance that falls short of a positive opinion for full marketing authorisation. Such an assessment could highlight key benefits and risks and propose risk management measures to assure safe use in emergency situations. Such a centralised EU opinion on emergency use would promote a more harmonised approach and provide assurance at Community level regarding the quality, safety and efficacy of products used to control important transboundary diseases.

12.1. Recommendations

Relevant legislation regarding veterinary medicinal products is not well suited to approve the use of vaccines in emergency situations. The current review is an ideal opportunity to introduce a mechanism for approving vaccines for emergency use at European level.

13. GENERAL ISSUES AND CONSIDERATION RELATED TO THE VACCINES INDUSTRY

To set up a vaccine or antigen bank, industry needs to respond to a relevant tender. There are many factors a manufacturer needs to consider when submitting an offer for an exotic disease. For EU-authorised vaccines, the size of the vaccine/antigen bank, the price per dose, the shelf life of the vaccine or antigen and renewal plans are important issues. For vaccines not yet authorised in the EU, they need to take into account the existence of a vaccine elsewhere in the world, the need to start a development programme, and standards relating to safety, quality and efficacy.

13.1. Vaccine bank

A cost-efficient method is a vaccine bank from 'rolling stock', where a company has ongoing production, and increases its reserve stock by the quantity of the tender. In this case, the company acts as the storage point for the tender stock. This avoids issues such as shelf life and is particularly efficient if the licence for the product is held by the owner of the bank. This could be the case with a Bluetongue vaccine, where the vaccine is being used on an ongoing basis in one EU region. A bank can be created for use should the disease spread to other regions, assuming an EU licence has been granted by the countries affected. However, the principle of rolling stock is only possible if the size of the bank is relatively small in relation to the regular supply of the vaccine to the regular market.

The advantage of a vaccine bank based on the rolling stock principle is that emergency vaccine manufactured, tested and released in accordance with European legislation can be supplied rapidly.

13.2. Antigen bank

Consideration needs to be given to the merits of creating an antigen bank versus a vaccine bank. An antigen bank has many advantages over vaccine banks. It is especially suitable if the vaccine has a short shelf life, or in case of diseases with antigenic variance, such as FMD, BTV, AI, AHS etc, where the formulation of the vaccine can be decided once the field virus has been typed.

However, the regulatory aspects of formulating an antigen into a vaccine in an emergency situation must also be considered. An antigen bank for emergency supply is based on the principle that the emergency vaccine can be produced within days and released for immediate use without re-testing the final product. The only disease for which there is a regulatory framework within the EU for rapid release of vaccine from pre-tested antigen is FMD (Ph. Eur. Monograph 0063.). To apply the principle of a vaccine or antigen bank for diseases other than FMD, the following regulatory aspects need to be addressed in European legislation:

- The legislation for a bank system should be applicable to banks held by a country, an international organisation, or vaccine-producing companies, and provide assurances to

stakeholders on the quality, safety and efficacy of the vaccine when used in the face of disease outbreaks.

- Industry should have rapid access to new and emerging disease pathogens to ensure rapid development of seed material as the first step towards developing safe, potent and effective vaccines against threats of disease in the EU.

- For emergencies involving new diseases or new serotypes, especially in the case of inactivated vaccines, methodology should be in place to authorise vaccines, based on a risk assessment, before all required preauthorisation tests are carried out. The release of vaccine batches on such a basis should be possible before all required batch release tests are carried out, as permitted for FMD in the Ph.Eur monograph.

- Legislation may be required to ensure release of emergency batches of authorised vaccine before all quality control (QC) test results are available. The principle of performing QC tests, including safety and potency, on a pilot vaccine batch representative for the emergency vaccine in 'peace time' enables release of authorised emergency vaccine within days following a disease incursion. The vaccine should be formulated, filled and shipped within days, and released on the basis of the pilot batch results.

- The shelf life of stored antigen or vaccine is determined on the basis of ongoing periodic testing and re-testing. It is important to take into account that banks need to be created for emerging diseases or emerging serotypes, for which there is often relatively little experience in antigen or vaccine bank construction.

13.3. Emergency development / conditional license / vaccine banks

When an outbreak of disease or new serotype occurs for which a vaccine is urgently required, there is much pressure from authorities to make vaccine available as early as possible and in as large a quantity as possible. Once vaccine is available, the political pressure significantly decreases and strict regulatory compliance takes the lead.

