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Working Document on

Eradication of Bovine, Sheep and Goats Brucellosis in the EU

accepted by the "Bovine" and "Sheep and Goats" Brucellosis subgroups of the

Task Force on monitoring animal disease eradication

This document does not necessarily represent the views of the Commission Services

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1. Introduction

The eradication of brucellosis offers considerable benefits to society by eliminating one of the most serious zoonoses, therefore improving animal health and welfare, trade and productivity.

The purpose of this document is:

- to review the general principles that constitute the basis for strategic planning for surveillance, control and eradication of brucellosis
- to propose short/medium/long term measures based on specific epidemiological situation in order to accelerate the progress of eradication programmes
- to draw conclusions on perspectives on eradication/control
- to stimulate discussion on future strategy
- to assist in the design and improvement of eradication programmes.

This document is primarily based on conclusions and recommendations presented in various Task Force meetings and on the experience of the Member States (MSs).

The Task Force for Monitoring Animal Disease Eradication (TF) was created in 2000 as an action foreseen in the Commission White Paper on Food Safety (action plan proposed by the Commission and agreed by all MSs).

The aim was to create an additional tool mainly to assist with management of eradication programmes co-financed by the Community.

The objective is to improve both the effectiveness and the cost-benefit of co-financed eradication programmes as well as provide technical support to those MSs that still not have co-financed programmes.

The TF operates through plenary meetings and experts subgroups: two brucellosis subgroups have been created (see point 1.3) to deal with Brucellosis in bovines and small ruminants respectively.

Any proposed disease eradication or control measures must be assessed based on the individual situation in each MS or region running an eradication programme for bovine and/or sheep and goats brucellosis, especially in those still having significant disease prevalence.

1.1. Definition

Brucellosis is an infectious and contagious disease caused by bacterial species of the genus Brucella (except B. ovis). It is a major zoonosis with an important social and economic impact (direct and indirect losses).

There are six species known to potentially cause human disease and each of these has preferred animal hosts: *B. melitensis* in goats and sheep, *B. abortus* in cattle and buffalo, *B. suis* in pigs, *B. canis* in dogs and *B. ceti* and *B. pinnipedialis* in marine animals. *B. microti* and *B. neotomae* occur in wild rodents but have not been implicated in human infection.

For the purposes of this document, discussion is restricted to brucellosis in bovines, sheep and goats.

1.2. EU Legal framework

The Community legal framework on bovine (BB) and sheep and goats (S&GB) brucellosis (**listed** in **Annex I**) is formed by:

- 1. Legislation on trade of bovine, ovine and caprine animals
- 2. Legislation on animal products for human consumption (meat and milk)
- 3. Legislation on Community co-financing of eradication programmes
- 4. Legislation on reporting of zoonoses
- 5. Legislation relating to official controls for the Brucellosis CRL
- 6. Legislation approving vaccines against bovine brucellosis within the framework of Council Directive 64/432/EEC (Commission Decision 2002/598/CE)

In addition the following standards of the Terrestrial Code and Manual of the World Organisation for Animal Health (OIE) have to be taken in account:

- World Organisation for Animal Health (OIE). 2008. Bovine brucellosis. Terrestrial Animal Health Code. 17 ed. Articles 11.3.1.-11.3.9.
- World Organisation for Animal Health (OIE). 2008. Caprine and ovine brucellosis (excluding *Brucella ovis*). Terrestrial Animal Health Code. 17 ed. Articles 14.1.1-14.1.9.
- World Organisation for Animal Health (OIE). 2008. Bovine brucellosis. Manual of Diagnostic Test and Vaccines for Terrestrial Animals (mammals, birds and bees). Chapter 2.4.3. pgs: 624-660.
- World Organisation for Animal Health (OIE). 2008. Caprine and ovine brucellosis (excluding *Brucella ovis*). Manual of Diagnostic Test and Vaccines for Terrestrial Animals (mammals, birds and bees). Chapter 2.7.2. pgs: 974-982.

The EU legislation on trade and the OIE Code for bovine, ovine and caprine brucellosis, which defines the areas, herd and animals status regarding brucellosis has been frequently used for eradication purposes with the aim of gaining free status as soon as possible. However, as presented in this document, eradication requires more specific measures which need to be adapted to the situation in each Member State or region.

1.3. Task Force subgroups on brucellosis, CRL and scientific input

Two brucellosis subgroups were created: the bovine brucellosis (BB) and sheep & goat brucellosis (S&GB) subgroups with the purpose of assessing and advising Member States and the Commission on brucellosis eradication programmes. Meetings take place in those Member States where programmes are in force and include central and local veterinary authorities, laboratories, veterinary practitioners and stakeholders. As a result, a report that includes conclusions and recommendations is provided to the Member States and published online, in order to strengthen programme performance.

Between 2000 and 2009, the BB TF subgroup has met 13 times and the S&GB TF subgroup met 11 times.

The large number of meetings held and the outcomes reflecting the different epidemiological situations in the MSs visited, have provided the opportunity to design/update strategies which are more adapted to the heterogeneous situation of the MSs that are implementing eradication programmes as well as for those MSs that still do not have a programme in place or that have to carry out control/surveillance.

Full information on the activity and the reports of the Task Force and of these subgroups is available at http://ec.europa.eu/food/animal/diseases/eradication/taskforce_en.htm.

In addition, the outcomes of the 2 workshops organised by the Community Reference Laboratory for Brucellosis (EU Community/OIE & FAO Reference Laboratory - French Food Safety Agency - AFSSA, Maisons-Alfort, France) held in 2007 and 2008 provide for, *inter-alia*, an important input for coordination for the diagnosis of Brucellosis (ring trial involving all National Reference Laboratory) in the MSs and candidate countries.

Some important outcomes from the "Brucellosis 2008 International Research Conference" held in London from 10 to 13 September 2008 and past similar conferences also provide well focused input for the design/update of future eradication strategies.

1.4. Brucellosis status of EU Member States

The current brucellosis status of the EU Member states is listed in **Annex II**. A more complete overview is available in the "The Community summary report on trends and sources of zoonoses, zoonotic agents, antimicrobial resistance and food-borne outbreaks in the European Union in 2007" prepared by the European Food Safety Authority (EFSA) (http://www.efsa.europa.eu). More detailed information at MS or regional level may be contained in the eradication programmes of the MS approved for co-financing which are available in the DG Health and Consumers website.

2. Strategies to enhance the effectiveness of brucellosis eradication programmes

The main aspects to be considered, based on the experience, conclusions and recommendations of the two brucellosis subgroups, are the need for a thorough evaluation of the epidemiological situation in the region or MS, correct choice of an epidemiological unit and sound epidemiological parameters. This should be done before a strategy is defined, or an eradication plan is designed and implemented. Other aspects of importance are the availability of high quality laboratory diagnosis; appropriate slaughter, stamping out and compensation policies; animal movement control; the ability to carry out checks in the entire animal population, use of vaccination, an adequate organisation of the veterinary service and good stakeholder involvement.

