

EUROPEAN GRASS SILAGE *IN VITRO* RUMEN UNDIGESTIBLE NDF MEASURES AT 30, 120, AND 240 HOUR COMPARISONS BETWEEN TWO LABORATORIES

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INTRODUCTION:

Forage *in vitro* rumen undigested aNDF, corrected for ash (uNDFom, % of DM) has been related to gut fill and dry matter intake in dairy cattle with recent studies (Cotanch et al., 2014; Grant et al., 2018). While Hall and Mertens (2012) evaluated repeatability of *in vitro* rumen NDF digestibility among 10 laboratories, the authors evaluated short term ruminal incubations (30 hours). Today's uNDFom measures utilize longer rumen incubations, up to 240 hours. Assay repeatability between laboratories for long-term incubations is unknown. The objective here was to evaluate uNDFom repeatability for multiple *in vitro* rumen incubation time lengths, between two forage analysis laboratories.

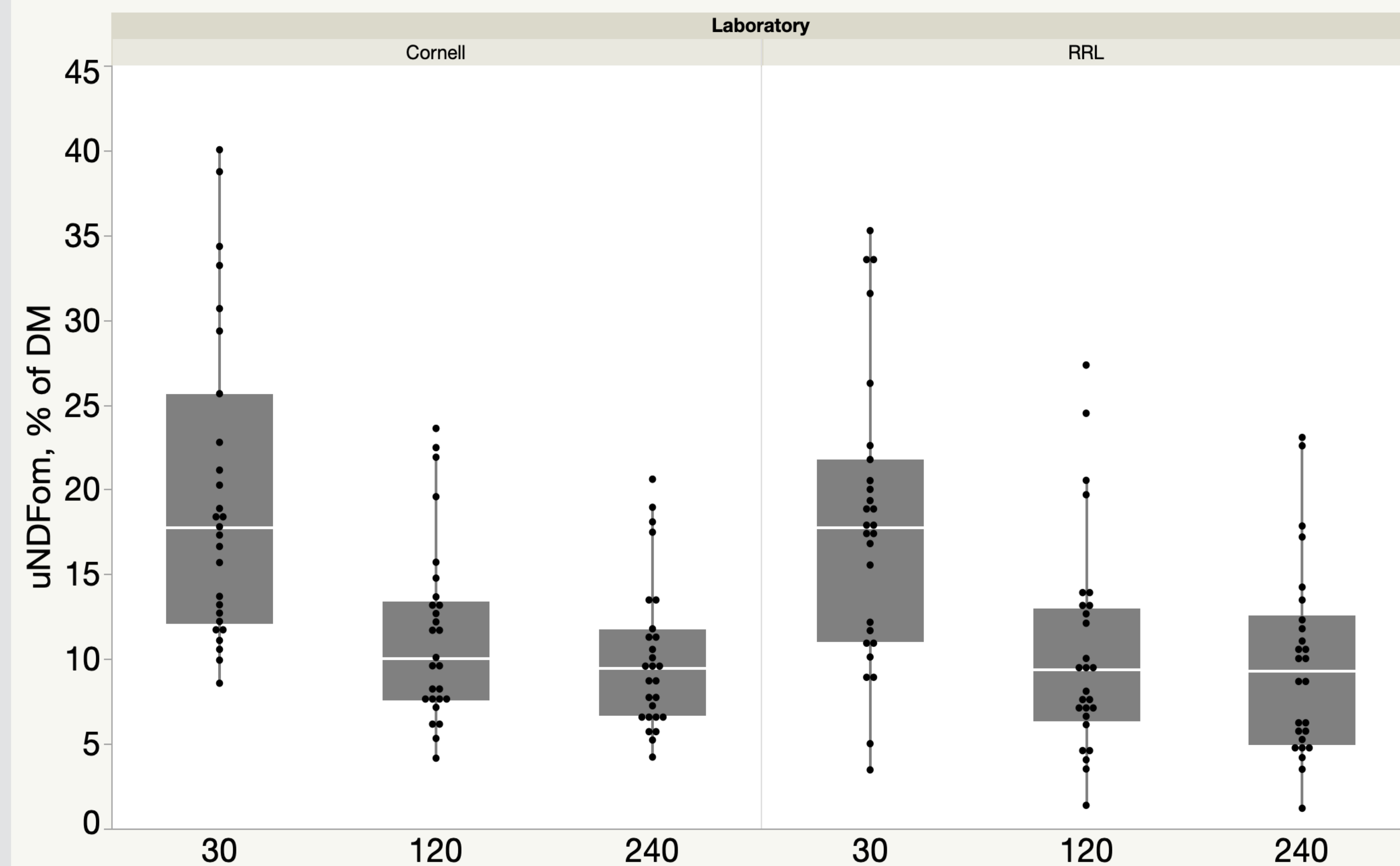
MATERIALS AND METHODS:

