

Chromium Analysis: Spectrophotometric Method

Materials:

5.5 cm aluminum pans
Drying oven, 100 °C
Muffle furnace, 470 °C
Micro-kjeldahl flasks, calibrated to 110 ml
Glass beads
Micro-kjeldahl digestion burners
Screw cap storage tubes
Spectrophotometer

Reagents:

Perchloric Acid digestion mixture: 5 g sodium molybdate (MCB Cat # SX690, FW 241.97, CAS # 7631-15-0 or Baker Cat # 3764-01, FW 241.95, CAS # 10102-40-6) dissolved in 1 liter 18 MOhm water, when dissolved, add 1 liter 70% Perchloric Acid (EMD Cat # PX0396A-7, FW 100.46, CAS # 7601-90-3).

Procedure:

1. Dry duplicate samples (0.5000 g high compound samples, % recovery samples; and 1.0000 g for low control samples, fecal, and duodenal samples) for 12-24 hours in a 100 °C drying oven. Cool in desiccator, weigh, and record. Include in sample run 3 of each of the following: chromium blank, chromium % recovery, high chromium pool, and low chromium pool.
2. Ash samples for 12-24 hours at 470 °C. Cool, weigh, and record weight.
3. Quantitatively transfer ash to 110 ml micro-kjeldahl flasks.
4. For standard curve, weigh 2.5-30 mg portions of pure chromium oxide powder and carry them through the following method with the samples.

Chromium standard series: 2.5 mg, 5 mg, 7.5 mg, 15 mg, and 30 mg

5. Add 2-3 glass-boiling beads to each flask.
6. Add 15 ml perchloric acid digestion mix to each flask.
7. Heat micro-kjeldahl flasks on digestion burners (medium heat, about 200 °C) until a yellow, orange or red color appears. After color change, heat for an additional 10-15 minutes (fumes should hang in the neck of the flask), unplug the burners and let flasks cool. **Do not boil dry.**
8. Dilute to volume (110) ml with 18 MOhm water. Stopper each flask and mix well by inversion. Samples may be transferred to screw capped storage tubes. Samples are stable indefinitely.
9. Read absorbance on a spectrophotometer with a wavelength set at 440 nm.

Calculations:

$$\% \text{ Recovery Pool} = \frac{0.007 \text{ g Chromic oxide}}{\text{g fecal sample}} = \frac{0.00479 \text{ g chromium}}{\text{g fecal sample}}$$

$$\% \text{ Recovery} = \frac{\text{Calculated \% Recovery } g/g \times 100}{0.00479}$$

$$\frac{\mu\text{g}}{\text{g}} \text{ unknown} = \frac{(\text{abs in sample})(10 \text{ ml})}{\text{Sample Dry Wt.}}$$

Abs = use regression curve to calculate

110 ml = total sample volume

Dilution Factor = 1 ml filtered sample diluted to a final volume of 10 ml gives a dilution factor of 10.

Reference

Fenton, T. W., and M. Fenton. 1979. An improved procedure for the determination of chromic oxide in feed and feces. *Can. J. Anim Sci.* 59:631-634.