Over recent years, the vaccine industry has shown strong commitment to putting massive resources into the generation of data for an emergency license (e.g. AI, BTV).

To keep the industry committed to emergency development of vaccines against new diseases or new serotypes, the negative consequences of a conditional license need to be addressed:

- Emergency development takes place under considerable political pressure both for the authorities as well as for the industry. The regulatory review needs to take unplanned product development into account when reviewing its application. This must happen not just in the first year, when political pressure is high, but also in subsequent years, when the applicant is required to supply data that were part of the conditions for the licence.

- Emergency development of a vaccine involves taking short-cuts in the research. However, some of these may turn out not to be optimal. Frequently, products which were developed in an emergency are reviewed against the same standards as those that apply to normal development. It is recommended that variations be reviewed with a pragmatic approach, taking into account the circumstances that applied during emergency development.

- Certain emerging diseases may come and go. It may be that there is an urgent need for a vaccine in year one and two of an outbreak, diminishing thereafter. Nevertheless, the conditional licence requires full product development, even when there is no longer an acute need for the vaccine. The applicant has only two choices at present: (1) to fulfil the conditions of the licence and to finalise complete development or (2) to withdraw the licence and destroy the efforts to obtain it. It is recommended that there is legislation to freeze the conditional licence and the commitments of the company, to take into account that there may not be an acute need at any given time, while protecting a potential future need for a licensed product.

- The conditional licence can be revoked at any time when a licensed product becomes available. The vaccine industry prepares antigen and vaccine stocks on the basis of commitments from national authorities to purchase vaccine. Production of an inactivated vaccine takes 5-6 months. Experience has shown that a conditional licence is likely to be revoked within days after a licensed product is available. As a consequence, the authorities stop companies with a conditional license supplying the vaccine, even when there are commitments to purchase it. As a consequence a company might have to face complete destruction of a production pipeline of 5-6 months. It is recommended that there be a reasonable selling-out period after a licensed product becomes available, and that authorities fulfil their formal or informal purchase commitments.

The worst-case scenario is where a vaccine has to be developed, where the shelf life is short, where there is no commercial market, and where the material is not licensed. In this case, public funds need to be used to develop a product. A contract manufacturer needs to be found, along with means of storing the bank. The bank needs to be restocked as the shelf life of a vaccine expires, with associated disposal costs. As the material is not licensed, various trade issues need to be resolved if the vaccine is to be used, and these may be insurmountable with resultant trade restrictions and costs as a consequence of use.

On the cost front, the experience of the Commission and Member States in running existing banks is the best source of real-time information. Speculative figures based on various assumptions could be made, but real data from the Commission and Member States would be more valuable. An antigen or vaccine bank means costs for production, testing, storage, regulatory requirements, full licence etc.

The vaccine industry can only justify developing a vaccine against a new disease or new serotype if there is a financial incentive. If there is no vaccine against an emerging disease and no existing market for such a vaccine, there is no incentive for the industry. For such emerging diseases, vaccine development with public funding is recommended, linked to a commitment for an antigen/vaccine bank.

Consideration needs to be given to the merits of creating an antigen bank versus a vaccine bank. An antigen may be more suitable if the vaccine has a short shelf life. However, the logistics of formulating an antigen into a vaccine in an emergency situation must also be considered.

On the regulatory front, many issues need to be considered:

1. Who has responsibility for the seed/vaccine/antigen bank once manufactured? Can the bank be kept by an institute, organisation or another manufacturer, and be released as

authorised vaccine under the regulatory framework? How to maintain GMP compliance and sign-off by a Qualified Person is of particular relevance.

2. Is there provision to extend the shelf-life of the antigen/vaccine? Can the potency of the antigen or vaccine be retested over time, with the possibility of extending shelf life, based on extrapolation, if the material appears to remain potent at a fixed time point?

It is estimated that a full dossier development programme can take five to seven years. With a lot of effort and luck, timeframes may be reduced. Safety, quality and efficacy issues link into licensing issues are considered elsewhere in this report.

In terms of the size of a bank, estimates need to be made regarding the potential speed at which a disease may spread, along with the density of susceptible animals.

It may be of interest to know that the U.S. is developing a RVF vaccine. In the human field, the U.S. authorities have part-funded the building of a vaccine factory as a means of increasing critical capacity.

