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Swiss Expert Committee for Biosafety SECB

SECB Recommendation

for work at sites potentially contaminated with anthrax spores

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1 Purpose and field of application

This SECB Recommendation is aimed at construction firms, companies dealing with contaminated sites, analytical laboratories and other companies working at sites that may be contaminated with anthrax spores, and also at the cantonal enforcement agencies responsible for such fields of activity. Possibly contaminated sites include former tanneries or knacker's yards (dumps for animal carcasses). The purpose of this Recommendation is to ensure a uniform procedure at such sites throughout Switzerland. The Recommendation provides information on investigations that must be made and on protective measures for employees. It also describes when soil samples should be taken, how this should be done, and where particular attention should be paid.

2 Anthrax: the basics

2.1 The pathogen: occurrence and distribution

Anthrax is an acute infectious disease cause by the spore-forming bacterium *Bacillus anthracis*. It primarily affects warm-blooded animals, especially ruminants. Anthrax is found worldwide but occurs predominantly in agricultural regions with livestock. Anthrax currently occurs primarily in Africa, Asia, South and Central America, Southern and Eastern Europe, the Caribbean, and the Middle East. In Switzerland, the last reported infection in a human was in 1991. Anthrax spores are extremely resistant and are able to survive for decades in the soil or in water, and then reproduce when environmental conditions become favourable or if transmitted to a new (animal or human) host.

2.2 Transmission

Animals become infected by ingesting *Bacillus anthracis* via feed, water and soil material. Transmission to humans takes place via contact with infected animals or contaminated animal products. Direct transmission from human to human is considered unlikely; human infection through contact with contaminated soil is extremely rare.

Species	Susceptibility
Sheep, goat, cow, buffalo, horse, camel, reindeer, ele- phant, mink	high
Dog, cat, rat, humans	moderate
Pig	low
Birds (exception: ostrich)	almost resistant

Susceptibility to infection varies from one species to another.

2.3 Forms of the disease in humans and treatment possibilities

Two properties of the anthrax pathogen cause disease. First, the bacteria form toxins that cause cell death and oedema; and second, bacteria may form a capsule and thus shield themselves from the host's immune system.

The incubation period is 1–6 days. Depending on the symptoms, anthrax may be treated more or less successfully with antibiotics. Since the fatal effect of anthrax is due to toxins, the production of which increases as the disease progresses, it is important to start treatment early. Although a vaccine exists, it is not officially approved in Europe.

Three forms of the disease occur in humans, distinguished by the method of transmission.

Cutaneous anthrax: infection takes place via contact with contaminated animals and animal products such as skins, wool or bone(meal), through cuts or other skin lesions through which the pathogen can penetrate. Blisters and pustules form at the entry points, and may develop into ulcers. Without treatment, lethality is 20%, with treatment it is less than 1%.

Gastrointestinal anthrax: infection takes place via contaminated meat and presents as an acute inflammation of the gastrointestinal tract, with symptoms such as nausea, fever, vomiting blood, diarrhoea. Without treatment lethality is 20–60%.

Pulmonary anthrax: infection takes place via the inhalation of fine dust containing spores or pathogens, or infected droplets. After initially nonspecific symptoms, a high fever develops, with chest pain and breathing difficulties, leading to pulmonary and cardiovascular failure, which untreated will cause death in 90% of cases.

The lethality of cutaneous and gastrointestinal anthrax remains high even after administration of antibiotics.

3 Historical investigation

Before construction activities can be carried out at former knacker's yards or tanneries (identified from the register of polluted sites) where contamination with anthrax spores cannot be ruled out, a range of investigations must be carried out.

If bone material, very organically contaminated soil (dark colouring), or foul-smelling zones are found during construction at sites that are not suspect, the work must nevertheless be stopped and the investigations given below carried out.

3.1 Records giving indications of anthrax cases

A historical investigation should include research at Federal Offices, hospitals, cantonal veterinarians or insurance companies into documents that give indications of the occurrence of anthrax in humans or animals in the region.

