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EFSA BIOHAZ Panel (EFSA Panel on Biological Hazards), 2013. Scientific Opinion on Bioreduction application

Hald, Tine; Baggesen, Dorte Lau; EFSA Publication

Link to article, DOI:
[10.2903/j.efsa.2013.3503](https://doi.org/10.2903/j.efsa.2013.3503)

Publication date:
2014

Document Version
Publisher's PDF, also known as Version of record

[Link back to DTU Orbit](#)

Citation (APA):
EFSA Publication (2014). EFSA BIOHAZ Panel (EFSA Panel on Biological Hazards), 2013. Scientific Opinion on Bioreduction application. Parma, Italy: European Food Safety Authority. (The EFSA Journal; No. 3503, Vol. 11(12)). DOI: 10.2903/j.efsa.2013.3503

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SCIENTIFIC OPINION

Scientific Opinion on Bioreduction application¹

EFSA Panel on Biological Hazards (BIOHAZ)^{2, 3}

European Food Safety Authority (EFSA), Parma, Italy

ABSTRACT

A method for on-farm containment of animal by-products (ABPs), called a 'Bioreduction' system, was assessed. The material for containment is of ovine origin and classified as a Category (Cat.) 1 ABP material. The proposed process consists of an aerobic degradation of the ABP material in a vented, leak-proof vessel. The parameters given by the applicant for heating and aeration rate are respectively: temperature 30-42 °C and aeration under a pressure of 40-55 kPa. The resulting material is finally disposed of according to standard methods for Cat. 1 ABPs. The Bioreduction system can reduce the risks related to pathogens such as non-spore forming bacteria and viruses. However, it is highly improbable that the risks related to more resistant biological hazards can be reduced. The application does not provide clear information about the location of the system and the origin of the material for containment. This has important implications on the risk related to the transport of the material. The design of the plant does not meet the requirements laid down in current legislation for handling of ABPs after their collection. Only a generic HACCP plan was provided and it was considered inadequate. Major deficiencies were noted in relation to the risks associated with interdependent processes, in particular, as regards to the biofilter, the opening of the bioreducer and the ability to sample for Transmissible spongiform encephalopathies surveillance. The biofilter was not considered effective in containing the risk of aerogenic transmission of biological agents and it is accessible to living vectors. Moreover, there is a risk of release of pathogens to the environment when opening the vessel. Therefore, the whole system cannot be considered as a closed system. The proposed Bioreduction method cannot be considered as a safe alternative method for on farm containment of animal by-products.

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KEY WORDS

animal by-products, alternative methods, containment, Bioreduction system

¹ On request from the Competent Authority of the United Kingdom, Question No EFSA-Q-2013-00609, adopted on 4 December 2013.

² Panel members: Olivier Andreoletti, Dorte Lau Baggesen, Declan Bolton, Patrick Butaye, Paul Cook, Robert Davies, Pablo S. Fernandez Escamez, John Griffin, Tine Hald, Arie Havelaar, Kostas Koutsoumanis, Roland Lindqvist, James McLauchlin, Truls Nesbakken, Miguel Prieto Maradona, Antonia Ricci, Giuseppe Ru, Moez Sanaa, Marion Simmons, John Sofos and John Threlfall. Correspondence: biohaz@efsa.europa.eu

³ Acknowledgement: The Panel wishes to thank the members of the Working Group on Bioreduction: Avelino Álvarez-Ordóñez, Reinhard Böhm, John Griffin and Christophe Nguyen-The for the preparatory work on this scientific opinion and, EFSA staff: Fulvio Barizzone for the support provided to this scientific opinion.

Suggested citation: EFSA BIOHAZ Panel (EFSA Panel on Biological Hazards), 2013. Scientific Opinion on Bioreduction application. EFSA Journal 2013;11(12):3503, 13 pp. doi:10.2903/j.efsa.2013.3503

Available online: www.efsa.europa.eu/efsajournal

SUMMARY

Following a request from the UK Competent Authority, the EFSA Panel on Biological Hazards (BIOHAZ) was asked to deliver a scientific opinion on the 'Bioreduction' application.

The application concerns a method for on-farm containment of animal by-products (ABPs) of ovine origin (such as fallen sheep and placentas) that is regarded as a Category (Cat.) 1 ABP material as defined in Reg. (EC) 1069/2009⁴ (the ABP regulation) for the purpose of the assessment.

