

**SUBMISSION OF ERADICATION PROGRAMMES FOR  
CATEGORY B AND C DISEASES OF TERRESTRIAL ANIMALS  
BASED ON GRANTING DISEASE-FREE STATUS AT  
ESTABLISHMENT LEVEL**

Template in accordance with Article 10 of Commission  
Implementing Regulation (EU) 2020/2002

1. Date of submission: 31/05/2021
2. Member State: Greece
3. Name of the disease: **Bovine brucellosis**
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5. Territorial scope with a description and demarcation of the geographical and administrative areas covered by the eradication programme and the names of the zones and regions, if more than one region is included in the territorial scope of the programme:

Programme	Region	Regional Unit
<b>Eradication</b>	Whole country	
<b>Eradication programme with RB-51 vaccine</b>	Thessaly	Larissa (Municipality of Elassona)
	Central Macedonia	Pella, Pieria, Thessaloniki
<b>Vaccination with Rev-1, only in the mountainous area of these RUs</b>	Western Greece	Aetoloacarnania
	Central Greece	Evrytania, Fokida, Fthiotida
	Epirus	Ioannina, Preveza, Thesprotia
	Thessaly	Karditsa, Larisa, Magnesia, Trikala
	Central Macedonia	Imathia, Pella, Serres, Thessaloniki
	Western Macedonia	Florina, Grevena, Kastoria, Kozani
	Eastern Macedonia and Thrace	Drama, Evros, Kavala, Xanthi

The Bovine Brucellosis eradication programme is a compulsory programme applied to all kept bovine animals in the whole territory of the country.



**Map I. Administrative division of Greece in Regions**



**Map II. Administrative division of Greece in Regional Units.**

6. A description of the epidemiological situation for each zone or region, if more than one region is included in the territorial scope of the programme:

Epidemiological situation for 2020 (Infection with <i>Brucella</i> spp.)										
Region	Health status									
	Unknown		Disease free*		Officially free		Officially free in suspension		Infected	
	N.E.	N.A.	N.E.	N.A.	N.E.	N.A.	N.E.	N.A.	N.E.	N.A.
Attica	1	18	0	0	32	2.302	11	123	5	829
Peloponnese	181	4.702	10	361	136	4.875	94	1.827	1	132
Western Greece	137	1.638	17	1.572	121	6.920	0	0	1	33
Central Greece	16	28	5	274	43	4.863	190	5.724	2	246
Epirus	143	4.871	16	578	90	6.270	39	1.610	1	70
Thessaly	35	1.880	0	0	57	1.707	150	4.480	1	250
Central Macedonia	276	10.945	16	839	2.172	120.667	149	3.670	17	1.207
Western Macedonia	0	0	0	0	664	22.380	27	152	4	109
Eastern Macedonia and Thrace	0	0	1	97	1.693	69.033	933	23.340	41	2.306
Northern Aegean	552	6.130	0	0	4	24	10	115	0	0
Southern Aegean	1.197	16.286	135	995	177	1.024	0	0	0	0
Crete	15	21	3	20	23	139	16	109	0	0
Ionian Islands	162	3.202	6	106	23	695	0	0	0	0
<b>Total</b>	<b>2.715</b>	<b>49.721</b>	<b>209</b>	<b>4.842</b>	<b>5.235</b>	<b>240.899</b>	<b>1.619</b>	<b>41.150</b>	<b>73</b>	<b>5.128</b>

\*All animals >12 months of age tested negative on the last test for brucellosis.  
N.E.: Number of Establishments, N.A.: Number of Animals

Health status regarding Bovine Brucellosis in animals. Greece 2020



Map III, Distribution of bovine animals, by health status for Brucellosis.

