

Department of Livestock Development THAILAND

Protocol Sample Submission to Laboratory for Brucellosis

via

International Airline

Prepared by: Brucellosis Laboratory

National Institute of Animal Health

Revised August 2016

The Laboratory for Brucellosis, National Institute of Animal Health, Department of Livestock Development, Ministry of Agriculture and Cooperatives is responsible for Brucellosis diagnosis and operating as the Laboratory for Brucellosis training center in country and upon the requested from neighboring countries. The specimens or infectious substances submission to the laboratory for diagnostic purpose are recommended to send to NIAH- Laboratory, Thailand by AIRFREIGHT ONLY. The destination airport is to SUVARNABHUMI INTERNATIONAL AIRPORT. The customs clearance process will be proceed by the DLD officers and collected by the authorized staffs from NIAH, Bangkok. In order to achieve the bio-safety and bio-security standard of samples or biological materials from oversea countries to NIAH-Brucellosis Laboratory, the packing and dispatch of samples or biological materials are required as this following.

Definition

Infectious substance definition: Infectious substance is known to contain, or reasonably expected to contain, pathogens. Pathogens are defined as microorganisms (including bacteria, viruses, rickettsia, parasite, fungi) and other agents such as recombinant (hybrid or mutant) that are known or reasonably believed to cause disease in humans or animals.

Non infection substance definition: Non infection substance is substances which do not contain infectious pathogens or have been treated, neutralized, inactivated so that they do not longer pose a health risk.

1. Collection of specimens

1.1 Tissues/milk/swab/others specimens

Specimens are collected using aseptic techniques and placed in sterile plastic bags of appropriate size to limit the amount of contained air which occupies unnecessary space and impedes refrigeration. Specimens should be refrigerated immediately after they are collected and frozen if they cannot be cultured within 24 hours. For transport, stout polystyrene foam containers, in which the specimens are packed on dry ice, are ideal. The number of variable *Brucella* cells in tissues stored frozen at -20 ° C or below remains constant for at least 18 months. (This applies to unprocessed tissues, not to tissues macerated and suspended in diluent and then frozen.)

Milk

Since infection of the udder with *Brucella* may be confined to one or more quarters, it is essential that the 10-20 mL of milk is obtained from all four quarters separately. The whole udder is washed and dried, the tip of each teat is disinfected with a swab of 70% ethanol and wiped dry. The first one or two streams of milk are discarded and the sample from each teat is milked directly into a sterile container. It is essential to avoid contact between the stream of milk and the milker's hand, both to protect the milker and to prevent cross contamination of specimens. The hands should be cashed in mild disinfectant between animals.

Vaginal swabs

Brucella organisms may often be recovered from a vaginal swab taken in the 6 week period following parturition or abortion. A guarded sterile swab is used and the swab should then be broken off in a sterile container if it is to be cultured immediately or into a vial of transport medium if there is to be a delay in before the swab is cultured.

Fetal membranes

In abortion due to *Brucella* infection, parts of the fetal membranes usually contain enormous numbers of *Brucella* organisms: this is also often true of membranes from infected full-term births. Special care is needed in handling, packing and transporting specimens. The membranes should be examined carefully and the least healthy-looking cotyledons should be removed from and transferred to the laboratory for culture. Infected cotyledons lose their normal bright red appearance and become a dirty grayish-yellow color.

Aborted fetus

The most valuable materials from the aborted fetus for cultural examination are the stomach contents, lung and spleen. The stomach contents, 10-20 mL, may be withdrawn with a pasteur pipette or sterile syringe. Pieces of lung and spleen are removed with aseptic precautions to sterile containers for transport to the laboratory.



Animal carcasses

The tissues from which *Brucella* can most often be isolated are those of the reticulo-endothelial system, the pregnant or early post-parturient uterus, the udder and its secretions, or the male reproductive tract.

In mature cows, about 90% of infections can be detected by culture of the mammary lymph nodes; the infection of samples from mandibular, medial iliac lymph nodes and the uterine caruncles, if present, will increase the chance of successful culture to almost 100%. In heifers, additional tissues need to be cultured to obtain meaningful results, these are medial retropharyngeal, parotid, superficial cervical (prescapular), mesenteric lymph nodes and spleen. In male animals, the tissue listed above should be cultured, with the following in place of the female reproductive organs: samples of testes, prostate, epididymes and seminal vesicles together with their associated lymph nodes.