13.4. Conclusions for vaccine banks

The regulatory environment for the authorisation of vaccines for emergency use has faced a number of challenges over recent years given threats to the EU from diseases such as avian influenza and bluetongue disease. At the time of these threats, there were no EU authorised vaccines. Although there is provision in EU regulations to use unauthorised vaccines in the event of a serious epizootic disease, in reality, veterinary authorities or governments are reluctant to use products of unsubstantiated quality, safety and efficacy. The legal consequences of such actions are unknown if adverse events occur in vaccinated animals, the environment (particularly for live vaccines), or users of the vaccines. The public and other stakeholders also increasingly expect use of regulated medicines. This is especially true when meat and dairy products from vaccinated animals are consumed.

Until recently, EU regulations for veterinary vaccines for emergency use and, in particular, for antigenically variable viruses, have not offered industry much incentive to develop vaccines for exotic diseases, or to submit applications for authorisation to anticipate disease outbreaks, even when the EU has faced actual outbreaks.

Changes to the Directive and Technical Annex on veterinary medicines have introduced amendments to facilitate the authorisation of such vaccines. The changes include innovations such as the multistrain dossier and the possibility of granting authorisations for limited markets or under exceptional circumstances, subject to certain conditions being met post-authorisation. However, these changes alone will not guarantee the availability of authorised vaccines for exotic diseases nor, necessarily, ensure the addition of new vaccine strains for authorised products in the time frame needed by EU authorities.

Additional funding is needed for research and development of vaccines for exotic diseases, where industry involvement is uncertain. Furthermore, funds are needed to establish vaccine banks for diseases that threaten the EU. Resources need to be invested in determining the most cost-effective structure for such banks, as either master seed, antigen or final product vaccine banks. Determining the long-term real-time stability of antigens and/or vaccines is a major undertaking, and requires investment and resources on the part of industry to establish the claims for their products.

The availability of new field isolates for industry is important for developing vaccine master seeds, but this is only one step of many towards developing new vaccines, even if seeds for other strains or serotypes are used in the manufacture of existing products. Field isolates need to be adapted to the production system before being laid down as new master seed and tested to ensure the absence of potential extraneous agents. Laboratory and field safety and/or efficacy studies may need to be performed with batches of vaccine manufactured from the new master seed. Finally, the data must be compiled and submitted to satisfy the requirements of EU regulators before the product is authorised as an Exceptional or Full Market Authorisation. Therefore, master seed and antigen banks should not be seen as a short-cut to providing emergency vaccines in the event of a disease incursion, as the full regulatory requirements will apply for all possible bank scenarios.

The regulatory framework for vaccines derived from master seed, antigen or vaccine banks may need greater flexibility in future to encompass the various possibilities for establishing such banks within the private and public sector. However, whatever structures and systems are developed to provide vaccines for disease emergencies in the EU, they must ensure that:

- Governments and stakeholders have confidence in the quality, safety and efficacy of the products.

- Vaccines supplied from final product vaccine banks or manufactured from master seed or antigen banks can be batch released as EU-authorised products to meet the specifications and requirements of the Market Authorisation, Good Manufacturing Practice and Qualified Person.

- Animal health and welfare is respected and where possible a ‘vaccinate-to-live’ policy is implemented, with no trade implications (development of DIVA vaccines and robust diagnostic assays).

- Meat and dairy products from vaccinated animals can be supplied to consumers through the usual supply channels without fear of boycotts or significant price variations.

14. GENERAL ISSUES AND CONSIDERATION RELATED TO THE DIAGNOSTIC INDUSTRY

The character of the diagnostics industry, which consists mainly of SMEs, makes the idea of a diagnostics bank useful. Diagnostics manufacturers, like many other life science industries, tend to keep stocks as low as possible. The availability of products is mainly based on forecasts. If an unexpected outbreak occurs, the diagnostic tools needed may not be in stock, and be available only after a delay of a couple of weeks, in which precious time is lost, giving a disease time to spread. A diagnostic bank offers immediate availability of products to control the spread of disease.

The limitations that were previously mentioned for the constitution of vaccine banks mainly apply to diagnostics. Most diagnostic kits have to be stored either cold or frozen. Shelf life is also limited, and kits generally expire within 6-24 months.

Today, there are diagnostics available for AI, FMD, and CSF. A tender should include the number of tests and the required time frame, as products usually expire within less than a year.

The vaccine bank should not be limited to DIVA diagnostics, as only few diseases could become candidates. Not many DIVA diagnostics have been developed recently, as they require close cooperation between vaccine companies and diagnostic manufacturers, which does not happen on a daily basis. This is partly due to the dissociation of the vaccine and diagnostics industries that has taken place. Cooperation between the industries is crucial and needs to be put in place while a vaccine and its associated DIVA diagnostic tool are being developed.