(d) Timeline with prevalence, incidence data and, where relevant, vaccination history covering at least the past 5 years:

Infection of establishments with <i>Brucella</i> spp.				
Year	Prevalence	Incidence	Vaccination with RB-51 (establishments)	Vaccination with Rev-1 (establishments)
2016	3,57%	1,27%	156	-
2017	2,18%	0,62%	107	1.372
2018	2,97%	1,59%	185	2.476
2019	2,60%	1,16%	122	1.803
2020	1,44%	0,95%	122	2.341

**7. A description of the disease control strategy of the eradication programme in accordance with Article 16 of Commission Delegated Regulation (EU) 2020/689 including at least:**

**(a) the sampling schemes and diagnostic methods to be used in accordance with Annex IV to Delegated Regulation (EU) 2020/689:**

**(i) for the granting of the disease-free status to establishments and the maintenance of that status:**

The Regional Competent Veterinary Authorities creates the official records about the classification according to the health status of herds of all kept bovine animals under the eradication program and keeps them up-to-date.

Registration and identification of kept bovine animals is compulsory.

An awareness campaign to farmers and other stakeholders is also implemented (e.g. leaflets, posters etc).

### **Classification of CATTLE HERDS & ESTABLISHMENTS**

- a) Type **B1** herds of cattle: Establishments of unknown previous clinical and vaccination history and serological status.
- b) Type **B2** herd of cattle: Establishments of known previous clinical and vaccination history and serological status, which are routinely serologically tested.
- c) Type **B3** herd of cattle: Establishments fulfilling the conditions for disease-free status (including vaccinated animals)
- g) Type **B4** Herd of cattle: Establishments fulfilling the conditions for official disease-free status (consisting of non-vaccinated animals)
- In selected Regional Units, vaccination with the RB-51 vaccine is approved on dairy cattle farms. Blood sampling of the animals is carried out immediately before their vaccination. All female bovines over the age of 4 months are vaccinated, once in their life. All bovines over the age of 12 months are blood tested **twice** in a period of 12 months, as the RB-51 vaccine has the advantage not to confuse the vaccine with the wild strain in the framework of the laboratory testing..
- In some mountainous areas, where livestock farming relies on extensive grazing, small ruminants (sheep and goats) and fattening bovines commingle on community pastures. In such areas, vaccinations with Rev-1 vaccine are implemented in those bovine animals, to protect them from the increased risk of being infected by *Br. Melitensis*. (Dec. 2002/598 (EC)).

• The official diagnostic methods for granting and maintaining infection-Free status from Brucella are described in Section 1 of Annex III to Regulation (EC) No. 2020/689/EU. The diagnostic methods used are as follows:

#### 1.1. Serological tests

##### (a) tests for blood samples

- (i) buffered Brucella antigen tests;
- (ii) complement fixation test (CFT)
- (iii) indirect enzyme-linked immunosorbent assay (I-ELISA)

##### (b) tests for milk samples

- (i) I-ELISA

An efficient means of screening dairy herds is by testing milk from the bulk tank. Milk from this source can be obtained more frequently than blood samples. When a positive test result is obtained, all cows contributing milk should be blood tested.

**As of the status free from infection with *Brucella abortus*, *B. melitensis* and *B. suis* with vaccination, this may be granted to an establishment keeping bovine animals, if the following conditions are met:**

1. (a) during the past 12 months, there has been no confirmed case of infection in kept bovine animals of the establishment;

(b) the entire bovine animals over 12 months of age present in the establishment at the time of sampling have tested negative to serological test, on two occasions as follows:

(i) the first test must be carried out on samples taken not earlier than 3 months after the removal of the last confirmed case;

(ii) the second test must be carried out on samples taken not earlier than 6 months and not later than 12 months following the date of sampling referred to in point (i);

(c) animals showing clinical signs, such as abortions, have been subjected to investigations with negative results;

(d) since the beginning of the sampling, as above mentioned, all bovine animals introduced into the establishment originate from establishments free from infection with *Brucella* without vaccination or free from infection with *Brucella* with vaccination and:

(i) originate from a Member State or a zone free from infection with *Brucella* for the relevant animal population;

(ii) are entire bovine animals over 12 months of age and have tested negative in a serological test on a sample taken

— during the 30 days prior to their introduction into the establishment; or

— during the 30 days following their introduction into the establishment provided they have been kept isolated during this period; or

(iii) are post-parturient females kept in isolation since their introduction into the establishment until they have tested negative in a serological test carried out on a sample taken not earlier than 30 days after parturition; and

(e) since the beginning of the sampling as above mentioned, all germinal products of bovine origin introduced into or used in the establishment originate from:

(i) establishments free from infection with *Brucella* without vaccination or free from infection with *Brucella* with vaccination; or

(ii) approved germinal product establishments.