For eradication programmes to be effective, the following characteristics of brucellosis must be taken into consideration:

- Disease spreads easily, particularly at time of calving, between and within herds and flocks;
- Clinical signs are not pathognomonic and may be unapparent;
- Infected females do not always abort;
- Latent carriers usually occur;
- Transmission occurs both horizontally and vertically; through direct or indirect contact;
- No diagnostic tool correctly identifies all infected or non-infected animals;
- Available vaccines dramatically reduce the spread of brucellosis but do not fully protect against infection.

Within a strategic eradication plan two groups of issues/measures are identified:

- Issues to be addressed in a general context;
- Short/medium/long term measures based on the epidemiological situation.

2.1. Issues to be addressed in a general context

The EU strategy is not aimed primarily to support control programmes but are focused on the achievement of the total eradication of brucellosis.

However, effective control of the disease may be essential as a preliminary step. The intermediate target is a rapid increase in the percentage of Free (BF) and Officially Free (BOF) herds. This leads to the recognition of BOF regions prior to the complete eradication of brucellosis from the territory of the EU.

Therefore a progressive strategy has to be set up, leading to the following objectives:

- 1. Primary objective in a very high prevalence area: control of the infection;
- 2. Mid / short-term objective: regular and rapid decrease in the true incidence and prevalence;
- 3. Final objective: eradication.

Four main issues in control/eradication programmes have been insufficiently addressed in several Member States where brucellosis remains as a problem. These are:

- Adequate organisation of veterinary services and stakeholder involvement.
- Epidemiological evaluation of the disease situation.
- Diagnostic capacities and coordination with veterinary services.
- Control of animal movements, implementation of the pre-movement testing in areas of high prevalence, animal/herd identification.

2.1.1. Adequate organisation of the veterinary services and ensuring stakeholder involvement

It is essential that the veterinary services are effective and organised adequately with sufficient staff and available budget to manage the programme so as to implement all the measures adapted to the specific epidemiological situation (whole population surveillance, movement control, animal identification, epidemiological investigation, etc.). Staff should be periodically trained and audited in order to guarantee the highest standards of performance.

Good organization, administration, coordination and supervision of the activities of the programme and an effective means of interaction and communication between all the parties concerned (appropriate capacity of laboratory network and veterinary authorities) are essential aspects.

The nomination of a single Programme Director/Coordinator could be of great help in countries with a decentralized political organization. The respective roles of the different competent authorities involved (central/regional/local) should be clear, recognized and reinforced when necessary. Full enforcement of current legislation is necessary along with an ongoing review and, when necessary, amendment, of the legislation.

A continuous review of the effectiveness of the measures implemented to ensure their adaptation when required should also be in place.

All stakeholders should be involved in the eradication programme, independent of their respective roles and responsibilities and actively commit and contribute to the full implementation of all the measures of the programme. Therefore, the programme should clearly define the tasks and duties for each of the partners. Co-operation with stakeholders is of paramount importance. Flowcharts of data, information and communication between stakeholders should be effectively established, and regularly revised.

An awareness campaign should be carried out by the central/local veterinary authority to ensure that each individual player is aware and has an understanding of his/her responsibilities and what is expected from him in the context of his/her involvement in the programme. This is better attained by adopting a holistic approach to the programme's management and delivery at the level of the farm, private veterinarians, official veterinary services, laboratories and local, regional and national administration. This overall approach should clearly define these responsibilities and ensure the sustained commitment of all concerned parties.

The efficacy of measures implemented at herd level depends on the active participation of private practitioners and farmers. The involvement of farmer associations may greatly help in this context.

In order to ensure that the stakeholders fully understand their role in ensuring the success of the eradication programme and to obtain the highest degree of commitment, the programme should include specific positive stimuli that apply to each participant so as to encourage their sustained participation and contribution to the progress of the programme. At the same time, sanctions or corrective actions should also be foreseen so as to avoid certain actions and/or to address any perceptions and attitudes that could limit the accelerated elimination of the disease.

Specifically, positive stimuli/sanctions should be linked to the maintenance of sanitary status and the fulfillment of legal requirements dealing with the implementation of compulsory measures (reporting of abortion, animal vaccination, disinfection activities, animal culling, etc).

The authorities in charge of the programme have an opportunity to re-design a programme that incorporates a system of "rewards" and "penalties" aimed at encouraging other stakeholders to take due account of their role in the eradication of brucellosis and to fully co-operate with the implementation of all of the measures contained in the programme.

2.1.2. Epidemiological evaluation of the disease

An adequate understanding of the epidemiology of brucellosis is critical to its successful control and eradication. This disease is not homogeneous in a given country/region because it occurs in different epidemiological situations and farming methods, therefore requires adapted control/eradication measures, which may differ between and within countries. The EU cattle, sheep and goat farming industries present an enormous variety of breeding systems and environmental conditions. This diversity affects the distribution and the evolution of the disease in the EU. As a consequence, the primary steps of an eradication programme are: to acquire knowledge of the situation and to define the epidemiological units of intervention.

The primary unit of concern or epidemiological unit for brucellosis is the herd, the flock or the group/holding including all epidemiologically-related susceptible animals.

"Herd" in the legal context of 64/432/EEC is defined as "an animal or group of animals kept on a holding (within the meaning of Article 2 (b) of Regulation 21/2004/EC) as an epidemiological unit; if more than one herd is kept on a holding, each of these herds shall form a distinct unit and shall have the same health status".

From an epidemiological point of view in the context of brucellosis eradication, the epidemiological unit should be defined as "any number of animals that are held, kept or handled in such a manner that they share the same risk of exposure to brucellosis" and therefore is the unit of concern for application of control and eradication measures.

In practical terms, two or more groups of animals belonging to the same owner but separately managed without any other link or contact between them could constitute two or more epidemiological units, whereas two groups of animals belonging to different owners but kept

together, or in contact with one another or with common management, constitute a single epidemiological unit. When production conditions result in direct or indirect contact between animals then consideration must be given to regarding them as one epidemiological unit.

From an epidemiological perspective, common grazing or fragmentation of holdings and/or the presence of management links between farms are obstacles to brucellosis eradication. Therefore it is necessary to define the epidemiological unit in a clear way, using appropriate criteria.

The use of appropriate indicators to evaluate the individual components/measures of the programme is necessary for a correct assessment of the management of the programme at regional/national level, with a view to identifying how different issues currently posing obstacles for eradication can be addressed. However, some of the more relevant indicators may require additional information to be recorded or analysed. In any case a good information system is required to enhance the quality of the epidemiological data.

Basic indicators for the follow-up of the co-financed eradication programmes are provided in Commission Decision 2008/940/EEC.

Measuring progress concerns more than simply assessing data that are easy to retrieve. It also requires an evaluation of the effectiveness of the measures currently in place. Therefore, more appropriate, and sometimes more sophisticated, indicators adapted to the needs of each programme, could be developed and applied at MS level. An appropriate epidemiological information system should be created connecting all levels of intervention.

If adequate information is not available, well designed epidemiological studies should be conducted to identify and quantify risk factors. The outcome of these studies should then be used to define the best strategies. It is important to have in place an adequate information system to ensure the quality of epidemiological data.

Changes in strategy should always be based on epidemiological evidence or informed by the best available knowledge.