Furthermore, there is not enough development of diagnostic tools for emerging diseases. This is because of the high risk as regards return on investment. Such insecurity means there is little stimulus to develop new diagnostic tools.

A diagnostic tool can be developed more quickly than a vaccine, potentially in as little as 6-8 months, provided that sufficient knowledge is available (nucleic sequences, specific antibodies and antigens), and development does not stumble on unforeseen complexities. This means a disease could be diagnosed and controlled by protective or prophylactic measures using diagnostic tools, especially if vaccines are not available.

The research funding programme of DG Research (FP7) should be involved in calls to which vaccine and diagnostics manufacturers could respond.

15. ANNEX 1: DISEASES OF TERRESTRIAL ANIMALS WHICH ARE SUBJECT TO NOTIFICATION ACCORDING TO COUNCIL DIRECTIVE 82/894/EEC OF 21 DECEMBER 1982 ON THE NOTIFICATION OF ANIMAL DISEASES WITHIN THE COMMUNITY

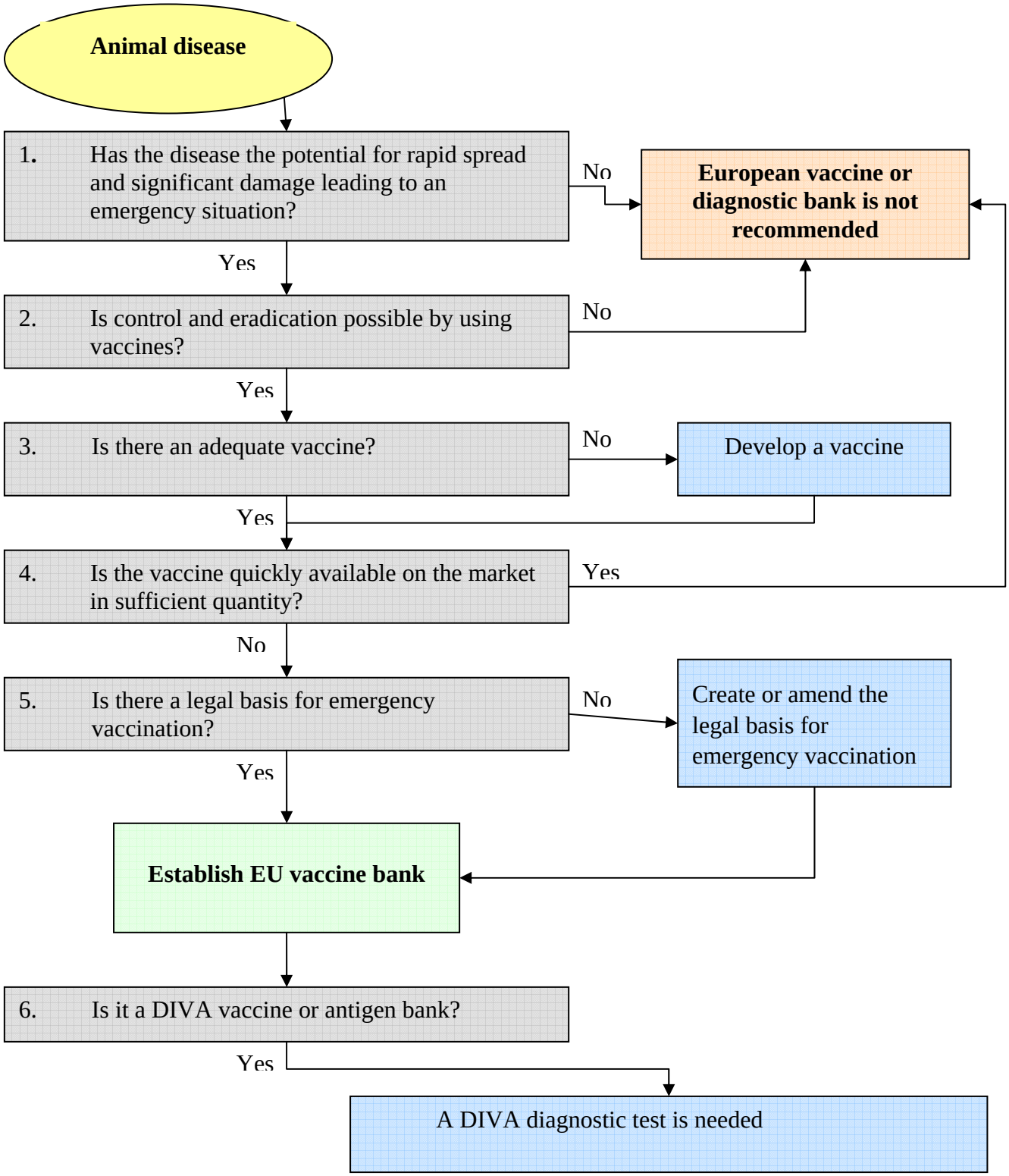
African horse sickness
African swine fever
Avian influenza
Bluetongue
Bovine spongiform encephalopathy
Classical swine fever
Contagious bovine pleuropneumonia
Dourine
Equine encephalomyelitis (of all types, including Venezuelan equine encephalomyelitis)

