

Selection and handling of the test manure

- Manure from control animals (not from storage tanks) animals kept in well defined conditions, description of feed should be given + vet history.
- Manure can be stored at -20 °C before the study is conducted.
- Acclimation period before spiking manure: 21 days (info should be given, why this is necessary!). Necessary to obtain anaerobic conditions
- Standard manure based on fixed dry matter content: 10 ± 1% cattle manure, 5% ± 1% pig manure (Missing information: data on beef cattle) 60% ± 5% chicken manure
- Info in literature is too limited to determine worst case or average conditions
- Preference can be given to the manure type of the production type in question
- If more target animals are involved select one manure type representative to other target animals

Matrix characterisation of the manure

- Proposed test parameters:
 - temperature
 - pH
 - (intestinal) microbial activity (BOD)
 - organic matter (TOC)
 - N and P content
 - Redox potential
 - dry matter content
 - moisture content
 - oxygen content?

Establishing test conditions

- Anaerobic (cattle and pig)
It should be demonstrated that an attempt is made to perform the study under anaerobic conditions, e.g. operational defined by discontinuous N flow
low redox potential
- Aerobic (poultry)
Start with a standard moisture content level (e.g. 40%)
or to mimic the stable conditions
- Preference is given to spiked manure, but it up to the applicant to deviate from it (and use manure from treated animals)

Establishing test conditions

- Redox potential
 - Cattle and pigs on stables → Anaerobic
 - Poultry (rabbit, duck?) → Aerobic

Can we define a threshold redox potential?
- Moisture content
 - Poultry (including turkey ?) → dry (50%)
 - Cattle and pigs on stables → wet
 - rabbit, turkey, duck → dry (%?)
- Test duration

100 days
- Sterile control useful to make distinction between biotic and abiotic degradation. May be more useful in case study is conducted with non-radio labelled substance. Question: will it remains sterile?
- Preferred test temperature is 20°C for all manure types (extrapolated to realistic field conditions)

Test substance

- Radiolabelled material preferred because:
 - mass balance is easier to achieve
 - facilitates the interpretation of test results
(recovery, transformation, fraction of bound residue)
 - mineralization easier detected
 - possibly improve the detection limits

Extraction and determination of the test substance, metabolites and non extractable residues

- The aim of the study is to demonstrate transformation (formation of metabolites / CO₂ production)
- Recommended to follow a sequential extraction method (including reactive method)
- If at T=0 less than xx % can be extracted, it will be difficult to demonstrate degradability
- BR is considered to be active ingredient
- Extraction should not disrupt the active ingredient or its metabolites.
- BR can only be disregarded when it can be demonstrated (by tox studies) that it is not available / does not cause toxicity

Extrapolation of biotic and abiotic transformation of VMPs within and between manure types of different target species

- In principle extrapolation from major species should be possible but info on manure from minor species will be useful.