In addition, there is a need to investigate any new outbreak, using an adapted epidemiological inquiry to identify contact animals or units. This will allow further identification of sources of infection that should be subject to control. A model of such an inquiry form has been prepared by the subgroups and is **presented in Annex III**

Finally, in officially free regions or MSs, efforts should be concentrated on identifying remaining potential sources of infection in order to prevent new outbreaks or the re-introduction of disease. This need, *inter-alia*, a permanent knowledge of the status of areas of destination of herds/flocks transhuming from free areas (exchange of information between provinces/regions), regular testing before/after movements and investigation of other potential reservoirs, including wildlife.

2.1.3. Diagnostic capacities and coordination with veterinary services

Diagnosis is another important tool in the control and eradication of brucellosis.

The European Commission supports MS in respect of brucellosis diagnosis, not only through direct financing but also by establishing a Community Reference Laboratory (CRL) for the disease, presently located in France (Laboratoire d'études et de recherches in pathologie animale et zoonoses – AFSSA, Maisons-Alfort). It is very important that the National References Laboratories (NRLs) are in regular contact with the CRL, attend the annual workshops organised by CRL and participate in the ring trials assays. The NRLs have to fully comply with their tasks.

Diagnostic activity should be based on a solid laboratory network of local and regional Labs coordinated by the NRL at national level, and by the CRL at European level. The co-operation amongst NRLs and the CRL is recognized as essential in order to achieve the best diagnostic standards in the European Union.

Whenever feasible, diagnostics efforts have to be focused on the confirmation of infection in the epidemiological unit, especially in the latter stages of eradication programmes. Unequivocal diagnosis of *Brucella* infections can be made only by isolation and identification of *Brucella* spp.

Bacteriological investigation requires good practice in organ collection and should include specific target organs such as head/mammary and genital lymph nodes, genital secretions and milk or udder and genitalia samples.

Several **serological tests** are available with good diagnostic performances at herd level. However, no single test is able to identify all infected animals (*i.e.* imperfect sensitivity), or assure with 100 % confidence that all tested negative animals are free of brucellosis. As a consequence, test combinations and repetition within a reasonable time may be necessary under certain epidemiological conditions.

Tests combinations (series or parallel) are a key tool to increase the predictive values of the test results. Testing in parallel will increase sensitivity and is therefore very useful in infected or high risk herds or regions. Test result interpretation should always take the following elements into account:

- Percentage of positive tests results, disease prevalence and incidence;
- Presence of clinical signs (abortion);
- Vaccination strategy;
- Known risk factors;
- Status of the herd, the area, the country.

The choice of tests and interpretation of results should always be based on epidemiological data and be set out in the eradication programme. For example, the diagnostic strategy may differ according to the objective (detection of infected herds, detection of infected animals, clarification of suspicion of false-positive reactors etc).

The performance of the available tests is not an obstacle for eradication if specific diagnostic strategies are adopted according to the epidemiological situations. However, there may still be a need for more research on the diagnostic field in order to address the remaining gaps.

False positive serological reactions may occur with any test. While of low importance in infected areas, these reactions are of concern in free or almost free areas. In these circumstances, strategies should be designed to rule out infection, by, for example, combining bacteriological investigation with epidemiological data and serological follow-up, etc.

2.1.4. Control of animal movements, pre-movement testing, animal/herd identification

The control of animal movements between herds, and especially from farms or regions with a high prevalence of disease is a basic principle of animal disease control and is a necessary and highly effective measure. The use of derogations provided for in EU legislation regarding pre-movement tests should be considered only for herds in low prevalence areas and in the context of a sound epidemiologically rational approach.

The application of pre-movement testing reduces the risk of spread of brucellosis between herds/holdings or within the herd (if several distant land parcels are used) and provides additional assurance for the purchaser in this regard. It also serves indirectly as an additional assessment of the

brucellosis status of the herd and region of origin. This particularly applies to the movement of animals into and between herds/holdings in areas or regions of high prevalence. Likewise, the movement of animals from unrestricted herds in these regions to herds outside these regions poses a relatively high risk of spread.

The permanent, individual identification of animals such as to allow a strict control of the animals/herds is a very important programme element. In spite of Community legislation, this aspect is often not fully addressed in the eradication programmes.

Movement restrictions result from the application of Directives 64/432/EEC and 91/68/EEC that restrict the movement of animals from herds/holdings not qualified as OBF/OBmF. These minimum movement restrictions alone may not be effective when particular epidemiological conditions prevail. Practices such as transhumance, the use of common grazing areas or the inclusion of a number of fragments of land as components of an epidemiological unit result in additional difficulties that are not always taken into account when the implementation of the movement restrictions is considered, especially in areas or regions of high prevalence.

2.2. Eradication measures

This section discusses eradication measures more specific to Brucellosis eradication which are largely in line with the conclusions and recommendations drawn up previously by the Task Force. The implementation of most of these is feasible and necessary to achieve eradication.

The epidemiological situation should drive the choice of measures for brucellosis control/eradication and results should be evaluated continuously.

Those measures are:

- Vaccination
- Slaughter/stamping out in infected herds
- Frequency /repetition of herd testing and the use of tests in association
- Appraisal of compensation scheme.

A justification for not applying specific basic measures should be provided by the competent authority.

2.2.1. Vaccination

Vaccination is often the first step in the control of infectious diseases. Vaccination against brucellosis increases the resistance to infection and decreases abortion risk and the excretion of the agent, therefore decreasing the disease incidence in the human population, but it is insufficient on its own to eradicate the disease.

There are advantages in maintaining vaccination up to or after eradication is achieved, because it ensures that the population remains protected against any unidentified remaining source of infection.

Effective vaccination is achieved when two requirements are fulfilled: vaccination coverage is >80% of the eligible animals and vaccination is performed for a period greater than twice the average production life (>10 years in sheep and goats).

The choice of the vaccine tool also should be linked to the capability of the veterinary services to efficiently implement a test-and-slaughter programme. Under conditions of high to moderate prevalence, inadequate movement control or limited diagnostic capabilities, mass vaccination of all

animals (including adults) is the optimal tool for reducing the level of infection. When used exhaustively in the whole flock incidence greatly decreases. Once the herd prevalence has been reduced, more effective control of the disease may be achieved through the implementation of a programme based on vaccination of young replacement animals combined with test-and-slaughter of adults. These programmes should be planned in the light of the area status, reflecting the risk of infection, instead of the holding status.

It should be stressed that having a population well immunised against brucellosis makes the implementation of other sanitary measures more effective. In this way, the cost effectiveness of the eradication programme can be greatly increased.

In a high-prevalence, endemic disease situation in small ruminants (>5% herd prevalence, depending on the epidemiological situation, where a test-and-slaughter programme cannot be properly implemented and/or progress is not observed), mass, i.e. whole flock, vaccination with Rev.1 vaccine is recommended as an emergency measure, which should be carried out in as short a time as possible. This should be reinforced with parallel measures such as movement restrictions and control of common grazing and later be followed by vaccination of replacement animals. The transition from mass vaccination towards young replacement vaccination should be planned in advance. Clear milestones for every major change in the programme should be set in advance and accepted by all involved parties.