The Bioreduction system consists of the aerobic degradation of ABPs in a vented, leak-proof vessel (called a "bioreducer" and directly buried in the soil), containing water, where the contents are heated and aerated. The parameters given by the applicant for heating and aeration rate are respectively: temperature 30-42 °C and aeration under a pressure of 40-55 kPa. These conditions create a favourable environment for bacterial degradation of carcasses resulting in their partial breakdown and a volume reduction through the loss of water vapour.

The remaining material, called liquor, is removed using a vacuum system and subsequently disposed of as a Cat. 1 ABP according to the provision of Art. 12 of the ABP Regulation.

According to the applicant, the bioreducer must be linked to a pipe for gaseous emissions equipped with appropriate filters to prevent the transmission of diseases communicable to humans and animals. Such filters consist of a biofilter bed placed outdoor and made of woodchip and compost. In addition, the bioreducer has to be placed at a dedicated site that ensures that there is no unacceptable risk for the transmission of diseases communicable to humans or animals.

The assessment of the application was performed taking into account the criteria laid down in Art. 20, point 5 of the ABP Regulation and it only considered biological hazards. The terminology used conforms to the EFSA Statement on technical assistance on the format for applications for new alternative methods for animal by-products⁵.

It was concluded that the Bioreduction system can reduce the risks related to pathogens such as non-spore forming bacteria and viruses. However, it is highly improbable that the risks related to more resistant biological agents (e.g. bacterial spores and Transmissible spongiform encephalopathies (or TSE) agents) can be reduced.

The application did not provide clear information about the location of the system (on-farm or outside the farm) and the origin of the material to be treated (only from the farm where the system is installed or also originating from other farms). This has important implications on the risk related to the transport of the material. Moreover, the design of the plant does not meet the requirements laid down in current legislation for the handling of ABPs after their collection.

Only a generic HACCP plan was provided and it was considered inadequate.

The Panel noted major deficiencies in relation to the risks associated with interdependent processes, in particular, as regards to the biofilter, the opening of the bioreducer and the ability to sample for TSE surveillance. The biofilter was not demonstrated to be effective in containing the risk of aerogenic transmission of biological agents. Moreover, it is accessible to living vectors. A risk of release of pathogens to the environment when opening the bioreducer was identified. Therefore, the whole system could not be considered as a closed system.

⁴ Regulation (EC) No 1069/2009 of the European Parliament and of the Council of 21 October 2009 laying down health rules as regards animal by-products and derived products not intended for human consumption and repealing Regulation (EC) No 1774/2002 (Animal by-products Regulation). OJ L 300, 14.11.2009, p. 1-33.

⁵ EFSA Panel on Biological Hazards (BIOHAZ) 2010. Statement on technical assistance on the format for applications for new alternative methods for animal by-products. EFSA Journal 2010;8(7):1680, 12 pp. doi:10.2903/j.efsa.2009.1680

It was concluded that the proposed Bioreduction method cannot be considered as a safe alternative method for on-farm containment of animal by-products.

The BIOHAZ Panel recommended that before considering authorisation of on-farm containment systems in the future it is essential that they comply with Annex IX to Reg. (EU) 142/2011⁶, have a fully operational HACCP plan according to Annex VII to Reg (EU) 142/2011 and there is provision for regular monitoring by the competent authority.

⁶ Commission Regulation (EU) No 142/2011 of 25 February 2011 implementing Regulation (EC) No 1069/2009 of the European Parliament and of the Council laying down health rules as regards animal by-products and derived products not intended for human consumption and implementing Council Directive 97/78/EC as regards certain samples and items exempt from veterinary checks at the border under that Directive. OJ L 054, 26.2.2011, p. 1-254.

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BACKGROUND AS PROVIDED BY THE UK COMPETENT AUTHORITY

On behalf of the Competent Authority for the Animal By-Products Regulations in Wales, I am sending you an application dossier for a full assessment by EFSA of Bioreduction as an alternative method for on farm containment of animal by-products.

The application dossier includes a number of supporting documents which have also been listed in the enclosed Index.

Please also find enclosed the competent authority's (Welsh Government) evaluation report on whether the attached application complies with the standard format for applications for ABP alternative methods as required by Reg. (EC) No 1069/2009, Art. 20, points (2) & (3) & Reg. (EU) No 142/2011, Annex VII.

