Method name: Urea
BIOVIT-nr.: Arb_1012_feed and muscle

1. Introduction

The method is intended for the determination of urea in feed and muscle.

The fish fed on diets that differed in levels of urea supplementation impact the incidence of salmon with ulcer, and subsequently its mortality that seems to relate to plasma osmolality. According to Rùrvik et al. (2000) the dietary urea supplementation may lead to reduced development of skin ulcers and will probably increase the proportion of market size salmon of superior quality.

Main instrument: RX Daytona + (Randox Laboratories Ltd, UK). 55 Diamond Road, Crumlin, County Antrim, BT29 4QY, United Kingdom. Kit: Urea (UR8334).

Urea concentration can be measured photometrically on a RX Daytona + spectrophotometer.
Reportable range: 0.50-62.0 mmol/L

2. Reagents:

Randox UREA Assay (R1 and R2)
Clinical Chemistry Calibrator Level 2 CAL2350
Clinical Chemistry Calibrator Level 3 CAL2351
Quality control (Assayed Chemistry Premium Plus Level 3, Cat No-HE1532)

3. Risk assements:

None.

4. Recommended equipment:

50 mL test tube (Greiner) with screw cap, and V bottom
Erlenmeyer flask
Filter paper (Whatman™)
Double Spatula
Tweezers
Pipettes and pipette tips 1-5 mL
Dispenser tips Combitips advanced® Eppendorf Quality, 50mL
Bench centrifuge with a swing-out rotor
Weighing scale

Vortex mixer
RX Daytona + instrument

A control sample containing 5.1-6.0 % urea, should be run with each batch.
For feed analysis, 1g ± 5 mg sample is needed. Degree of grinding: 0.5 mm.
For urea muscle analysis, a 15-20 g ± 50 mg of homogenized sample is needed.

Feed samples must be stored from 2 to 8 °C.
Muscle sample must be stored at -20 °C.

5. Job description

a) If feed sample, weigh in about 1 g into 50 mL plastic tube w/ screw cap, or weigh in about 20 g if muscle sample.
b) Make a note of the exact sample weight.
c) Add Milli-Q water up to 50ml mark in the sample tube to the sample and shake it well for 10 sec on Vortex. Note the exact amount of added water to each sample.
d) Centrifuge the samples for 15 min at 2500g and 10°C.
e) Filter the sample and collect the supernatant in Erlenmeyer flask.
f) Pipette 300 mL of the supernatant into Eppendorf tubes without caps.
g) The samples are analyzed on a RX Daytona + fully automatic analyzer (See in MpsArb_2020 Kliniske analyser Daytona +).

6. Calculation

% Urea= ((abs sample*MW) *0,05) / mg sample)/10
abs. sample = urea absorbance read on spectrophotometer (mmol / l)
60,06 = MW-molecular weight of urea (mg / mmol)
0.05 = Amount of ml water added
mg sample = weighed sample
10 = the calculation in %

7. Literature:

https://journals.physiology.org/doi/full/10.1152/advan.00027.2002

https://acutecaretesting.org/en/articles/urea-and-the-clinical-value-of-measuring-blood-urea-concentration

https://www.researchgate.net/publication/230107985_Urea_in_feeds_for_sea_water_farmed_Atlantic_salmon_Effect_on_growth_carcass_quality_and_outbreaks_of_winter_ulcer