In a **high-prevalence situation in cattle** a vaccination programme of replacement females with S19 or RB51 vaccine combined with test-and-slaughter could be applied.

If mass vaccination is necessary, RB51 is the only available tool because of the absence of interference with conventional serological diagnosis (RBT and/or CFT). Cattle can be re-vaccinated twice if required. Mass vaccination should be followed by vaccination of replacements, with the transition being planned in advance. Clear milestones for every major change in the programme should be set in advance and accepted by all involved parties. Additional measures such as depopulation, movement restrictions and control of common grazing should also be considered.

When a re-emergence of disease occurs in a territory where the disease was controlled, and initial measures are not enough to control the spread disease, emergency vaccination should be envisaged.

In case of **moderate to low herd/flock prevalence** (for example, <5%, depending on the epidemiological situation) and the presence of important risk factors (movements, outbreaks), a programme combining vaccination of young replacement animals (with S19 and Rev.1 vaccines, for cattle and for sheep and goats respectively) with test and slaughter in adults is recommended. Where risk factors cannot be controlled (for example, under conditions of transhumance), vaccination is recommended even when the prevalence is lower.

It must also be pointed out also that the RB51 vaccine for cattle has been used with success in some parts of the EU and could be a useful tool for eradicating bovine brucellosis from well-controlled epidemiological units, provided that it is applied massively and regularly for a sufficient period and in conjunction with a severe test-and-slaughter programme. In other conditions, S19 vaccine should be preferred.

The subsequent test and slaughter policy has to be based on the chosen vaccination strategy, taking into account the persistence of residual antibodies due to the vaccine. As a general rule, animals should be tested six to twelve months after vaccination, depending on the route of administration of the vaccine. For sheep and goats, the use of the conjunctival route minimises interference with serological testing. Tests interpreted in parallel to increase sensitivity will hasten eradication but will increase costs due to the use of multiple tests and an associated reduction in specificity.

The use of parallel testing enables swifter eradication and protection of herds/flocks. The requirements for success are:

- Regular testing (at least annual, better twice a year) of all adult animals, always in line with the provisions of Dir 64/432/EEC and Dir 91/68/EC;
- Rapid culling of positive animals (depopulation if needed);
- Re-test positive herds at short intervals (every 2 months).

The currently available vaccines, which are officially authorised at European level for the prophylaxis of brucellosis (S19 and RB51 strains for cattle, and Rev.1 for sheep and goats) can potentially provoke two types of adverse effects: the induction of abortion in pregnant females, and in the case of the smooth strains (S19 and Rev.1) the persistence of residual antibodies to the classical tests (RBT and CFT). This latter that may cause diagnostic difficulties in certain situations but can be avoided by the use of conjunctival route for vaccine administration and restrict the age of application to 3-4 months, maximum 6 months.

2.2.2. Slaughter/stamping out in infected herds: criteria, application and assessment

When the prevalence of the disease is very low or even close to zero, an exclusive test and slaughter policy or stamping out should lead to the eradication of the disease and gaining Officially Free status. To ensure success, such eradication programmes require proper organisation of the veterinary services and laboratories, good co-operation with the stakeholders as well as movement control, individual identification, well-organised national data-base and an adequate budget. Disinfection of premises and leaving infected pastures empty for at least two months are other essential additional measures.

Stamping out of the infected epidemiological unit is a major but very efficient disease eradication measure, provided that the infected herd is rapidly removed from the farm and that restocking is not a means of re-introducing the infection.

In the case of depopulation, is very important to pay attention to complementary measures such as thorough cleaning and disinfection of the premises, a delay on the re-use of common grazing areas, checked and strengthened bio-security measures, an empty period before repopulation, amongst others.

The decision whether to use stamping out should be made by the veterinary services, based on the epidemiology of the disease and the capacity of the veterinary services. Factors that should be taken into consideration include the disease prevalence in the area, the within-herd prevalence, contact with other susceptible species, the interval before restocking, herd size, enterprise type and the type of husbandry, prevailing bio-security measures, farm security in relation to contiguous holdings and the ability and willingness of the herd-owner(s) to conform with conditions of the stamping out protocol. Also, account should be taken of any additional criteria assessed by the local veterinary services regarding the decision to proceed with stamping out. The capacity of local veterinary services for a concerted action between all entities involved in stamping out and their capability to maintain adequate surveillance of herds/flocks is also an important consideration for the decision to use stamping out.

The decision to adopt a stamping out policy should be formalised in the form of guidelines that identify the criteria to be considered when extensive spread of infection within a herd or group of herds or epidemiological units is diagnosed. Such guidelines would be required for detailed consultation by local or regional veterinary officers when considering whether or not stamp out.

2.2.3. Frequency/repetition of herd testing/use of tests association

The minimum frequency of tests on herds/animals is laid down in Community legislation.

However, an increase in the frequency of testing (as well as the use of parallel testing), together with rapid removal of positive animals, reduces the incidence and the prevalence of the disease, by enabling earlier detection of infected herds/animals and their early elimination.

Another aspect that should be considered is the procedure for a herd/unit re-gaining the OBF status under certain conditions. The minimum procedure laid down in Directives 64/432/EEC and 91/68/EEC may not be the optimum in all cases. More stringent strategies such as a longer interval between tests before this status is regained, or the introduction of an additional test before OBF status is regained, could be applied under certain difficult conditions. These strategies could be applied in MSs which are not OBF and in particular, in the case of herds in areas or regions of high prevalence, as a further means of reducing the likelihood of re-infection and movement of infected cattle out of such areas.

2.2.4. Appraisal of compensation schemes

Compensation schemes should be linked to the level of the herd-owner's cooperation with the eradication programme as well as to the progress of the eradication.

Once adequate compensation is approved, its payment should be swiftly accomplished. Adequate compensation implies that the compensation paid does not pose an obstacle for the progress and success of the programme.

The compensation scheme should be aimed at modifying the behaviour of the farmers in a way that they do their best to avoid the reintroduction of the disease in their herds. Consequently, a level of compensation that is perceived by the farmers not to be sufficient to allow them to adjust to their new circumstances following the loss of their stock and the imposition of animal movement restrictions, along with concern associated with the possible reintroduction of the infection, jeopardizes the progress of the programme by engendering an attitude of non-cooperation.

Accordingly, it is extremely important to ensure that the level of compensation is appropriate and serves to encourage farmers to respond to their situation in an appropriate manner that will prevent or considerably reduce future risk of infection. Furthermore, compensation should always be at a level below (to a reasonable or, sometimes, significant extent) that of the current market price of comparable healthy animals.

3. Conclusions and Recommendations

Eradication of brucellosis in both cattle and sheep and goats is achievable, as it has been demonstrated in most countries and regions within the European Union. No single strategy is the optimal applicable to all MSs, due to the great diversity of farming systems and associated differences in the epidemiological situation of brucellosis that occur. Legislation defines the conditions under which countries, regions or herds/flocks can be recognised as free of brucellosis and establishes trade rules. However, further measures, as outlined in this document, are required to be implemented to accelerate eradication of the disease.

