

DETERMINATION OF ESCAPE PROTEIN FROM OMASAL SAMPLES

Updated September 2013

I. References:

Carulla, J.E., V.A. Wilkerson, D.H. Shain, T.J. Klopfenstein and R.A. Britton. 1994. Omasal sampling to estimate escape protein of forages. J. Animal. Sci. (Supplement 1). 72:85.

Hart, F.J. and J. Leibholz. 1990. A note on the flow of endogenous protein to the omasum and abomasum of steers. Animal. Prod. 51: 217-219.

II. Personal Protective Equipment:

- A. Lab coat
- B. Safety glasses/goggles
- C. Latex gloves

III. Procedures:

- A. Evacuate rumen and reticulum
- B. Locate reticulo-omasal orifice
- C. Introduce finger into omasum and grab omasal sample (200 g - fresh weight)
- D. Freeze-dry sample

IV. Required Analysis:

- A. Total N
- B. Purines
- C. Indigestible Acid Detergent Fiber
- D. Acid Detergent Insoluble Nitrogen (ADIN)

V. Principles and Calculations:

A. Escape protein (EP) is considered as the dietary protein that is not degraded in the rumen. In forages, it is considered as the plant protein (PP) that escapes rumen digestion. The mathematical expression of this concept is:

1. $EP = (PP \text{ flowing to small intestine } / PP \text{ intake } g) * 100$

The omasal technique to estimate escape protein of forages assumes that particles of forage found in the omasum will not undergo further degradation in the rumen (e.g. particles will not go back into the rumen). Therefore:

2. $PP \text{ flowing to small intestine } (g) = PP \text{ flowing to omasum } (g)$

3. $EP = (PP \text{ flowing to omasum } (g) / PP \text{ intake } (g)) * 100$

4. $PP \text{ flowing to omasum } (g) = DM \text{ flow to omasum } (g) * \% PP \text{ in omasum DM}$

5. $PP \text{ intake } (g) = DM \text{ intake } g * \% PP \text{ in forage}$

6. Therefore:

a. $EP = \frac{DM \text{ flow to omasum } (g) * \% PP \text{ in omasum DM}}{DM \text{ intake } (g) * PP \text{ in Forage}}$

The flow of DM to omasum over DM intake is considered the Undigested DM, therefore

7. $EP = \text{Undigested DM at omasum} * (PP \text{ in omasum} / PP \text{ in forage}) * 100$

To estimate the Undigested DM at the omasum, we have used an internal marker in the forage, indigestible acid detergent fiber (IADF). The internal marker is a

fraction of the forage that is considered not degradable and therefore:
 Intake of IADF (g) = Flow of IADF at omasum (g) or,
 Intake of DM (g) * % IADF forage = Flow of DM (g) * % IADF in omasal DM

And, (% IADF forage)/ (% IADF omasal DM) = (Omasal DM flow (g))/ (DM intake (g))

8. (% IADF forage)/(% IADF omasal)= Undigested DM at omasum

Combining equations 6 and 7, we can estimate EP

$$EP = ((\% \text{ IADF forage}) / (\% \text{ IADF omasal DM})) * (\% \text{ PP in omasal DM} / \% \text{ PP in forage}) * 100$$

Estimation of Plant Protein in Omasal Samples: In the omasal technique, we have assumed that there are three major fractions of N in the sample

- 1) Plant N
- 2) Microbial N
- 3) Endogenous N

The plant N is estimated by difference:

$$\% \text{ Plant N} = \text{Total N} - \text{Microbial N} - \text{Endogenous N}$$

Microbial N is estimated independently in the sample using purines as microbial marker. Bacteria from the rumen are isolated by differential centrifugation. Total N and purines in the sample are determined. Bacterial N to purine ratio is determined. Omasal samples are analyzed for purines.

Bacterial N in omasal DM g/g = Purines (g/g DM) in omasal sample * Bacterial N to purine ratio: (g N/g purine)

Multiply by 100 to get %

Endogenous N is considered to be proportional to DM intake (2.2g N/kg DM intake). In cases where DM intake is not known, the endogenous N can be estimated based on the undigested DM at omasum.

$$\text{Endogenous N} = \text{DM intake} * 0.22$$

$$\text{Endogenous N at omasum} = \text{DM at omasum} * X$$

$$\text{DM intake} * 0.22 = \text{DM at omasum} * X$$

$$(\text{DM intake} / \text{DM at omasum}) * 0.22 = X$$

$$(1 / \text{undigested DM omasum}) * 0.22 = X$$

$$(\% \text{ IADF omasal DM} / \% \text{ IADF forage}) * 0.22 = X$$

Where X is the percent N of endogenous origin at the omasum as a percent of DM.

Therefore:

$$PP_{om} = \text{Total } N_{om} - \text{Mic } N_{om} - X$$

Remember that all fractions have to be expressed in the same units. It is recommended to use %.

Finally, if EP wants to be expressed as % of DM. The equation will be:

$$EP = \text{Undigested DM at omasum} * (\text{Total } N_{om} - \text{Endogenous } N_{om} - \text{Microbial } N_{om})$$

Or:

$$EP = (\% \text{ IADF forage} / \% \text{ IADF omasal}) * (\text{TN}_{om} - X_{om} - \text{Mic } N_{om})$$

Attention should be given to use the right units for every calculation in order to make no mistakes!!!!

Remember the ratio of IADF of feed and omasal contents will give you a coefficient ranging from 0 to 1. So there is no need to divide per 100 when multiplying the coefficient for the % plant protein in omasal sample. If you wish to do that you may multiply the coefficient by 100 first. That will give you grams of undigested feed at omasum per 100 g of ingested forage.

Adjustments for ADIN should be done after the value of escape protein as a percent of dry matter have been calculated.