3.2 Former knacker's yards and tanneries

3.2.1 Time scale

The time period when a tannery or knacker's yard was present must be clarified. If the company has not been related to cases of anthrax and has been out of operation for at least 20 years, it can be assumed that there is no risk. The preliminary investigation is considered to be negative (see 3.5).

3.2.2 Tanneries

The following additional points should be observed for tanneries:

- Origin of the skins: If only fresh domestic hides have been processed, the risk of anthrax spores is small. However, the hazard potential rises if they originated in Africa, Asia or South America. There remains a residual risk even if no cases of anthrax have been documented, as spores may still be present.
- Production period: During industrialisation, many sites used imported goods, sometimes cheap goods, particularly in large-scale enterprises. The risk of spores being present is therefore greater at such tanneries.
- Size of company: Small companies generally process rawhides from local sources, and not imports. This substantially reduces the risk.

3.3 **Positive preliminary investigation**

If the preliminary investigations are positive, the site should be considered to be potentially contaminated. The provisions of the Ordinance on Protection of Employees from Dangerous Microorganisms (PEMO) must be observed. This Ordinance contains a requirement for hazard identification and risk assessment. Soil sampling, as described in Chapter 4, may be part of this hazard identification and risk assessment. The protective measures for the construction phase are in line with this hazard identification and risk assessment.

Measures to protect the surroundings should also be established.

3.4 Unclear preliminary investigation

If some points are unclear or there are other reasons to suspect the presence of anthrax, prophylactic soil sampling (see Chapter 4) or an investigation of the bones found (see Chapter 5) should be considered.

If prophylactic soil sampling is waived, the minimum protective measures against anthrax infections must be observed for the construction work (see Chapter 6.1).

3.5 Negative preliminary investigation

If there are no records of a tannery or knacker's yard, or of cases of anthrax, the construction phase may be commenced or continued; the generally applicable protective measures (i.e. not including anthrax-specific protective measures) must continue to be observed.

4 Exploratory investigation

Once it has been determined that a site is potentially polluted, samples must be taken before commencing construction. In some cases, this may be appropriate as a general precautionary measure, for example if there are indications of severe organic pollution in the soil. The objective of the exploratory investigation is to estimate how large the potentially contaminated area is.

In general, specialists (in contaminated sites, geologists, soil scientists) should determine where samples are taken.

The following factors should be considered when taking samples:

- 1. Spores favour soils that are rich in calcium and nitrates, or with a mildly alkaline pH. The top layer of soil will show hardly any spores, as they are generally destroyed by the effect of UV radiation as well as by natural soil organisms.
- The wash-out ability of bacterial spores in the soil is considered to be limited in stable and even terrain. We can assume that any anthrax spores will be located close to organic material.
- In uneven terrain, spores can be washed out and transported to the soil surface under certain circumstances (e.g., very heavy rains, terrain displacement).

To take samples, boring should be carried out to a depth of 3 metres. The cores should be tested for the sections contaminated with organic material, and this material examined. A fresh, sterile spoon should be used for each section in order to prevent cross-contamination.

Sites where a tannery once operated are generally also contaminated with chemicals (primarily chromium and arsenic). If soil samples are to be investigated for particular pollutants in a chemical laboratory, and if anthrax is suspected, the samples should first be checked for the presence of spores This test may be omitted if the soil to be investigated has previously been autoclaved. The autoclaving program used should be validated for soil.

If the soils are disposed of as special waste, the anthrax identification can be waived, as long as minimum protective measures (Chapter 6.1) for staff are observed and the special waste is directly incinerated.

4.1 Targeted sampling

To obtain a meaningful result, it is important to take samples where anthrax spores are most likely to be found. The selection of suitable sampling places is therefore essential. On a large site it is however often very difficult to carry out a risk assessment and establish the places that have a high probability of being contaminated. Aerial photos may contain valuable information, such as where a relevant operation was located at a particular time.

If the site is a former knacker's yard and if the location of the pit for animal carcasses, which generally measured approx. 5 x 5 metres, is known, five borings should normally be enough. Spores are often found inside bones, which are therefore good indicators. Theoretically, one bone is sufficient, but it is better to examine bones from various animals.