TERMS OF REFERENCE AS PROVIDED BY THE UK COMPETENT AUTHORITY

The UK competent authority asked EFSA to assess the Bioreduction method as an alternative method for on farm containment of animal by-products.

ASSESSMENT

1. Introduction

The terminology used in this assessment conforms to the “Statement on technical assistance on the format for applications for new alternative methods for animal by-products” (EFSA Panel on Biological Hazards (BIOHAZ), 2010). The assessment only considered biological hazards. Other hazards (e.g. physical, chemical or radiological) are not considered.

The assessment of the application received was performed taking into account the criteria laid down in Art. 20, point 5 of Reg. (EC) 1069/2009⁷ (the Animal By-Products Regulation).

2. Full description of the process

According to Annex VII to Reg. (EU) 142/2011⁸ the applicant is required to provide a full description of the process to be assessed.

The following text provides a summary of the information received.

The application concerns a system (Bioreduction) for containment of animal by-product (ABP) material of ovine origin (e.g. fallen sheep carcasses and placentas).

The Bioreduction system consists of the aerobic degradation of ABPs in a vented, leak-proof vessel (called a bioreducer, see Figure 1), containing water, where the contents are heated and aerated. The carcasses and other material are added as they become available. Water has to be regularly added into the bioreducer to facilitate carcass degradation. According to the applicant, the water level must be maintained so that at least two-thirds of each carcass is submerged at all times.

The parameters given by the applicant for heating and aeration rate are respectively: temperature 30-42 °C and aeration 40-55 kPa. The information provided in Figure 1 appears to indicate that a pressure of 50 kPa is applied for 45 minutes of every hour. According to the applicant, these broad temperature/aeration combinations allow for some flexibility in the system but maintain lightly aerated, mesophilic conditions. These moist and warm conditions create a favourable environment for bacterial degradation of carcasses resulting in their partial breakdown and a volume reduction through the loss of water vapour. However, it is noticed that in several parts of the application it is mentioned that the vessels may be switched off to reduce costs.

⁷ Regulation (EC) No 1069/2009 of the European Parliament and of the Council of 21 October 2009 laying down health rules as regards animal by-products and derived products not intended for human consumption and repealing Regulation (EC) No 1774/2002 (Animal by-products Regulation) OJ L 300, 14.11.2009, p. 1-33.

⁸ Commission Regulation (EU) No 142/2011 of 25 February 2011 implementing Regulation (EC) No 1069/2009 of the European Parliament and of the Council laying down health rules as regards animal by-products and derived products not intended for human consumption and implementing Council Directive 97/78/EC as regards certain samples and items exempt from veterinary checks at the border under that Directive. OJ L 054, 26.2.2011, p. 1-254.