- Equine viral encephalomyelitis
- Eastern equine encephalomyelitis
- Western equine encephalomyelitis
- Venezuelan equine encephalomyelitis
- Borna virus disease
- WNF? (not in the Netherlands)

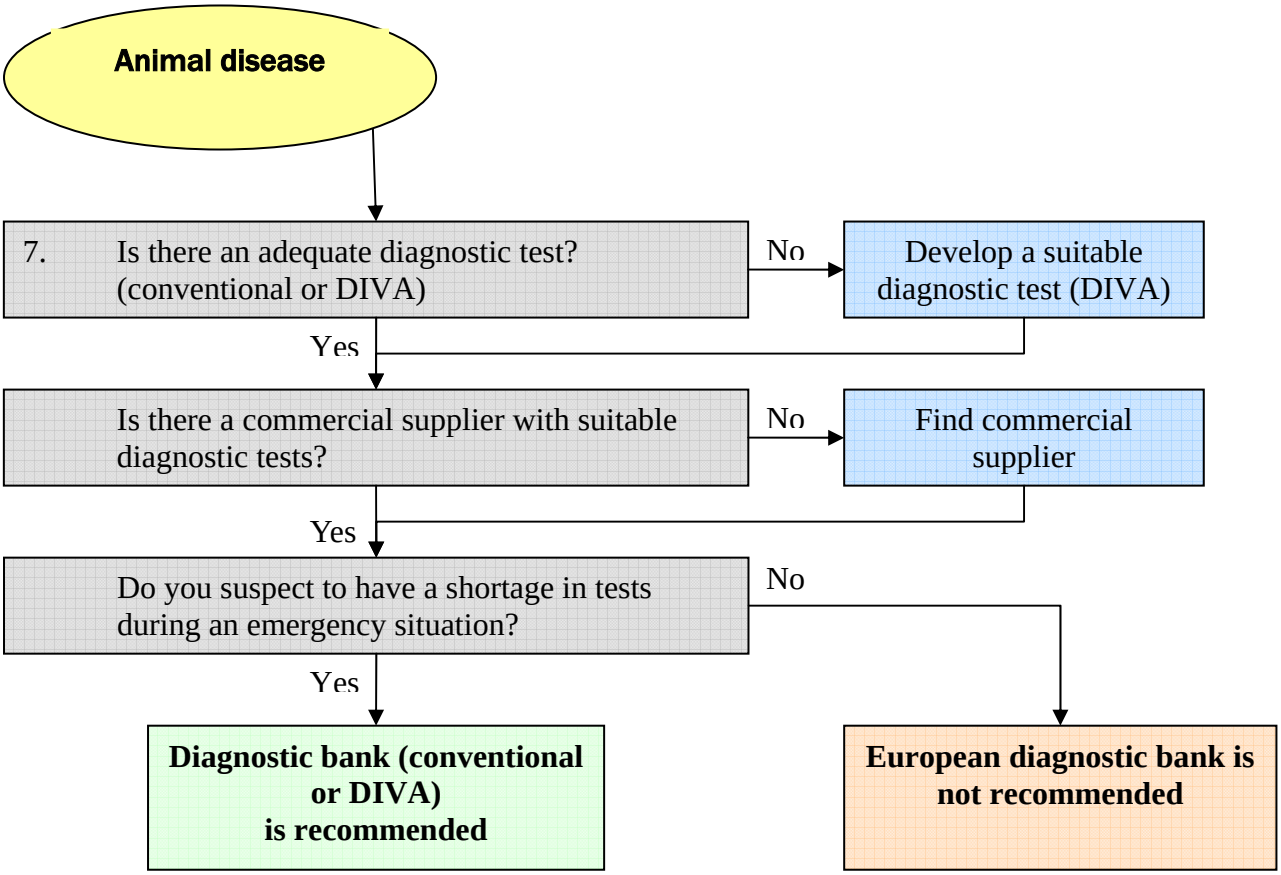
Equine infectious anaemia
Foot-and-mouth disease
Glanders
Lumpy skin disease
Newcastle disease
Peste des petits ruminants
Rift Valley fever
Rinderpest (cattle plague)
Sheep and goat pox (Capripox)
Small hive beetle (Aethina tumida)
Swine vesicular disease
Tropilaelaps mite
Vesicular stomatitis