If the site is a former tannery, the drainage and waste situation should be investigated if operating documents are available. The probability of finding spores is greatest where the hides were stored and washed. The samples would need to be taken at these locations. The best samples are taken from sediments from the drainage system, tanning pits or waste collection points. Possible landfilling with tannery waste and the former effluent disposal, e.g. trickling surfaces, should also be taken into account.

4.2 Random sampling

If the precise contaminated site is unknown, it is sensible to take bore samples every five metres.

4.3 Protective measures during sampling (occupational safety)

During the exploratory investigation, the minimum protective measures must be observed (see Chapter 6).

4.3.1 Construction workers

The awareness of the workforce about anthrax should be raised before construction commences: they must be informed about possible hazards, routes of infection and symptoms of anthrax, and the protective measures that must be taken (see Chapter 6). If injuries or symptoms occur, the workers should visit a doctor and inform him or her of the situation.

The construction machines and plant should be decontaminated after use, even if no evidence of the presence of anthrax is yet available. Formalin (10% formaldehyde solution) or other decontamination solutions can be used, and should be allowed to take effect for one hour. A breathing mask should be worn to protect against the vapours. For some equipment or parts (bucket of the excavator) a thorough heat treatment with a flamethrower may also be a good method to reduce anthrax spores. The temperature of all the material should reach at least 200 °C for a few seconds. The air filters of the driver's cab should be incinerated directly or autoclaved.

4.3.2 Laboratory staff

Staff taking the samples should be protected at least to a level equivalent to a Biosafety level 2 laboratory, in accordance with the Containment Ordinance (ContainO; SR 814.912) or the PEMO (SR 832.321); this means aprons (or better, a disposable protective suit), gloves (latex gloves on the skin, and over them robust rubber gloves for rough work), and boots should be worn. Exposed skin wounds (e.g., on the face) should be covered with an airtight dressing. If there is any danger of mud splashes, a face shield should be worn. Afterwards, soiled clothing and all equipment should be either disposed of or decontaminated. A breathing mask (FFP3) is required if dry dust samples are being collected. The risk of aerogenic infection is very small, since the necessary concentration for a pulmonary infection is not reached in the open air under natural conditions. There are no known cases in which the collection of soil or other environmental samples have led to an aerogenic infection. However, cutaneous infections have been described, although a skin lesion must be present. Gloves are indispensable to protect against skin lesions.

5 Diagnosis and evaluation

5.1 Screening

Employees may be screened for *Bacillus anthracis* at the National Reference Centre for Highly Pathogenic Bacteria (NABA).

If anthrax is found in an employee, the cantonal physician and the Regional laboratory responsible must be notified.

Initial diagnosis of *Bacillus anthracis* from environmental samples may be carried out in a BSL-2 laboratory. The test is performed both by culture detection on agar plates and by PCR detection of the toxin gene on the plasmid pX01. The samples must be prepared in different ways according to their composition. If the result of the diagnostic procedure is contradictory (e.g. PCR positive and culture negative, possibly caused by inhibition of growth), mice must be inoculated. Confirmation testing and any further characterisation of the cultures should be carried out in a BSL-3 laboratory.

The detection limit is generally <100 spores / gramme of soil, depending on the soil matrix. It does however vary, and is determined for each approach separately.

The National Reference Centre for Highly Pathogenic Bacteria (NABA) can test soil samples for anthrax spores by prior arrangement using bacterial cultures and molecular tests. Please address requests to:

National Reference Centre for Highly Pathogenic Bacteria (NABA) SPIEZ LABORATORY Bacteriology Austrasse CH-3700 Spiez Tel: 058 468 1400 e-mail: laborspiez@babs.admin.ch

5.2 Positive samples

If the result is positive and the suspicion of contaminated soil is confirmed, there are four possible courses of action, depending on the size of the site:

- 1. Concreting over the premises. The site remains on the register of polluted sites, or is entered there;
- 2. The contaminated material may remain on the site, if the surface is sealed with a lawn or other appropriate covering. Furthermore, in these cases no unmonitored earth-moving can be carried out. The site remains in the register of polluted sites, or is entered there;
- 3. Excavated material is treated thermally in a special waste facility or decontaminated using e.g. formaldehyde. Material may be transported in bulk on lorries, which must be carefully covered with tarpaulins;
- 4. Decontamination of small areas on site, for example through heat or with formaldehyde. Five litres of 10% formaldehyde per square metre are sufficient to decontaminate up to a depth of 5 cm. However, the efficacy of this also depends on the soil material.