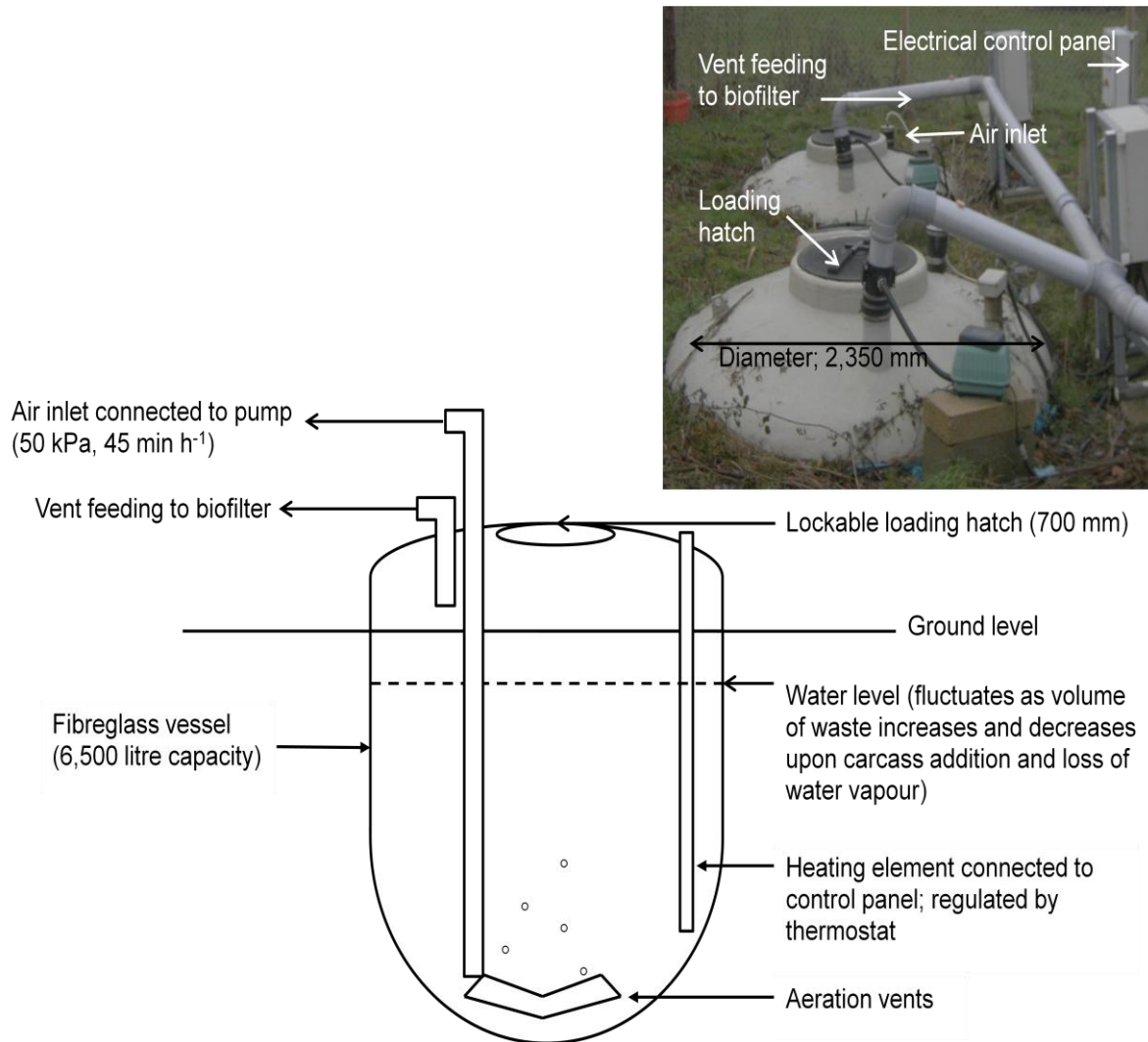


Figure 1: Photograph and cross-sectional diagram of a bioreduction vessel as provided in the applicant's report.

The material to be contained should be collected on a holding for which the Competent Authority has authorised the use of the method. From the application received it is unclear if the material to be stored can also be collected outside the holding. However, it appears that the Bioreduction system can also be placed outside livestock holdings. This raises questions about the vehicles that would be used to transport the material and on their management.

The material must then be placed as soon as possible into a bioreducer with the following characteristics:

- have a device to close and lock it;
- be water-proof and leak-proof;
- be coated in a way which prevents corrosion;
- be equipped with a device for controlling emissions;
- be supplied with a source of heating and air;

According to the applicant, the bioreducer must be constructed and laid out in accordance with European Union legislation for the protection of the environment and must be linked to a pipe for gaseous emissions, which must be equipped with appropriate filters to prevent the transmission of diseases communicable to humans and animals. In the Bioreduction system, as described by the applicant, the gases generated within the vessel exit through the venting system into a biofilter bed comprised of woodchip and compost (approximately 70:30 by volume). The biofilter bed is mainly designed to prevent odours and is approximately 1 m × 1.5 m × 1 m in dimension. The biofilter should be disposed of as Cat. 1 ABP material.

According to the applicant, the bioreducer has to be placed at a dedicated site that ensures that there is no unacceptable risk for the transmission of diseases communicable to humans or animals. However, no specific criteria are mentioned in the application to ensure that an unacceptable risk does not arise. It is important to highlight that the system is placed outdoors, which raises concerns about the accessibility of the biofilter to living vectors (e.g. birds and rodents). However, no detailed information is given on how to protect the biofilter from these living vectors. Also, no information is provided in relation to the replacement of the biofilter.

The design of the proposed Bioreduction plant does not meet the requirements laid down in Annex IX, chapter II of Reg. 142/2011 (e.g. the plant must have a covered space to receive and dispatch animal by-products and must be constructed in such a way that it is easy to clean and disinfect).