3.1. Conclusions

- The measures in the eradication programme (national/regional legislation) should empower the veterinary services to define the appropriate epidemiological unit as the primary unit of concern for all measures within the programme. This key issue should be addressed, taking into account the prevailing practices used in animal production in each MS/region. Expertise on epidemiology is needed at MS level to identify and provide the most appropriate indicators for each epidemiological situation.
- The herd prevalence level of the region and the type of movements where the pre/post-movement test should be implemented has to be defined. In addition, stricter specific procedures for regaining the OBF/OBmF herd/holding status in high prevalence areas or, in restricted herds/holdings before the return to trading, may need to be applied.
- Individual and permanent identification that permits a strict check of the animals/herds is necessary. This aspect is often not fully addressed in the eradication programmes of the MS.
- Appropriate guidelines that are epidemiologically reliable and which take into account all
 relevant criteria should be drawn up at national /regional level and formally applied when
 deciding whether or not to implement herd depopulation as a component of the eradication
 programme.
- The optimum ratio of herd prevalence and the frequency/repetition of testing on herds in regions of high prevalence, and the use of tests combinations, taking into account the local prevailing conditions, needs to be established and continually reviewed in line with the progress of eradication in these regions. The reproductive cycle and husbandry practices should be used to determine when tests should best be carried out. Epidemiological models could help to establish a theoretical optimum frequency of testing under certain epidemiological circumstances.
- Compensation schemes should be thoroughly and constantly reviewed, taking into account
 local conditions so as to ensure that the amounts to be paid are adequate and that the payment is
 linked to full compliance with stated measures aimed at the prevention of re-infection. Overcompensation should be avoided. Farmers have to be promptly informed on future changes.

3.2. Recommendations

- 1. Each country's eradication programme should be based on a strategy that reflects the epidemiology of the disease, the farming system and veterinary infrastructure present in the region or country. The strategy should be regularly reviewed and the programme adapted accordingly.
- 2. The veterinary services should define the appropriate epidemiological unit as the primary unit of concern for measures applied within the programme.
- 3. Expertise in epidemiology should be available at MS/regional level to identify the most appropriate indicators for each epidemiological situation.
- 4. The testing strategy (test type, frequency and test combinations) should be defined to optimise sensitivity or specificity according to the objectives outlined in the strategic plan. It must also address the risk of false sero-negative results in infected animals (e.g. latency, incubation period; pregnancy) and false-positives in the late stages of eradication. Specific more stringent procedures for regaining the OBF herd status in high prevalence areas with certain epidemiological conditions or, in restricted herds before the return to trading, may need to be applied.

- 5. Appropriate guidelines that take account of all the relevant epidemiological criteria should be drawn up at national /regional level to determine the eradication programme (vaccination, test-and-slaughter, stamping out) to be implemented within countries or regions.
- 6. Compensation schemes should be regularly reviewed to ensure that the amounts payable are reasonably sound and that the payment is linked to full compliance with stated measures. Overcompensation should be avoided.
- 7. Experience acquired by the MSs and the Commission after more than twenty years of cofinanced programmes in brucellosis and the present working document leads to the conclusion that the existing EU legislation could usefully be revised.

4. ANNEX I

European Union Legal Framework on bovine and sheep and goats brucellosis

A. Legislation on trade of bovines, sheep and goats

- Council Directive 64/432/EEC lays down measures on:
 - Requirements for trade of bovines based on herd qualification. The procedures for gaining, maintaining, suspending, withdrawing or regaining the Officially Brucellosis Free (OBF) status (based on serological/milk tests at herd level) (Annex A, II) A Member State or part of a Member State may be declared OBF
 - Diagnosis of bovine brucellosis in cattle (Annex C modified based on EFSA opinion 2006 by Commission Decision 2008/984/EC).
- Council Directive 91/68/EEC lays down measures on:
 - Requirements for trade of sheep and goats based on holding qualification. The procedures for gaining, maintaining, suspending, withdrawing or regaining the *Brucella melitensis* Officially Free (OBmF) status, based on serological/milk tests at holding level. (Annex A) A Member State or part of a Member State may be declared OBmF
 - Diagnosis of S&G brucellosis (Annex C)

B. Legislation on animal products for human consumption (meat and milk)

- Regulation (EC) No 853/2004 of the EP and of the Council: laying down specific hygiene rules for food of animal origin (Annex III, section IX, chapter I).
- Regulation (EC) No 854/2004 of the EP and of the Council: laying down specific rules for the organisation of official controls on products of animal origin intended for human consumption (Annex I, section IV, chapter IX).

C. Legislation on vaccines

- Council Directive 91/68/EEC, Council Directive 64/432/EEC and Commission Decision 2002/598/EC approving vaccines against bovine brucellosis within the framework of Council Directive 64/432/EEC

D. Legislation on Community co-financing of eradication programmes

- Council Directive 77/391/EEC which introduced Community measures for the eradication of TB, Brucellosis and EBL in cattle: MSs are obliged to draw up programmes for accelerated eradication (financial contribution Community). Amended and completed by Directives 78/52/EEC, 82/400 and Decision 87/58/EEC (provide additional legal framework for the eradication TB, Brucellosis and EBL in cattle.
- Council Decision 90/242/EC which introduced Community measures for the eradication of Brucellosis in sheep and goats: MSs may submit eradication programmes (financial contribution Community)
- Council Decision 2009/470/EC on expenditure in the veterinary field defines Community financial measures on eradication and monitoring programmes aimed at progressively eliminating animal diseases that are endemic in certain areas of the Community
- Standardisation of eradication programmes:
 - Community criteria for eradication and monitoring programmes (Commission Decision 2008/341/EC)
 - Standard requirements for the content of the programmes (Commission Decision 2008/425/EC
 - Standard reporting requirements for the programmes (Commission Decision 2008/940/EC

5. ANNEX II

Current brucellosis status of the EU Member States

A. BOVINE BRUCELLOSIS

Officially Bovine Brucellosis-free (OBF) MSs

(data June 2009)

Belgium, Czech Republic, Denmark, Germany, France, Ireland, Luxembourg, Netherlands, Austria, Poland, Slovakia, Slovenia, Finland, Sweden, Spain (Canary Islands autonomous region), Italy (65 provinces), United Kingdom (Great Britain), Portugal (6 Autonomous region of Azores). June 2009: Ireland

Member States with Community co-financed eradication programmes in 2009

Spain, Italy, Malta, Cyprus, Portugal and United Kingdom

B. SHEEP AND GOATS BRUCELLOSIS

Officially B. melitensis free (OBmF) MSs

(data June 2009)

Belgium, Czech Republic, Denmark, Germany, Ireland, Luxembourg, Hungary, Netherlands, Austria, Poland, Romania, Slovenia, Slovakia, Finland, Sweden, United Kingdom, Spain (Canary Islands autonomous region), Italy (54 provinces), France (64 Départements) and Portugal (Autonomous region of Azores)

• Member States with Community co-financed eradication programmes in 2009

Spain, Italy, Cyprus and Portugal

6. ANNEX III

Epidemiological investigation of brucellosis outbreaks/suspicions Task Force Brucellosis sub-group (March 2001)

A Introduction

The epidemiological investigation constitutes an essential tool for the control of brucellosis, whatever the sanitary status of the considered zone and the control strategy. It enables, at herd/flock level, (i) the confirmation or invalidation of a suspicion, (ii) the determination of the origin of infection, (iii) the search for other herds/flocks epidemiologically related to an infected one and (iv) the evaluation of disease spread and the holding characteristics to decide on control strategies (i.e. slaughter of reactors or depopulation). At local level, the analysis of data collected in a group of holdings contributes to the identification of patterns of disease and the monitoring and adjustment of strategies.