2. By way of derogation from point 1, the status free from infection with *Brucella* with vaccination may be granted to an establishment if all bovine animals originate from establishments free from infection with *Brucella* without vaccination, or free with vaccination, and:

(a) originate from a Member State or a zone free from infection with *Brucella abortus*, *B. melitensis* and *B. suis* for the relevant animal population;

(b) are entire bovine animals over 12 months of age and have tested negative in a serological test carried out on a sample taken:

(i) during the 30 days prior to their introduction into the establishment; or

(ii) during the 30 days following their introduction into the establishment provided they have been kept isolated during this period; or

(c) are post-parturient females kept in isolation since their introduction into the establishment until they have tested negative in a serological test carried out on a sample taken not earlier than 30 days after parturition.

**As of the maintenance of the status free from infection with *Brucella abortus*, *B. melitensis* and *B. suis* with vaccination:**

The status free from infection with *Brucella* with vaccination of an establishment keeping bovine animals may be maintained if only the provisions of Section 2, of Chapter 2, of Part I, of Annex IV of Commission Delegated Regulation (EU) 2020/689 are met:

1) all kept bovine animals must have been free from clinical symptoms of brucellosis for the last 12 months,

2) all kept bovine animals over 12 months of age must have tested negative by serological testing, twice within a period of 3-12 months, according to the type of production, the situation of the disease and the identified risk factors,

3) bovines that are introduced in the holding must come from a holding of the same health status, while animals over 12 months of age must have tested negative 30 days before or 30 days after their entrance in the holding. In the second case, the animal/s must be quarantined until the negative result/s,

4) animals under 12 months of age that are introduced to the holding must originate from an (officially) disease-free holding, while animals over 12 month of age must come from an (officially) disease-free holding and must have tested negative 30 days before or 30 days after their entrance in the holding. In the second case, the animal/s must be quarantined until the negative result/s.

**(b) disease control measures to be applied in the event of a confirmed case:**

1) Exit from and entrance in the affected premises of animals is prohibited, with the exception of those destined for slaughter; in this case, the procedure is verified with the appropriate documents (according to national legislation) for movement to the slaughterhouse, under “slaughter in the framework of eradication programme”, issued by the Local Veterinary Authorities and accompanying animals throughout transport.

2) Infected animals are marked with a "Δ" shape punch in the right ear and must be in isolation from the other animals until their slaughter. A special, clearly separated space is defined for the isolation of positive and suspect animals.

3) An epidemiological investigation must be conducted so that, the source of infection is determined and the herds linked with the outbreak are also determined and investigated. The results are registered in the “Epidemiological Investigation Fact sheet”.

4) Regarding other animals of species susceptible to *Brucella*, if present (eg. sheep and goats, swines, dogs, horses), measures are applied as foreseen. More specifically, in case of suspicion in the targeted animal population, other animals of species susceptible are tested and, if positive, they are culled or euthanized.

5) All establishments in a radius of 500m from the infected one are checked in high priority if they have not been already checked in the framework of the programme.

6) In case the positive holding utilizes a pasture, no other holding is permitted to move to it until 60 days have passed since the departure of the positive holding from that pasture.

7) In case that more than one cattle holding coexists in the same livestock establishment, then the Veterinary Services of the Regional Unit can recommend the slaughter of the whole cattle population of the establishment if the percentage of cattle that tested positive exceeds 50% of the animals of all establishments, or if the prevalence of the disease is 0,1-1% among the establishments of the specific area, or if the infected animal or holding is placed in an officially free area.

**(c) biosecurity and risk mitigating measures to be implemented:**

1) Aborted fetuses, placenta, animals that were stillborn die right after their birth are collected and destroyed according to the current legislation concerning the management of animal by-products. Appropriate parts are collected as samples (with every necessary precautions and biosafety) and sent to

the National Reference Laboratory (NRL) for Brucellosis (in Larisa) so that the necessary laboratory tests for the confirmation of the disease can be performed.

2) Milk coming from infected animals is prohibited to be used for human consumption. It must be collected in separate vessels and used only under the procedures of Reg.1069/2009.