Specimens for bacteriological examination are collected aseptically. Each tissue should be packed separately, identified and cooled or frozen immediately. A separate set of sterile forceps and scissors is for each tissue, or at least for those from each animal. It is sometimes practicable to flame the tissue overlying and surrounding the lymph nodes or tissues to be collected. The specimen is dissected out from the surrounding fat without rupture of the capsule if possible.

1.2 Blood /serum samples

Collection of blood sample should be collected under sterile conditions and serum part is separated by centrifugation and kept it in a screw cap vials with Orings. Serum samples should be kept at freezing condition (-20°C) before dispatch to Brucellosis Laboratory.



2. Packaging and specification

Infectious substance must be transported in packaging which meets the UN Specifications for Division 6.2 that meet the requirement of Packing Instruction 620 (PI 620) of IATA Dangerous Goods

Noninfectious substance or diagnostic specimens must be transported in packaging that meets the requirements of Packing Instruction 650 (PI 650)

The samples must be packed in a watertight primary and secondary IATA approved containers or a strong container so that the samples arrive in good condition and do not present any hazard to persons or animals during shipment. It is essential that the contents of containers which break or leak during the transit, do not contaminate the outside layer of the parcel.

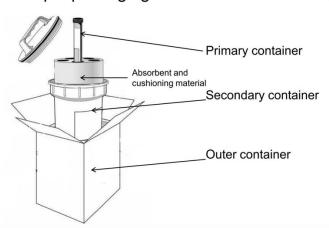
The procedure for packing of samples are recommended as this following

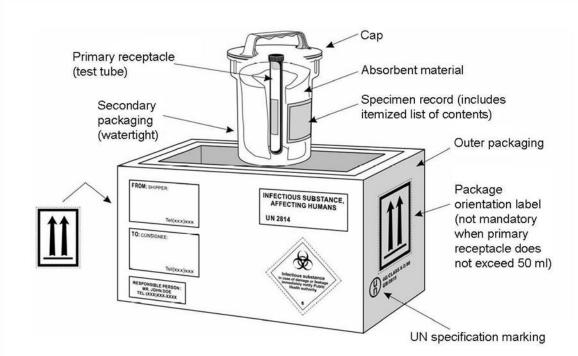
- 1. The samples must be put a primary container (glass or plastic tubes or bottles) with screw caps and wrapped with paraffin film or adhesive tape individually in order to prevent leakage of fluid. The wrapping of bottle or primary container should be completed in clean surrounding by using effective disinfectant before proceeding further.
- 2. Primary container must be packed in watertight secondary packaging, which should be strong crushproof and leak-proof metal containers with absorbent cotton wool that can absorb entire contents of all primary containers.
- 3. The secondary packaging must be placed in outer containers which should be a foam box covered with hard box or IATA proved container.

- 4. Sufficient information and list of samples or materials should be enclosed in an envelope covered with plastic bag and placed between the secondary packaging and outer box.
- 5. The FREEZER PACK is recommended to put outside the secondary packaging to ensure that all materials are kept cool during shipment. The prefrozen packs at -20oC is necessary to be prepared before packing samples.
- 6. In case of packing with dry ice, it must be put outside the secondary packing. Please do not place dry ice directly in the primary containers, it may break the sample tube.

Category A – UN 2814/UN 2900 Packing Instructions 620 (PI 620)

Triple packaging – 3 levels of containment





PI620 (the smallest external dimension shall not be less than 10 cm)
PI650 (at least one surface of the outer packaging must be 10x10 cm minimum)

3. Marking Labeling

The outer package of parcel must be clear labeled with all details as this following:

- The name and address of Institute,
- Telephone numbers
- Flight number, arrival time
- Airway bill number (usually filled out by the transport company)
- Dangerous goods declaration (completed and signed by the shipper (usually transport companies pre-fill the form for the shipper with several original copies)
- Import/export permit (the shipper and the receiver should check on their respective country's requirements)
- List of contents
- Invoice (for customs)
- Dry ice label (if necessary)
- Others such as Perishable, fragile, keep cool or keep at -20 ° C, etc.