The efficacy of eradication measures implemented in outbreaks depends on its adequacy in relation to real situations. The correct evaluation of this depends on the quality and the exhaustiveness of the investigation. It is therefore essential to carefully collect all the necessary information in order to make the good choice for action. Furthermore, the increasing awareness for the importance of a deeper participation of farmers and assistant veterinarians in programmes activities, makes the epidemiological investigation a very important tool: the direct observation at the farm of management practices and other factors that might constitute risk for brucellosis spread, is a important moment for sanitary education and involvement of these key partners.

The objective of the present work is to help Member States to improve its own investigation procedures by setting up a series of considerations on brucellosis investigation, especially on the design and implementation of questionnaires at farm level. The principles orienting the investigation, possible methods, data collection and management have to be adapted to each epidemiological and structural situation but a minimum level of analysis of situations is desirable to improve the achievement of the objectives of brucellosis programmes.

This document consolidates the principles and methods discussed through the enumeration of important risk factors for brucellosis infection and transmission, the examples of a "minimum" and a "extended" questionnaires and, in annex, the questionnaires currently in use in some Member States, with different perspectives, reflecting the epidemiological situation of each country and the organisation of operational procedures.

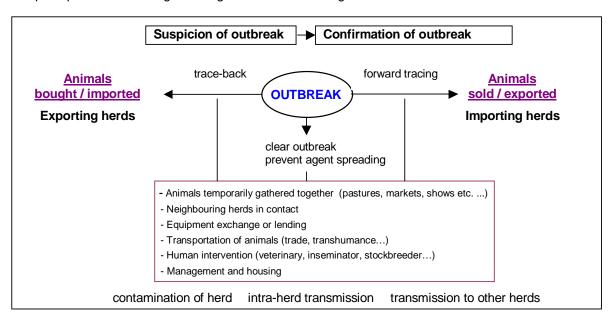
B Implementation of the epidemiological investigation

1 – Principles (WHY?)

The main objectives of the epidemiological investigation are:

- Determining the origin of a brucellosis outbreak (trace-back);
- ➤ Identification of risk factors at farm level for intra-herd transmission, evolution of outbreak, possible escape routes of the agent and risks for human health;
- Identification of other herds/flocks epidemiologically linked to the initial outbreak (forward tracing or follow-up).

The principle of the investigation is given on the following chart.



➤ <u>Trace on survey</u>

The objective of this trace on investigation (forward tracing) is the identification of herds/flocks or places of accommodation of animals potentially contaminated from the initial outbreak. It provides the opportunity to identify potentially exposed populations, which may lead to control measures designed to prevent secondary or tertiary outbreaks.

It is necessary to search for:

- Animals sold/exported since the assumed date of the start of infection,
- Neighbouring herds/flocks in contact (pastures, etc.),
- Herds/flocks at least temporarily gathered together with the infected herd/flock (common pastures, transhumance, shows, markets, etc.)
- Animals transportation (trade, transhumance)
- Equipment/materials exchange or lending
- Contamination through human intervention (veterinary, inseminator, stock-breeder, etc.)

Taking in to account the incubation period for brucellosis, date of parturitions/abortions and production cycles (mating period, synchronization of oestrus, transhumance period, etc.), the survey should search for data for the two years before the outbreak finding date (or even more when history of the holding is not reliable),

➤ Trace back survey

This trace back survey is designed to locate the origin of an outbreak, *i.e.* to identify the probable source(s) of the outbreak and consequently other outbreaks having the same origin. It would be necessary to exclude previously the possibility of resurgence of the disease in a previously infected herd/flock.

The period of inclusion would be of at least 2 years. All animals' introductions and the common grazing periods for several herds/flocks should be investigated for the inclusion period.

The possibility of contamination from materials lending or exchange and human interventions should be taken in account as well.

In case of irregular introduction or absence of movement authorisation, the sanitary status of the herd/flock of origin should be controlled. When no obvious origin can be assumed, the last-two year's history of the sanitary status of the herds/flocks of origin of all animals introduced for the last two years should be traced and checked.

In case of an important period of time between the exit of the animal(s) out of the selling/exporting breeding and the entrance in the buying/introducing one, it would be necessary to investigate the different places of accommodation of the animals and to take in account all the risks linked with the trade and transportation of animals.

> Evolution of outbreak and intra-herd transmission assessment

Data on occurrence of serological positivity and some risk factors considered important for the persistence of brucellosis have to be evaluated in order to evaluate the likelihood of having infected incubating animals disseminating infection and in order to adjust possible measures (decide on stamping out, change of management practices, cleaning and disinfection, etc.).

Some of these factors are:

- Size of herd as a conditional factor for management;
- Existence of segregation facilities (premises, fences) and disinfection feasibility;
- Seasonality of parturitions: extended lambing seasons allow for the existence of females in different susceptibility status while others are giving birth; for example, young females mated some months after adult females might be in late pregnancy when adult females start to calve, being highly susceptible to infection;
- Place of parturition, determining the level of contamination of the environment and possible disinfection practices;
- Disposal of infected material (afterbirths, abortions, etc.);
- Management of new-born and replacement females and its exposure to infection;
- Dates of parturitions, abortions, inseminations.

2 - Methods

a. Investigation protocol

The investigation protocol should define who is in charge for the activity, in which conditions should it be applied and in what form, how the work should be performed and evaluated. The epidemiological investigation is based on data collection using the available sources, including the implementation of a questionnaire at farm level, the analysis of data and the production of useful information for helping the clearance of the outbreak, the trace-back and the forward tracing.

WHO should apply the protocol?

The investigation protocol should be developed by the official veterinarian in charge of implementing the restriction status at the farm, after the suspicion/identification of positive animals.

In case of situations where only vaccination is applied, the investigation is reduced to a minimum for the establishment of the profile of the herd. The field veterinarian can do this work.

To WHICH farms should the protocol be applied?

The investigation protocol should be performed in all herds with suspicion/confirmation of brucellosis, as a result of a positive direct or indirect diagnostic test result.

> In areas of high disease prevalence

In areas of high prevalence the implementation of an epidemiological investigation might not be practical, requiring a great investment in terms of human resources and time and being of limited importance for the determination of the origin of the disease. However, considering the other objectives and advantages of such work, a simple questionnaire, collecting data on the most important management aspects and risk factors, should be implemented. The follow-up of an infected herd should not be only a routine of blood sampling, test and slaughter: it is necessary to make interventions in other factors facilitating the persistence of brucellosis and this is based on a correct identification of problems at farm level.

> In areas with low prevalence

In low prevalence areas and in pre-eradication phase, the epidemiological investigation if very important to identify all possible sources of infection and all possible spreading of the agent beyond an infected farm. A careful investigation will allow the identification of false positive herds as well as the rapid confirmation of infection with the quick implementation of containment measures.