3) Milk coming from non infected animals of the holding may be used according to Reg. 853/2004 of the EU, annex III part IX ch. 1 par. 3-5.

4) Manure and bedding must be collected in a separate place and treated with relevant disinfectant, unless covered with soil. It is forbidden to transport these materials outside the holding. In case these are moved, they are characterized as category 2 materials and are treated according to the procedures of Reg. 1069/2009.

5) Materials that have come to contact with abortion or delivery fluids, placentas or foetuses are disinfected by coiling or spraying or dipping in the suitable disinfectant.

6) The slaughter of infected animals must be carried out according to Reg. 2019/627, Section 4, Article 34, at most within 30 days from the announcement of the positive test results. Transport of these animals to the slaughterhouse is carried out according to the regulations in place and after all necessary measures to contain the infection have been taken.

7) The carcasses of infected animals undergo a detailed inspection. Samples are also taken and sent to the corresponding Veterinary Lab.

8) In 1-7 days after the last infected animal has been culled, cleaning and disinfection of the premises and the equipment of the holding takes place. At most, in 15 days after the culling of the last positive animal and at least 7 days after the completion of the first disinfection, the procedure is repeated. Cleaning and disinfection are carried out under veterinary supervision and a certificate of disinfection is signed in two copies. One copy is given to the farmer and the other is kept in the archive of the Local Veterinary Authority.

9) The importance of the zoonoses, for the public health is pointed out to the farmer. Compliance to the aforementioned measures is a prerequisite for the successful implementation of the programme and the compensation of the farmers, who should be notified in writing about the obligation of biosecurity measures in their establishment.

**(d) type of vaccine(s) to be used and the vaccination scheme, if relevant:**

- Rev-1 Vaccination Programme in fattening bovines of extensive grazing which commingle with small ruminants (sheep and goats) on community pastures in specific mountainous areas of the country was decided, because *Br. melitensis* strains were isolated from aborted foetuses of these animals.

Moreover, according to the programme, vaccination of all healthy female and non-pregnant calves over four (4) months of age is mandatory.

Pregnant or sick females who were not vaccinated are recorded and vaccinated as soon as possible after births or at the next visit of the veterinarian occupied within the Programme. Male cattle are not vaccinated and those over 12 months are annually tested for Brucellosis.

- As an additional preventive measure in order to rapidly reduce the prevalence of Bovine Brucellosis is the vaccination programme using the RB-51 vaccine.

RB-51 Vaccination programme is implemented in the Regional Units (RUs) of Thessaloniki, Pella, Larisa and Pieria. The vaccination with RB-51 vaccine is implemented concurrently with the eradication programme. According to the programme all healthy, non-pregnant female cattle over 4 months of age are vaccinated. Prior to vaccination, a blood sample is collected and tested for *Br. abortus* from all the animals over 12 months of age. Vaccination is implemented once in the lifetime of each animal. Vaccination is mandatory for all unvaccinated female animals over 4 months of age in infected establishments.



**(e) measures to be implemented as regards additional animal populations, if relevant:**

Regarding other animals of species susceptible to *Brucella*, if present (eg sheep and goats, swines, dogs, horses), measures are applied as foreseen. More specifically, in case of suspicion in the targeted animal population, other animals of species susceptible are tested and, if positive, they are culled or euthanized.

**(h) targeted animal populations and when applicable, additional animal populations:**

The targeted animal population for the programme is all kept bovine animals. As bovine animals are considered all animals of the species: *Bos taurus* and *Bos indicus*, *Bison bison* and *Bubalus bubalus*.

The brucellosis programme is also implemented to dairy Equidae over 12 months of age. All equines over 12 months must be blood-tested two times in a period of 3-12 months. Treatment or vaccination for equine brucellosis is prohibited.

**8. A description of the organization, supervision and roles of the parties involved in the eradication programme including at least:**

- (a) the authorities in charge of coordinating and supervising the implementation of the programme;
- (b) responsibilities of all stakeholders involved.

A) The Directorate of Animal Health of the Directorate General of Veterinary Services, of the Ministry of Rural Development & Food is responsible for the preparation and publication of the Joint Ministerial Decision under which the costs for the implementation of the programme are covered.