3.1 Name and address:

Laboratory Epidemiology Section c/o Brucellosis Laboratory, Immuno-serology Section National Institute of Animal Health, Department of Livestock Development 50/2 Pahonyothin, Kasetklang, Ladyao Chatuchak, Bangkok 10900 THAILAND Tel: +66 (0)2 579 8908 to 14, Fax: +66 (0)2 579 8918 to 9 Email: niah@dld.go.th,

preecha.w@dld.go.th

3.2 Others marking:

The marking on the package is an essential source of information to communicate to everyone involved in the transportation process, such as

"DIAGNOTIC SPECIMENS" or "BIOLOGICAL SUBSTANCE"

"PATHOLOGICAL MATERIAL OF NO COMMERCIAL VALUE"

"PERISHABLE" "FRAGILE" "KEEP AT 4 ° C"

3.3 Hazard Labels:

The label must be legible and must not be covered. The label must be attached to one side of the box and not overlap.

Infectious Substance label



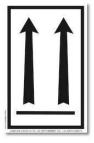
Cargo aircraft only

Must be used for all shipment that cannot ride on passenger aircrafts



3.4 Oriental label

The arrow label must be used when shipment liquid samples in the container, indicates to hold the package this way up, this label must be placed on 2 sides of the package (opposite side)



4. Route and destination of the shipment

All biological materials, Diagnostic specimens or Infectious substances must be sent by AIRFREIGHT ONLY direct to SUVARNABHUMI INTERNATIONAL AIRPORT, BANGKOK ONLY before dispatch the sender must notify the National Institute of Animal Health, fax (Fax number +662 579 8918 to 19) for the details of the Airway Bill Number, flight number, time and date of arrival of the parcel or package in Bangkok, Thailand.

5. Customs clearance process

The staffs of Department of Livestock Development (DLD)/courier (authorized by DLD) will clear the parcel through the customs at the airport, then the parcel will be collected and taken to the laboratory at NIAH by the courier.

6. References

- OIE. 2013. Manual of Diagnostic Tests and Vaccines for Terrestrial Animals 2013. Chapter 1.1.1. Collection, submission and storage of diagnostic specimens (NB: Version adopted in May 2013)
- OIE. 2013. Manual of Diagnostic Tests and Vaccines for Terrestrial Animals 2013 Chapter 1.1.2. Transport of specimens of animal origin (NB: Version adopted in May 2013)
- Regional Reference Laboratory for FMD in South East Asia, National Institute of Animal Health, Department of Livestock Development, Pakchong, Nakhonratchasima 30130, THAILAND //www.dld.go.th/rrl
- The 4th FAO-APHCA/OIE/DLD Regional Workshop on Brucellosis Diagnosis and Control in Asia and Pacific Region Proficiency Test and Ways Forward for the Region Chiang Mai, Thailand, 19-21 March, 2014
- World Health Organization 2006. Brucellosis in human and animals. Produced by the World Health Organization in collaboration with the Food and Agriculture Organization of the United Nations and World Organization for Animal Health. Editors: M.J. Corbel, S.S. Elberg, and O. Cosivi. 89 pages.

Example: Packaging the diagnostic samples for shipment via courier





















BRUCELLOSIS SAMPLE SUBMISSION FORM

Sender details	Owner Details
Country:	Owner's name:
Your Reference No.:	Address:
Name of Institute:	
Address:	
	District/Province:
	Region:
Tel:Fax:	Country:
Email:	Tel:Fax:
1.Sample Details:	
Species: Number of animal samples:	
☐ Serum ☐ Milk ☐ Tissues ☐ Bacterial colony ☐ DNA	
□ other	
Animal sample identification: Attachment	
Date collected:/ Date of dispatched:/	
History:	
Thistory.	
2. Vaccination history:	
2. Outhwell investigation.	
3. Outbreak investigation:	
4. Comments:	
4. Comments:	
5. Signature	Date
5. Signiture	Dute