

Requirements for Measurement and Validation of Biochemical Methane Potential (BMP)*

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†For more information and other documents, visit <https://www.dbfz.de/en/BMP>. For document version history or to propose changes, visit <https://github.com/sashahafner/BMP-methods>.

1 Introduction

This document presents the minimal requirements for measurement and validation of biochemical methane potential (also called biomethane potential) (BMP) in batch tests. It represents the consensus of more than 50 biogas researchers. The list of requirements is the same as in the open-access commentary by [Holliger et al. \[2021\]](#). For details on development of these requirements see the open-access papers [Holliger et al. \[2016\]](#) and [Hafner et al. \[2020c\]](#).

2 Requirements for BMP measurement

2.1 Analysis of substrate and inoculum

Volatile solids (VS) content of inoculum and substrate is needed to determine quantities for a selected inoculum-to-substrate ratio (ISR) and for calculation of BMP. For details on measurement of total solids (TS) and VS see the detailed document from the US EPA [\[EPA, 2001\]](#) (open-access) or other sources (e.g., [Strach \[2020\]](#) (open-access) or [Baird et al. \[2017\]](#)).

1. Total solids (TS). Measure for the inoculum and all substrates, by drying at 105°C in triplicate. TS is needed only for determination of volatile solids (VS) content.
2. Volatile solids (VS). Measure for the inoculum and all substrates by igniting the dried sample at 550°C in triplicate. VS is determined from mass loss.

2.2 Test setup and duration

1. Samples. All BMP trials must include three types of samples: batches with only inoculum (“blanks”), with inoculum and microcrystalline cellulose as a positive control¹, and with inoculum and substrate.
2. Replication. All tests must include at least 3 batches (bottles) for each condition.² The minimum number of batches used in a BMP test with one substrate is therefore 9 (3 blanks, 3 cellulose, 3 substrate).
3. Duration. Terminate BMP tests only after daily methane (CH₄) production from individual batches during 3 consecutive days is < 1.0% of the net accumulated volume of CH₄ from the substrate (substrate batch minus average of blanks). For manual or other methods where measurements are

¹Other positive control substrates could be used in the future [\[Koch et al., 2020\]](#), but only cellulose has had extensive testing that was used to develop the validation criteria described below in Section 4 [\[Hafner et al., 2020c\]](#).

²If a bottle is lost through, e.g., breakage, resulting in $n = 2$ for any condition, results cannot be validated. Therefore it is prudent to include 4 replicates, especially for blanks. Outliers can be eliminated if there is good reason to suspect there was an error in measurement (e.g., leakage) but the remaining number of replicates must be at least 3.

not made every day, termination can take place at the end of the first measurement interval of at least 3 days where the rate of production drops below the 1% maximum (or two or more intervals that sum to at least 3 days, all with rates below the 1% maximum). If different substrates are tested, each substrate can be terminated when the slowest of the 3 replicate batches has reached the termination criterion. Blanks must be continued as long as the slowest (latest) batch with substrate. Continuing tests beyond this 1% net duration is acceptable and can help ensure that validation criteria are met (Section 4).

3 Calculations

1. Data processing. Standardized CH₄ volume (dry, 0°C, 101.325 kPa) is calculated from laboratory data using standardized methods.³
2. BMP units. BMP is expressed in standardized CH₄ volume (dry, 0°C, 101.325 kPa, referred to as “normal” volume) per unit mass of substrate organic matter added (typically VS but sometimes chemical oxygen demand (COD)) (often written as NmL_{CH₄} g_{VS}⁻¹).
3. Calculation of BMP. BMP of all substrates (including cellulose) is calculated by subtracting inoculum CH₄ production (determined from blanks) from gross (total) CH₄ production from substrate with inoculum, and normalizing by substrate VS mass. Any differences in inoculum or substrate mass among batches must be reflected in calculations. Calculations must follow a standardized approach.⁴
4. Calculation of BMP standard deviation. The standard deviation associated with each mean ($n \geq 3$) BMP value must include variability from both blanks and batches (bottles) with substrate and inoculum, along with uncertainty in the mass of substrate VS added.⁵

4 Validation criteria

BMP results that meet *all* the following criteria can be described as “validated”.⁶ Otherwise, results are not validated, and tests must be repeated.

1. All required components of the BMP measurement protocol listed above (Section 2) are met (including duration) and calculations are done as described above (Section 3).

³Detailed descriptions of calculations are available for the following measurement methods in the Standard BMP Methods collection (<https://www.dbfz.de/en/BMP>): volumetric (document 201) [Hafner et al., 2020f], manometric (document 202) [Hafner et al., 2020a], gravimetric (document 203) [Hafner et al., 2020g], and gas density (document 204) [Hafner et al., 2020d].

⁴Calculation of BMP is described in detail in document 200 [Hafner et al., 2020b].

⁵See document 200 [Hafner et al., 2020b].

⁶The criteria listed above are duplicated in document 101 [Hafner et al., 2020e], which was created to simply make it easier to find these required criteria.

2. Mean cellulose BMP is between 340 and 395 NmL_{CH₄} g_{VS}⁻¹.
3. Relative standard deviation for cellulose BMP (standard deviation, including variability in blanks, substrate bottles, and added substrate VS, divided by mean BMP) is no more than 6%.

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