European grass silages were selected based upon NIR predictions, to range in both aNDF and uNDFom. Samples (n=27) were microwave oven dried at low intensity to approximately 90% dry matter, ground (1mm), mixed and then split for analysis at both Rock River Laboratory (RRL) and Cornell University (CU) forage analysis laboratory. Both respective laboratories digested samples using *in vitro* rumen techniques for 30, 120, and 240 hour time points. Cornell University and Rock River Laboratory utilized the Raffrenato et al. (2018) technique, with RRL also employing the following modifications: processing and standardizing rumen fluid inoculum as described by Goeser and Combs (2009) and then re-inoculating samples to be digested for 240h time period (at 120h), and using an Ahlstrom 141 glass fiber filter (3.1- μ m pore size) to filter samples post digestion and refluxing. Samples were digested in triplicate, within *in vitro* run, at each time point, and in 2 runs at CU and 1 run at RRL. The single run was chosen at RRL to mirror commercial feed analysis procedures.

Table 1: Main fixed effect least square means for laboratory or *in vitro* rumen incubation time point. *Means with differing connecting letters differ at P < 0.0001.

MAIN EFFECT	UNDFOM, % OF DM	STD. ERROR	CONNECTING LETTERS
Cornell University	13.76	0.769	NS
Rock River Laboratory	12.99	0.769	NS
30 hour	18.90	0.941	A
120 hour	10.97	0.941	B
240 hour	10.26	0.941	B

Figure 1: *in vitro* rumen undigested aNDFom, % of DM, box plots for European grass silages analyzed by two laboratories, Cornell University and Rock River Laboratory.



KEYWORDS:

Undigestible Fiber, Laboratory Analysis

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STATISTICAL ANALYSIS:

Following sample analyses, uNDFom was related to laboratory, time point, and the interaction using the Fit Model function in JMP v14.0. Laboratory and time point were treated as fixed effects. The interaction was not significant and was removed. Residuals were assessed visually for normality, no trends were apparent.

RESULTS AND DISCUSSION:

Following linear modeling, uNDFom was related to time (P<0.0001) but not laboratory (P > 0.4). Model adjusted R-square was 0.23 and RMSE was 6.9. The least square means (lsmean) for European grass silage uNDFom at each digestion time point (% of DM) were 18.9, 11.0, and 10.3 for 30, 120 and 240h, respectively, across both labs. The lsmean uNDFom for each laboratory were 13.8 and 13.0 for CU and RRL, respectively. The lsmeans are also presented in Table 1. Raw data box plots are presented in Figure 1 to view population statistic results for each laboratory.

CONCLUSIONS:

Based on these observations for a select group of European grass silages, *in vitro* rumen uNDFom measures at 30, 120 and 240h appear repeatable between two different laboratories. Further research is warranted with additional comparisons, including alternative forage, feed types, and additional laboratories to better understand repeatability for long-term (120 to 240h) incubated samples.

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