The liquor resulting from the Bioreduction process has to be collected and disposed of as a Cat. 1 ABP material according to the provision of Art. 12 of the ABP Regulation. According to the applicant, the frequency of liquor removal will depend on usage of the container but should be a minimum of once a year in order that integrity checks on the facility can be made. A minimum time from the addition of the last material into the bioreducer to the removal of the hydrolysed material is not given. The applicant reports that the solid fraction remains within the tank until liquefaction. That would imply that the vessel will never be completely emptied. Moreover, it is questionable whether all the solid material (e.g. bones and soil) will liquefy. No information is given about intervals and possible methods of total cleaning of the bioreducer. The applicant reports that the liquor is removed using a vacuum system but does not provide technical explanation on how this is performed and on how it is ensured that contamination of the exterior with the removed material is avoided.

A schematic diagram of the Bioreduction process is available in Figure 2.

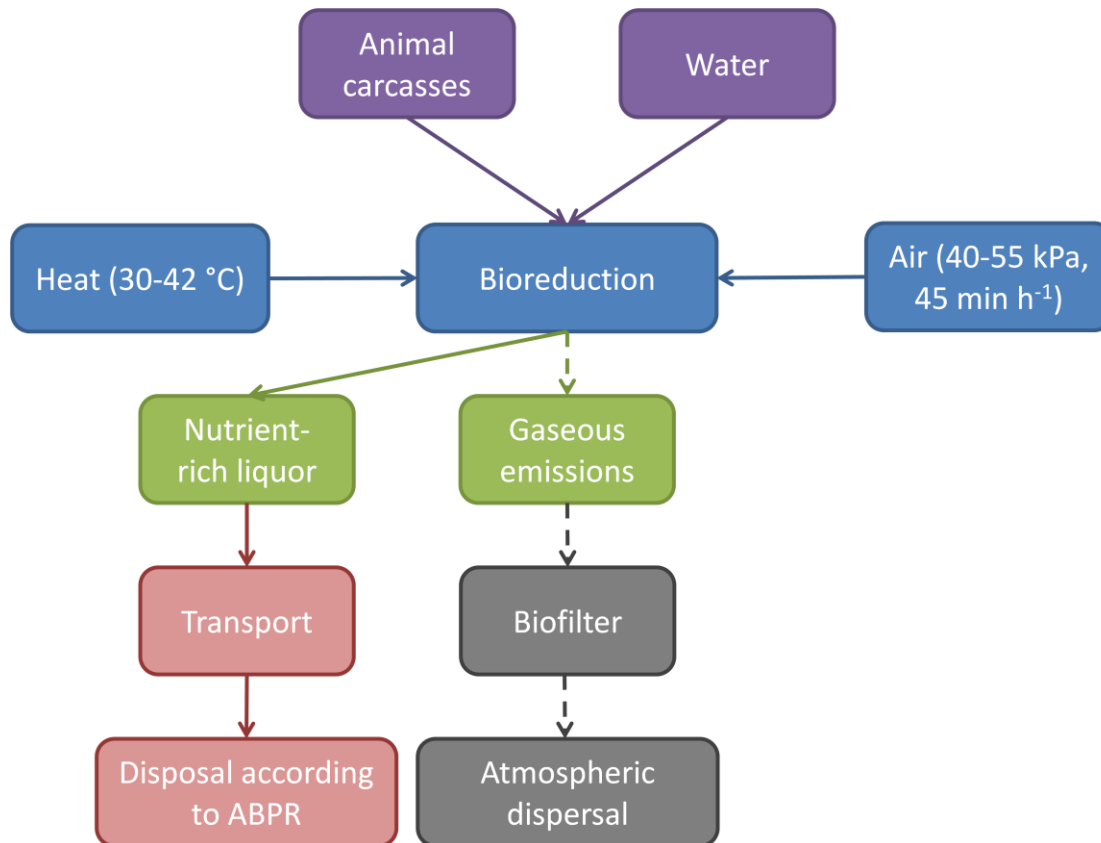


Figure 2: Schematic diagram of the bioreduction process as provided in the applicant's report.

It is important to highlight that, in general, long term storage of fallen animals on farm could result in a failure to identify and report infectious diseases to the veterinary authorities (EFSA Panel on Biological Hazards (BIOHAZ), 2012). Also, in this case, it may compromise the surveillance programme for Transmissible spongiform encephalopathies (TSEs) in sheep. There is no information available in the application about sampling for TSEs surveillance when the Bioreduction system is applied.