➢ In brucellosis free areas

This investigation appears as well essential in brucellosis free areas when it is necessary to determine whether positive serological results could be due either to an infection with *Brucella* or to a cross-reacting bacterium (false-positive serological reactions as those due to *Yersinia enterocolitica* O: 9). In those brucellosis free areas, where the intervals between two successive controls are generally enlarged, the detection of an outbreak could be delayed. Therefore it is essential to implement such investigations in order to identify rapidly all herds/flocks likely to be infected.

HOW should the protocol be applied?

After clear definition of the objectives of questionnaires and the information to be obtained by the epidemiological investigation, determine what are the necessary data to be collected and design the data collection forms and questionnaires.

Analysis of questionnaires to obtain the necessary in formation should also be planned in advance and databases should be designed (using database management programs or spreadsheet programs) and adjusted to the questionnaires.

The following phases are the data collection and the implementation of questionnaires, to check them and introduce into database. Data can be collected at the official veterinary services, the laboratory and the field veterinary services (either public or private). It is always of advantage to have the farm profile and history prior to the farm visit.

The final phases are data analysis and comment of resulting information.

b. Epidemiological questionnaire

The epidemiological survey is performed using a standardised questionnaire as those proposed in the following section.

Questionnaire design theory can be found in most epidemiology books. Questions should be formulated taking into consideration the advantages and disadvantages of each type of questions (open, closed, open-ended, etc.).

The model questionnaires presented in the following session, which apply to a detailed investigation (low prevalence area), consists of two parts:

- The first part corresponds to a first evaluation of the epidemiological context and is filled taking in account the information issued from the herd/flock's file made up by the veterinary services. Other data could be collected from other agricultural services, organisations, stockbreeders' associations, etc.;
- The second part could be filled by data collected during a visit performed by the veterinary services on the holding.

The questionnaire should be filled out in a uniform way in order to standardise the survey methods and to allow the subsequent analysis of the collected data.

For this purpose, the explanation of the objective of each question, the meaning of data to be collected and the best way to collect should be discussed with all the professionals involved.

The detail on data collection on each item can be adapted to the epidemiological situation and the capacity of implementing and analysing the data collected.

A list of important aspects for the epidemiology of brucellosis is also presented, in Table 1. These aspects are divided into 7 subjects, and classified with 1 to 3 stars according to the level of detail required in high prevalence areas (less detail) and low prevalence areas (more detail).

It is important to take into account the possible existence of more than one group of ruminant species in the same holding, i.e. small ruminants, cattle, buffaloes, that might be kept in different facilities within the same holding, with different control strategies and identification systems. This might require the use of different sources of data but the epidemiological questionnaire should clarify the situation in order to make sure that serological testing and restrictions are applied to all susceptible animals living in the holding.

Table 1. Aspects of importance in brucellosis epidemiology

Subject	items	import
	identification (number, name of owner), geographical location	***
	veterinary services (producers organisation, name of vet)	***
The farm	sanitary status (date of last status, serological history, restrictions, vaccination)	***
	human cases	**
	products leaving the farm	***
	herd structure (number of animals by class)	***
	change in herd size (dates, reasons)	**
The herd / flock	sanitary status of animals (vaccinated, non vaccinated)	**
	clinical signs (abortion, stillbirths, infertility, etc)	**
	other species and sanitary status; contacts with herd	**
	purchasing of animals (number, dates, purposes, origin) (2 breeding cycles)	***
	other introductions of animals (offers, lending, borrowed back, etc.)	**
	transhumance and common pastures / watering points	***
Trace back	status of neighbouring farms and contacts	**
	other possible contacts (markets, shows, common milking, AI, etc.)	**
	contacts with other animal species	*
	human intervention (vets, inseminator, visitors) and equipment exchange	*
	animals sold (number, dates, destiny)	***
	contacts (transhumance, common pastures, milking, markets, neighbours, etc)	***
Forward tracing	selling of milk, fresh cheese, matured cheese, and other products	**
	manure and other possibly contaminated by-products	**
	human intervention and equipment exchange	*
	lambing season (duration, different seasons for adults and young females)	***
	parturition (place and hygiene)	**
	management of lambs and young replacement females	**
Intra-herd transmission	milking practices	**
1141131111331011	manure and waste material	*
	cleaning and disinfection	**
	wildlife	*
	resurgence	***
	neighbouring	***
	introduction of animals	***
Conclusions	common pastures or watering points	***
	transhumance, mixing flocks	***
	other sources of infection	***
	progress of trace back	**
Measures	progress of forward trace	**
	progress of outbreak clearance	**

c. Consequences

- In case of suspicion, the results of the survey will support the sanitary decision. The qualification of all herds/flocks epidemiologically linked to the outbreak should be suspended
- Serological controls should be performed in these herds/flocks on all adult animals.
- The sanitary authorities concerned with herds having animals issued from the infected herd/flock(s) should be informed rapidly.
- Corrective measures following the identification of management practices favouring the spreading of the agent should be implemented.
- The analysis of data collected will allow the correct evaluation of the epidemiological situation of brucellosis in the area.
- Adjustments of the programme made with good quality data will improve the achievement of expected results.

QUESTIONNAIRE FOR EPIDEMIOLOGICAL INVESTIGATION IN CATTLE HERDS AND/OR SHEEP AND GOATS FLOCKS SUSPECTED TO BE INFECTED OR INFECTED BY BRUCELLOSIS

- Billy goats:

- Number of vaccinated animals: sheep...... goats goats

Other spe	cies			01-1	Para Danas Hara'	_
Cattle - C	Cows			Status regard	ding Brucellosi	S:
- (calves <1 y					
Pigs:						
Poultry:		- husbandr	y methods: .			
Dogs:						
☐ free		ied up stray do				
- rece	nt serological	controls: No Ye	es (dates, r	esults, treatme	ents):	
Horses:						
Cats:						
Other spec	cies:					
Presence of	of wildlife:					
Plac	es of breedir	ng other than the f	f arm No	Yes	Place:	
• Presen	t sanitary sta	tus of the herd/flo	ock			
□ oʻ	fficially free		□ suspecte	ed of being info	ected 🖵	other:
☐ fr	ee		☐ infected			
Date of	beginning of p	oresent sanitary sta	atus/	/		
		·				
• Is the c	lata collected	revealing brucell	osis in the l	nerd?		
	⊒ No					
	⊒ Yes	Date:/	1	Origin:		
_	1 163	Date/	./	Origin		
		oats tested:				
		sitive animals: ons due to <i>Brucella</i>		acad?	☐ Yes	□ No
	Slaughter:		a been evidei	icea?	u res	□ NO
	3 33	□ Partial				
			- No. slaug	htered animals	S:	
	01 1 1					
	• Sneds nav	ve been cleaned ar No			whom:	
	◆ Date of th	o hord ro-qualificat	ion: /	1		
		e herd re-qualificat ons:				

•	Last	annual	controls	results

Date	No. tested animals/total	Serological results	Comments

• Introductions (purchase/importation)

	Regularly	Irregularly	Explain when irregular
Animals purchased/introduced within the year			
Animals purchased/introduced within year y-1			
Animals purchased/introduced within year y-2			
Animals purchased/introduced within year y-3			

- ◆ Compare the list of the last two-years' movements with the list of controls for trade and the identification Nos. of the herds/flocks of origin.
- Has the flock/herd been transhuming for the last 3 years?