16. ANNEX 2: DECISION TREES

16.1. European vaccine bank



16.2. European diagnostic bank



17. ANNEX 3: OVERVIEW TABLE OF VACCINES AND DIAGNOSTICS

Diseases	**1. Spread/ severity /likelihood**	**Number of relevant serotypes**	**2 Emergency vaccination strategy useful**	**3 + 4 Vaccine available**	**Vaccine Storage time (years)**	**5. Legislative Framework**	**Size of recommended vaccine bank**	**6. DIVA possible**	**7. Regular diagnostic tests available**	**Test shortage suspected = diagnostic bank recommended**
African horse sickness	High	9	Yes	Yes (KV in Egypt, LV)	?	needed	150.000	In house tests	Yes	
African swine fever	High	1	-	No	-	-	Needed but not possible	-	Yes	
Avian influenza	High	2 (H5&H7)	Yes	Yes (KV)	Limited	Yes	40.000.000	Yes	Yes	
Blue tongue	High	24	Yes	Yes (KV, LV) Limited available	Limited	Yes	??	Limited serology + PCR	Yes	
Classical swine fever	High	1	Yes	Yes (KV, MLV)	Yes	Yes Needs amended	2.000.000	Yes Genetic DIVA	Yes	
Contagious bovine pleuro-pneumonia	Low			Yes (LV)						
Dourine				No						
Foot-and-mouth disease	High	7	Yes	Yes (KV)	≥ 5	Yes	Exists and needed	Yes	Yes	
Glanders				No						

Diseases	**1. Spread/ severity /likelihood**	**Number of relevant serotypes**	**2 Emergency vaccination strategy useful**	**3 + 4 Vaccine available**	**Vaccine Storage time (years)**	**5. Legislative Framework**	**Size of recommended vaccine bank**	**6. DIVA possible**	**7. Regular diagnostic tests available**	**Test shortage suspected = diagnostic bank recommended**
Lumpy skin disease	Low	1	No	Yes (MLV, LV)	?	No	No	No	Limited	
Newcastle disease	Low to high	1	Yes preventive strategy	Yes (KV)	?	Yes	0 in sufficient quantities available	No	Yes	no
Pest de petit ruminants	Medium	1	No	Yes (MLV,LV	?	?	0	No	Yes	
Rabies	High	**1**	Yes preventive strategy	yes			0 in sufficient quantities available			
Rift valley fever	High	?	No	Yes (KV, MLV)	?	No		No	Limited	
Rinderpest	Low		Yes	Yes (LV)		No	0			
Sheep and goat pox	Low to medium			Yes (MLV, LV)						
Small hive beetle	Low	1	NA	No	No	NA	N A	N A	Yes	
Vesicular stomatitis	Low	2	-	Yes (KV in Colombia and Venezuela)	-	No	0		Yes	

Diseases	**1. Spread/ severity /likelihood**	**Number of relevant serotypes**	**2 Emergency vaccination strategy useful**	**3 + 4 Vaccine available**	**Vaccine Storage time (years)**	**5. Legislative Framework**	**Size of recommended vaccine bank**	**6. DIVA possible**	**7. Regular diagnostic tests available**	**Test shortage suspected = diagnostic bank recommended**
West nile fever	Medium	1	No	Yes (KV)	1	No EU leg framework	0	yes	yes	

1. Has the disease the potential for rapid spread and significant damage?
2. Is control plus/minus eradication necessary and possible by using vaccines?
3. Is there an adequate vaccine?
4. Is the vaccine quickly available on the market in sufficient quantity?
5. Is there a legal basis for emergency vaccination?
6. Is it a DIVA diagnostic test?
7. Is there an adequate diagnostic test?

MLV = modified live vaccine
LV= Live vaccine
KV = Killed vaccine
NA= Not applicable