3. Full description of the material to be treated

Cat. 2 and 3 material of ovine origin identified in Article 9(f)(i), (ii) and (iii) and in Article 10(h) of the ABP Regulation will be treated. Since that material also comprises fallen sheep containing Specified Risk Material (SRM), which is classified as a Cat. 1 material according to Art. 8(b)(i) of the above-mentioned regulation, the whole material is regarded as a Cat.1 ABP material for the purpose of this assessment.

4. Hazard identification

The applicant does not provide a comprehensive list of pathogens for humans and animals which could be present in the material to be treated (e.g. spores from bacterial pathogens such as *Bacillus anthracis*, *Coxiella burnetii*, *Toxoplasma gondii*, foot and mouth disease virus etc... are not mentioned). However, *Campylobacter*, *Escherichia coli*, *Salmonella*, parvovirus and prions are identified by the applicant as potentially present in the material to be treated. Prions must be considered as the most resistant biological hazards potentially present in the material to be treated.

5. Level of risk reduction

In principle, a new proposed process should be able to reduce the amount of the most resistant biological hazards associated with the category of the material to be processed for a defined final use to an acceptable level. However, this application concerns the storage of material that will be disposed

of according to standard methods for Cat. 1 ABP material; hence all the risks would be eliminated by this last step. Therefore, a validation experiment of the risk reduction in the material is not needed in this case.

The storage of ABP may change the microbiological properties of the material (including increasing or decreasing the number of pathogens) and this aspect was considered in this particular case. In this context, a number of studies were undertaken to establish the fate of microorganisms within a bioreduction vessel (Williams et al., 2009; Gwyther et al., 2012; Gwyther et al., 2013). In the latter study, a number of bacterial agents (e.g. *Enterococcus faecalis*, *Salmonella* spp., *Escherichia coli* O157) and parvovirus were seeded within the bioreduction vessel under normal operating conditions and when no aeration and heating were applied. Under normal operating conditions, the results showed (i) an initial sharp decrease for *E. faecalis* and a subsequent progressive increase until the end of the study, (ii) a >5 logs reduction in *Salmonella* spp. numbers, (iii) a reduction in *E. coli* O157 to below the limit of detection by enrichment, and (iv) a 3-log reduction in parvovirus infectivity by day 7 of the study. According to Gwyther et al. (2013), the increase of *E. faecalis* was due to the addition of new carcasses rather than to the growth of the inoculated strains. The results of this experiment indicate that pathogens having comparable survival patterns (e.g. non-spore forming bacteria and viruses) would follow similar inactivation kinetics and would not proliferate. However, no information is provided about the inactivation of bacterial spores and TSE agents.

The efficacy of the containment of the hazards within the vessel must also be considered. The possibility of escape of pathogens through the biofilter and when opening the vessel is a critical aspect of the application.

In the study carried out by Gwyther et al. (2013), the biofilters were examined for the presence of *Salmonella* spp. and *E. faecalis*. Under normal operating conditions, high numbers of both microorganisms were found in the biofilter samples after inoculation of the vessels and addition of the carcasses, and although bacterial numbers declined over time, *E. faecalis* and *Salmonella* spp. were still detected more than 60 days after the beginning of the study. In addition, low numbers of *E. faecalis* and *Salmonella* spp. were detected from bioaerosols above the biofilter (3.3 CFU/m³ on day 0.04 and 3.3 CFU/m³ on day 8, respectively). Hence, the biofilter used is not appropriate to avoid the external contamination with the pathogens potentially present in the material to be treated.

Another source of environmental contamination would be the opening of the container during operation. Low numbers of bacteria were detected in bioaerosols released from the opening hatch (*Salmonella* spp. were detected until day 8 at 2.2 CFU/m³). As the vessel can be continuously fed with new carcasses, there is the risk of release of pathogens in the environment during this operation.

No tests were performed on the containment of prions. Instead, an expert elicitation was conducted that highlighted aerosol production during operation as a concern.

The Bioreduction system should be considered as an open system.

6. HACCP plan

A HACCP plan was submitted as part of the application but it was considered inadequate for a number of reasons, including the following:

- the critical limits were not always directly related to the critical control points, e.g. the critical control point for dealing with the hazard “vermin gaining access to carcasses awaiting to go into the vessel” was “boundary to site” and the critical limit is “100% carcasses into the vessel without delay”;

- the procedure for monitoring the critical limits for some critical control points were inadequate, e.g. a daily pressure gauge check to monitor that the air pressure in the tank was higher than 50 kPa for 45 minutes of every hour;
- in some cases, the verification procedures (Step 6) seemed to be the same as the monitoring procedures (Step 4);
- some of the corrective actions presented in the plan were considered not feasible (e.g. cleaning and disinfection of soil after a spillage);
- critical aspects concerning the containment of the hazards (such as the integrity and disinfection of the vessels and functioning of the biofilter) were not addressed.