B) The Department of Zoonoses (DZ) of the Directorate of Animal Health, of the Directorate General of Veterinary Services of the Ministry of Rural Development & Food is responsible for:

- (i) programme planning,
- (ii) the designation of the implementing authorities and the definition of the responsibilities for the needs of the programme,
- (iii) centralized control and coordination of the authorities involved in the implementation of the programme,
- (iv) the provision of appropriate instructions and clarifications to the implementing authorities of the programme,
- (v) collecting and evaluating data and results of the programme, reporting and informing the competent authorities of the Commission,
- vi) the timely supply of vaccines (RB-51 and Rev-1) and diagnostic reagents in collaboration with the Directorate General of Financial Services of the Ministry of Rural Development & Food.

C) The Directorate of Animal Protection & Veterinary Drugs of the Directorate General of Veterinary Services, of the Ministry of Rural Development & Food is responsible for the implementation of the system of identification and registration of the establishments according to the Regulations (EC) 1760/2000 and 911/2004.

D) The Regional Veterinary Directorates are responsible for:

- (i) the coordination, monitoring and evaluation of the implementation of the programme in their region,
- (ii) the cooperation with the respective Veterinary Departments of their region and with the Department of Zoonoses of the Directorate of Animal Health,
- (iii) the monthly / bimonthly collection of the Monthly / Bimonthly Surveillance Fact sheets of the Bovine Brucellosis programme from their region and sending to the Department of Zoonoses of the

Directorate of Animal Health. They are also responsible for collecting and processing of any statistical and financial data from the Regional Unit of their province, on a semi-annual and annual basis.

iv) support for the implementation of the programme to the Veterinary Services of the Regional Units of the country.

E) The Veterinary Services of the Regional Units are responsible:

(i) to designate a veterinarian responsible for the implementation of the programme,

(ii) to carry out official activities (checking the marking of animals, the special marking of infected animals, checking the records of establishments, all the necessary bulletins relating to the implementation of the programme and the issuance of Health Certificates, for the movement of animals or herds) and to cooperate with creoscopic veterinarians, for the needs of the programme, in the territory of their responsibility,

(iii) for the proper implementation (vaccinations / blood sampling / milk sampling) and the evaluation of the implementation of the programme at the level of Regional Unit and in cooperation with the approved private veterinarians,

(iv) for the dispatch of the required doses of vaccines, their receipt, proper keeping and administration. Also, for the information of the Department of Zoonoses at the end of the year regarding the annual needs for vaccines.

(v) for the sending of the requested statistical data of the programme to the Veterinary Directorates of the Region and to the Department of Zoonoses of the Directorate of Animal Health, on a regular basis, as planned.

(vi) for the completion and timely sending of the Bimonthly Surveillance Fact sheets of their Regional Unit and the updating of the online veterinary database for vaccinations and sampling,

(vii) to monitor the implementation of the programme by approved private veterinarians,

(viii) to inform and to cooperate with all stakeholders and other competent authorities (breeders, veterinarians, livestock and agricultural cooperatives, Public Health Authorities) in the framework of the implementation of the programme,

(ix) for the disinfection and sanitation procedures and the restoration of the health status of livestock of infected establishments,

(x) for compiling of a list by health status for brucellosis of all establishments of the area of their responsibility and the notification of the Veterinary Directorate of their Region and the DEPARTMENT OF ZOONOSES , on a six-month and annual basis.

F) The NRL of Brucellosis in Larissa (Directorate of the Veterinary Center of Thessaloniki - Department of Veterinary Laboratory of Larissa) is responsible for:

(i) the approval of the results of the evaluation of the tests used for the diagnoses of brucellosis,

(ii) the calibration of the secondary national reference serum standards (working standards) based on the primary national serum standard,

(iii) the organization of the annual comparative tests (ring tests) among official national laboratories,

(iv) the cooperation within the EU network with the national reference laboratories for brucellosis,

(v) the provision of scientific and technical assistance to the Directorate of Animal Health for the implementation of coordinated control plans,

(vi) the collection of the semi-annual and annual results and statistical data from the laboratories participating in the program and their submission to the Directorate of Animal Health,

(vii) the coordination and the standardization of the diagnostic methods applied in the diagnostic laboratories for brucellosis of the country. For that reason:

1) plans the supply of diagnostic reagents,

2) checks the quality of all reagents,

3) performs bacteriological examinations,

4) receives blood samples / milk samples and pathological material for bacteriological examinations from the Veterinary Departments of the Regional Units.

