METHOD SPECIFICATION Faculty of Biosciences, NMBU

Method name: Tryptophan BIOVIT No .: Met 1051

1. Analysis method / Principle / Main instrument

The method determines the total content of tryptophan in feed and faeces. The method does not distinguish between D and L- tryptophan.

To determine total tryptophan, the sample is hydrolyzed under alkaline conditions (in a saturated barium hydroxide solution) at 110 °C for 20 hours. An internal standard is added before hydrolysis. The concentration of tryptophan and internal standard in the hydrolysate is determined by liquid chromatography (HPLC) with fluorescence detector.

<u>Main instrument:</u> Ultimate 3000 HPLC with auto-injector (Thermo Scientific) and fluorescence detector (Shimadzu).

2. Reference and any modifications

Commission Regulation (EC) No 152/2009. 27 Jan 2009. Laying down the methods of sampling and analysis for the official control of feed. Annex III, P, Official Journal of the European Union L54 / 1 from 26/02/2009

3. Requirements for grinding and storage

Samples must be grinded to 0.5 mm. Moist samples must either be air-dried (at a temperature not exceeding 50 °C) or lyophilized before grinding. Samples with a high fat content, e.g. fish feed with an extra high (> 40%) fat content, are extracted with petroleum ether (b.p. 40-60 °C) before grinding. The weighed analytical sample should contain approx. 10 mg nitrogen.

4. Contact persons

Lab manager: Hanne Kolsrud Hustoft Responsible for analysis: Elin Follaug Johnsen

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