7. Risk associated with interdependent processes

Some risks associated with interdependent processes related to:

- the identification and reporting of diseases;
- the possibility to carry out sampling for TSE surveillance;
- the waste air treatment;

have already been described under points 2 and 5 of this assessment.

Plans to control pests (birds, rodents, insects and other vermin), as set out in Reg. 142/2011, are mentioned but not described in detail.

In addition, there is no possibility to inspect the vessels for leakage since they are directly buried in the soil without protection and no precautions are foreseen to protect the environment in case of an accidental leakage.

Generally, the application indicates that the Bioreduction system would be placed on-farm. If only fallen animals from that farm are stored in the bioreducer, the risk of transmission of infectious diseases from farm to farm due to transport would be limited. However, as already mentioned, the system could hinder the identification and reporting of diseases to the competent authorities.

A part of the document deals specifically with the requirement that would apply if the system is located on a livestock holding, suggesting that the Bioreduction system can also be placed outside livestock holdings. It is not clear what vehicles would be used to transport the carcasses to such sites and whether these vehicles would comply with the requirements (e.g. use of covered leak-proof containers or vehicles) set out in Annex VIII Regulation (EC) 142/2011. In addition, the application form does not explicitly state whether or not a Bioreduction system on a particular farm would be used exclusively for storing ABP material originating on that farm. It is important to note that the movement of such material to a dedicated site outside the farm or between farms could present a risk of transmitting infectious agents.

8. Risk associated with the intended end use of the product

The material resulting from the Bioreduction process will be treated according to standard methods for Cat. 1 ABP and these methods ensure the appropriate inactivation of the possible hazards present in the hydrolysed material.

CONCLUSIONS AND RECOMMENDATIONS

CONCLUSIONS

- The application concerns a containment system for animal by-products of category 1 as defined under Regulation (CE) 1069/2009. The system consists of the aerobic degradation of ABP in a vented, leak-proof vessel. The resulting material is finally disposed of according to one of the standard methods for category 1 animal by-product material.
- The Bioreduction process can reduce the risks related to pathogens having survival patterns comparable to the test organisms used in the validation (i.e. non-spore forming bacteria and viruses). It is highly improbable that the risks related to more resistant biological agents (e.g. bacterial spores and TSE agents) can be reduced using this method.
- The application received does not provide clear information about the location of the system (on-farm or outside the farm) and the origin of the material to be treated (only from the farm where the system is installed or also originating from other farms). This has important implications on the risk related to the transport of the material.
- The design of the plant does not meet the requirements laid down in current legislation for the handling of animal by-products after their collection.
- Only a generic HACCP plan was provided and it was considered inadequate.
- The Panel noted major deficiencies in relation to the risks associated with interdependent processes, in particular, as regards to the biofilter, the opening of the bioreducer and the ability to sample for TSE surveillance.
- The biofilter was not demonstrated to be effective in containing the risk of aerogenic transmission of biological agents. Moreover, it is accessible to living vectors. There is also a risk of release of pathogens to the environment when opening the bioreducer. Therefore, the whole system cannot be considered as a closed system.

Answer to ToR

- The proposed Bioreduction method cannot be considered as a safe alternative method for on-farm containment of animal by-products.

RECOMMENDATIONS

- Before considering authorisation of on-farm containment systems in the future it is essential that they comply with Annex IX of Reg (EC) 142/2011, have a fully operational HACCP plan according to Annex VII to Reg (EU) 142/2011 and there is provision for regular monitoring by the competent authority.

DOCUMENTATION PROVIDED TO EFSA

1. Bioreduction application dossier. June 2013. Submitted by the Animal Health and Veterinary Laboratories Agency, London, UK.

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- EFSA Panel on Biological Hazards (BIOHAZ) 2010. Statement on technical assistance on the format for applications for new alternative methods for animal by-products. *EFSA Journal* 2010;8(7):1680, 12 pp. doi:10.2903/j.efsa.2009.1680
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