5.3 Negative samples

If the result is negative even though preliminary investigations have been positive, the site should still be considered potentially contaminated. Construction work should be carried out under special protective measures, in line with the hazard identification and risk assessment (see 3.3). In addition, the workers should take note of any changes to exposed skin.

6 Protective measures during the construction phase (occupational safety)

Animals should not be kept in the vicinity. The soil and the earth excavated should be continually inspected visually. Excavation should not be carried out when it is dry and the wind is strong, unless the soil is kept moist to prevent the dispersal of dust.

The historical preliminary investigation and exploratory investigation produces the following decision diagram:



6.1 Minimum protective measures (soil sampling; for preliminary investigation without soil sampling)

- Expose as few members of staff as possible;
- Select equipment and working processes to reduce or avoid aerosols, dust and splashes (e.g. do not excavate if weather is dry or windy, keep the soil moist);
- Select equipment that is easy to clean;
- Changing facilities (store work clothes separately from street clothes);
- No eating, drinking or smoking on the building site; provide separate facilities if this is possible without endangering health;
- Washing facilities and hygiene plan (for hands and equipment);
- Personal protective equipment according to activity: protective suit, gloves, breathing mask (FFP3) if aerosols, dust or mist are expected, goggles if splashes are expected;
- Provide wound care, cover skin wounds (no open wounds on the building site);
- Inform the workers about the possible hazard of anthrax bacteria;
- Raise the workforce's awareness of anthrax before construction starts: they must be informed about possible hazards, infection routes and symptoms of anthrax, and the safety measures to be taken. If wounds or symptoms occur, workers should visit the doctor and inform him or her of the situation.

6.2 Protective measures if preliminary investigation is positive

These measures depend greatly on the hazard identification and risk assessment. The German publication "Handlungsanleitung zur Gefährdungsbeurteilung nach Biostoffverordnung -Tätigkeiten mit Boden sowie bei Grundwasser- und Bodensanierungsarbeiten" (Instructions for assessing risk under the German Ordinance on Biological Substances – Activities using soil or when remediating groundwater and soil, in German, BGI 583) by the Berufsgenossenschaft der Bauwirtschaft (BG Bau) is helpful. In general, the following measures should be considered in addition to the minimum requirements given above:

- Black-white separation;
- Disposable protective suit category III, Type 5 or 6;
- Extended hygiene measures: wash parts of the body and hair not covered by the disposable protective suit;
- Clean and decontaminate equipment and building machinery (see Protective measures for soil sampling);
- Disposal of potentially contaminated protective suit in accordance with the OOSB;
- Include the problem of anthrax in the emergency plan (ensure that medical advice is sought immediately if there is a suspicion of infection, and that information is provided about any possible contact with anthrax).

7 Literature

7.1 Information on polluted sites in Switzerland

- Register of polluted sites <u>https://www.bafu.admin.ch/bafu/de/home/themen/altlas-</u> ten/fachinformationen/altlastenbearbeitung/stand-der-altlastenbearbeitung-in-derschweiz/online-kataster-von-kantonen-und-bundesstellen.html (in German)
- Altlasten Kataster 2001, Zielsetzung, Grundsätze für die Erstellung, Systemgrenzen, Mengenschwellen, Kriterien und Vorgehen zur Erfassung. Publisher: Federal Office for the Environment <u>https://www.bafu.admin.ch/bafu/de/home/themen/altlasten/publikationen-studien/publikationen/erstellung-kataster-belastete-standorte.html</u>
- Milzbrand im Kanton Zürich zwischen 1878 und 2005. Schweizer Archiv für Tierheilkunde, Band 149, Heft 7 (2007), p. 295-300 <u>http://www.researchgate.net/publication/230838094 Milzbrand im Kanton Zrich zwischen 1878 und 2005</u>