(viii) the testing of blood samples from other species of animals susceptible to brucellosis (eg. dogs, equidae),

(ix) the performing of the serological tests of the blood samples / milk samples received from the Veterinary Departments of the Regional Units of their area.

G) The Department of Pathological Anatomy, Histology, Microbiology and Physiopathology of the Breast, the Department of Veterinary Laboratory of Iraklio, the Department of Veterinary Laboratory of Rhodes and the Department of Veterinary Laboratory of Tripoli of Veterinary Institutes in Athens. The Department of Avian and Bee Pathology, Microbiology, Infectious Diseases and Brucellosis, the Department of Veterinary Laboratory of Ioannina and the Department of Veterinary Laboratory of Kavala of the Veterinary Institute of Thessaloniki are responsible for:

(i) the laboratory examination and the performance of the serological tests of blood samples / milk samples from the Veterinary Departments of the Regional Units of their area in accordance to the instructions of the National Reference Laboratory (NRL). In case of a positive result, they inform the competent veterinary authorities within 24-48 hours.

(ii) the sending of statistical and financial data to the NRL and the Department of Zoonoses,

(iii) participation in the annual ring test organized by NRL.

H) The Laboratory of Microbiology and Infectious Diseases of the Aristotle University of Thessaloniki, as well as the Clinic of Veterinary Pathology of the Veterinary School of AUTH is responsible for the scientific support of the programme.

I) Private veterinarians working in livestock / agricultural cooperatives, dairy and cheesemaking industries may participate, under certain conditions, in the implementation of the programme, after an approval from the Department of Veterinary Medicine of the relevant Regional Unit. Then, the private veterinarian must submit a weekly program with the names of the breeders and the codes of their establishments, the number of animals that will be vaccinated / blood samples / and / or the milk samples that will be collected, the exact dates of the above veterinary operations, as well as the exact number of vaccine doses required for each holding.

J) Private veterinarians may be involved in the vaccination programme of fattening bovines of extensive grazing against *Br. melitensis* in specific mountainous areas of the country, as well as in the equine brucellosis eradication programme.

K) The farmers of bovine establishments are responsible for:

1) the registration of their bovine establishments and their livestock in the Identification and Registration System

2) the cooperation with the competent Veterinary Authorities of the Regional Units for the implementation of the programme. In particular, they shall inform the Veterinary Services of the Regional Unit, in no later than 48 hours, in case of abortions,

3) the cooperation with the Veterinary Authorities and the participation to assist vaccinations, blood / milk sampling, epizootiological research,

4) the compliance with the restrictive, sanitary and biosecurity measures imposed on their establishment,

5) keeping up-to-date the establishment's registries,

6) keep all the necessary documents and the results of laboratory tests in the establishments' registries,

7) maintain biosecurity measures on their establishments in order to prevent contamination of the animals.

L) A Central Committee is set up for the coordination, control and effective implementation of the programme, which acts as an advisory body to the Department of Zoonoses.

The Central Brucellosis Committee (CBC) consists of:

- 1) the Head of the Directorate of Animal Health as the President,
- 2) the Head of the Department of Zoonoses Animal of the Directorate of Animal Health as the Vice President,
- 3) the responsible veterinarian of the programme from the Department of Zoonoses.
- 4) a veterinarian responsible for the Integrated Information System of the Directorate of Animal Protection, Medicines and Veterinary Applications of the Ministry of Rural Development & Food,
- 5) the veterinarian in charge of the National Reference Laboratory.
- 6) a veterinarian specializing in Veterinary Pathology,
- 7) a veterinarian specializing in Veterinary Microbiology,
- 8) a veterinarian - epidemiologist.

**9. Estimated duration of the eradication programme:**

The estimated duration of the eradication programme is ten (10) years (+/- two (2) years).