□ No	Yes
------	-----

Dates	Area	Other herds/flocks' owners	Other herds/flocks' sanitary
(from to)		names	status

Relationship with neighbouring	pastures c	luring the period of tr	ranshumance:
No 🗖	Yes □ (Give a m	(Sheep/Goats □ ap)	Cattle □)

Actual conditions of the transhumance

a) On foot way

Date	Route	Other breeders a	associated E	Breeders having preceded t	he herd
	List of stops	during the ro	ute *	on the route * *	
Names of br	eeders whose herds/flo	ocks have been mixed	with the conce	rned herd/flock during the	Α
edestrian route	COGOTO WITOGO HOTGO/III	one have been mixed	With the conce	inoa nora/nook aaning tir	•
	eeders whose herds/fl	ocks have preceded th	ne concerned h	erd/flock on the same	
edestrian route.		·			
າ) Livestock tru	ck transportation				
Nama and addra	as of the transporter				
vame and addre	ss of the transporter:		•••••		
_ivestock trucks	cleaned before the load	ding of animals Yes	□ No		
		g			
Places and dates	s of animals unloading:				
c) Events occur	red during transhuma	ance			
Albanda a 🗖		End to divin	П		
Abortions ☐ Presence of stray	, dogs □	Early lambing Presence of wi			
rieselice di sila	y dogs 🗖	Flesence of wi			
Serological contr	ols on sheepdogs	Yes □ No □			
Green een					
Comments: .					
		. l lli			
• Lenaing, renti	ng, boarding on othe	r nolaings			
Date	Identification Nos. of	Name of importing	Sanitary sta	tus of Controls perforn	ned afte
(from to)	moved animals	herd/flock	importing he		
,					
· · · · · · · · · · · · · · · · · · ·					
Has the herd s	status been withdraw	n for administrative r	easons?		
		n for administrative r	easons?		
Has the herd s	☐ Yes	n for administrative r	easons?		

- Grounds:

• Are serolog	gical tests for	trade control positi	ve?	
□ N □ Y	es	- Date:/ - Details:		
+Observation	ns:			
• The suspic	ion is based o	n:		
* annual s	erological testir	ng		
□ N □ Y		- Results:		
* epidemic	ological link with	an outbreak		
	es - Informatio - Place of t - Date of ou - Kind of ep	on date:// he outbreak: utbreak discovery: oidemiological link:	/	erd identification number:
- Results of herd/flock	serological ·	tests performed in	n case of epiden	niological link with another infected
Date	Test	No. tested animals	No. positive animals	Comments

• Supplementary information

- Map giving the location of pastures
- Map giving the location of sheds, pens
- Map giving the location of all pastures of the area (especially those for transhumance).

QUESTIONNAIRE FOR EPIDEMIOLOGICAL INVESTIGATION IN CATTLE HERDS AND/OR SHEEP AND GOATS FLOCKS SUSPECTED TO BE INFECTED OR INFECTED BY BRUCELLOSIS

Date:	2 nd Section	on – SURVEY PER Name of inve	-	HOLDING	
А	Il data collected in the first	section should be o	checked with the he	erd-owner/holder.	
• Locat	ion of livestock buildings	\$			
Breedii	ng methods				
	. Permanent sheepfold (no	grazing)		. Use of common paths $\ \square$	
	. Winter sheepfold			. Summer transhumance	
	. Summer sheepfold			. Winter transhumance $\ \square$	
	. Permanent grazing	(Renting Owr	ner 🗆)		
- Wateri	ng . water conveyance . well . possible contact with o	other flock: 🔲 Yes	. pond . river s 🖵 No		
- Manur	e				
	. Storage: on the cou		mulated litter on lands under	other cultivation	ed 🗖
- Premis	ses' cleaning frequency:				
- Disinfe	ecting frequency:	Chemica	ls used:		
	Service provider (give doc	umentary evidence):		
- Numbe	er of premises per species	(ownership or rentir	ng):		
Sh	eep: Goa	ts:	Cattle:	Other species:	
- Map Io	ocating the premises (1/100	000):			
- Numbe	er of pastures plots:				
- Мар о	f pastures				
- Presei	nce of a herd for fattening:	☐ Yes	□ No		
	dispensation:	☐ Yes	□ No		
<u>C</u>	bservations:				

- Census of animal p			
- Checking of the bree	eding registe	er (stock list of an	imals):
- Anomalies evidence	ed and obse	rvations:	
Other species			
 Introductions for 2 	, youro (way	ing, boaraing, ic	
- Complete info		lected in section 1	
- Complete info		lected in section 1	
- Complete info		lected in section 1	
- Complete info		lected in section 1	
	ormation col	lected in section 1	
- <u>Animals sold for 3</u>	years		:
- <u>Animals sold for 3</u>	ormation col	date of sale	dealer or introducing herd identification N
	years		:
- Animals sold for 3	years		:
- Animals sold for 3	years		:
- <u>Animals sold for 3</u>	years		:
- <u>Animals sold for 3</u>	years		:
- <u>Animals sold for 3</u>	years		:
- <u>Animals sold for 3</u>	years		:
- <u>Animals sold for 3</u>	years		:
- <u>Animals sold for 3</u>	years		:

·		<u>ras</u> (transnumance, lending, boa									
- Is the herd/flock gathered together at least seasonally with other herds/flocks?											
□ No	☐ Yes		⊒Yes □ No								
Precise and complete if necessary the data collected in section 1											
• Neighbouring contacts											
List of the nearest holdings (premises and pastures)											
Herd identification No.	Place	Status regarding brucellosis	Remarks on possible pathological events								
Map locating neighbouri	ng holdings										
- Premises	- Pastu	ires									
(point out roads, lanes, tra	cks, waterways, wate	ring places, etc.)									
List of risk factors for pro	obable contamination	on from external plants or holding	gs								
- Artificial insemination: No	Yes 🗆	Observations:									
- Natural mating: No	o □ Yes □	Observations:									
etc.):		tainment apparatus, tractor, manur									
		eterinary, technicians, inseminator's									
		**									

- Herds gathered together:

- Transhumance:
It would be useful to point out on a map (at least 1/25000): - waterways, - the holding's pastures (including those far away from the main holding), - the location of neighbouring holdings (use the same No. as in the previous table).
◆ Identification of cases of brucellosis on people in contact with animals
Observations made by the veterinary
• <u>Observations made by the farmers' organisation</u> (it would be useful that the visit be done together with a representative of this organisation)
CONCLUSIONS
◆ Results of the survey
Risk factors identified from the survey performed on the holding:
☐ Evidence of a previous brucellosis outbreak
☐ Animal(s)' introduction from an infected herd/flock
□Lack of serological control of introduced animals
☐ Herd probably gathered with animals from infected or unqualified herds/flocks
☐ Probable neighbouring contact with an infected herd/flock
□Others (give information please):