7.2 Information on polluted sites in Germany

- Erkundung ehemaliger Gerbereistandorte; hrsg. vom Umweltbundesamt Deutschland, Fachgebiet III 3.6, Berlin (1998)
- Ergebnisbericht über die vertiefte Untersuchung und Bewertung von Gerbereistandorten im Hinblick auf eine Gefährdung durch Milzbranderreger in ausgewählten Teilgebieten des Landkreises Reutlingen im Rahmen der Nacherhebung altlastenverdächtiger Flächen. Authors: Dirk Maisel and Ulrich Stahl

7.3 General information on anthrax

- Federal Office of Public Health: Anthrax (in German) <u>https://www.bag.admin.ch/bag/de/home/krankheiten/krankheiten-im-ueberblick/anthrax-milzbrand.html</u>
- Federal Food Safety and Veterinary Office: Anthrax (in German) <u>https://www.blv.ad-min.ch/blv/de/home/tiere/tierseuchen/uebersicht-seuchen/alle-tierseuchen/milzbrand-beim-tier-und-beim-menschen.html</u>
- Federal Office for Civil Protection (SPIEZ LABORATORY): Factsheet Anthrax <u>https://www.spiezlab.admin.ch/content/spiezlab-internet/en/leistungen/biolo-</u> <u>gie/naba/ jcr content/contentPar/tabs copy/items/373 1574688246009/tabPar/down-</u> <u>loadlist copy 2040558320/downloadItems/459 1597655205454.download/Fact-</u> <u>sheet Anthrax en.pdf</u>
- Anthrax in Humans and Animals, fourth edition of the WHO guidelines https://www.who.int/publications/i/item/9789241547536
- Robert-Koch-Institut Deutschland: Anthrax (in German) <u>http://www.rki.de/DE/Content/InfAZ/A/Anthrax/Anthrax.html</u>
- Inactivation of *Bacillus anthracis* Spores <u>http://wwwnc.cdc.gov/eid/article/9/6/pdfs/02-0377.pdf</u>
- *Bacillus* spp., in: Mikrobiologische Diagnostik, 2. Auflage, 2009, Hrsg.: B. Neumeister, H. K. Geiss, R. W. Braun, P. Kimmig., S. 386-397, **ISBN: 978-3-13-743602-7**
- In: Qualitätsstandards in der mikrobiologisch-infektiologischen Diagnostik, MIQ 26: Hochpathogene Erreger/Biologische Kampfstoffe. Kapitel: *Bacillus anthracis* (Milzbrand), Wolfgang Beyer, Carsten Bartling, Heinrich Neubauer. ELSEVIER, MIQ 26, 2008, Teil I, S. 38-55

7.4 National and cantonal agencies

- National and cantonal agencies <u>https://www.bafu.admin.ch/bafu/en/home/topics/contaminated-sites/services.html</u>
- National Reference Centre for Highly Pathogenic Bacteria (NABA) <u>https://www.spiezlab.admin.ch/en/leistungen/biologie/naba.html</u>

- FOEN Publications on contaminated sites <u>https://www.bafu.ad-</u> <u>min.ch/bafu/en/home/documentation/publications/contaminated-sites.html</u>
- Ordinance on the Remediation of Contaminated Sites <u>https://www.fedlex.ad-min.ch/eli/cc/1998/2261_2261_2261/en</u>

7.5 Occupational safety / Health protection / Environmental protection

- Ordinance on Protection of Employees from Dangerous Microorganisms (PEMO) (in German) <u>https://www.fedlex.admin.ch/eli/cc/1999/445/de</u>
- Ordinance on the Contained Use of Organisms (ContainO) <u>https://www.fedlex.ad-min.ch/eli/cc/2012/329/en</u>
- Handlungsanleitung zur Gefährdungsbeurteilung nach Biostoffverordnung (BioStoffV) DGUV Information 201-005 <u>https://publikationen.dguv.de/regelwerk/dguv-informatio-nen/344/handlungsanleitung-zur-gefaehrdungsbeurteilung-nach-biostoffverordnung-biostoffv-taetigkeiten-mit-bo</u>