

# AOCS/SQT Amino Acid Round Robin Study

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# Phase I Study

08/2009 - 06/2010

Goal of study can be described in two parts:

1. Determine the major differences between the various amino acid (AA) determination methods in terms of accuracy, reproducibility, and repeatability.
  - This will be determined independent of amino acid hydrolysis.
2. Make recommendations and improvements to existing methods through the cumulative data collected from all study participants.

# Phase I Study

08/2009 - 06/2010

## AA testing, Two steps

AOAC method for acid hydrolysis (*AOAC 994.12*)

Instrumental quantitation of hydrolysate (*HPLC, UPLC, LC/MS/MS, etc.*)

## Experimental Design

18 samples for each lab

15 amino acids to report (ug / ml) (*alanine, arginine, aspartic acid, glycine, glutamic acid, histidine, isoleucine, leucine, lysine, phenylalanine, proline, serine, threonine, tyrosine and valine*)

Accuracy (*AA-standard (Sigma), NIST food reference material, animal feed and spiked animal feed samples*)

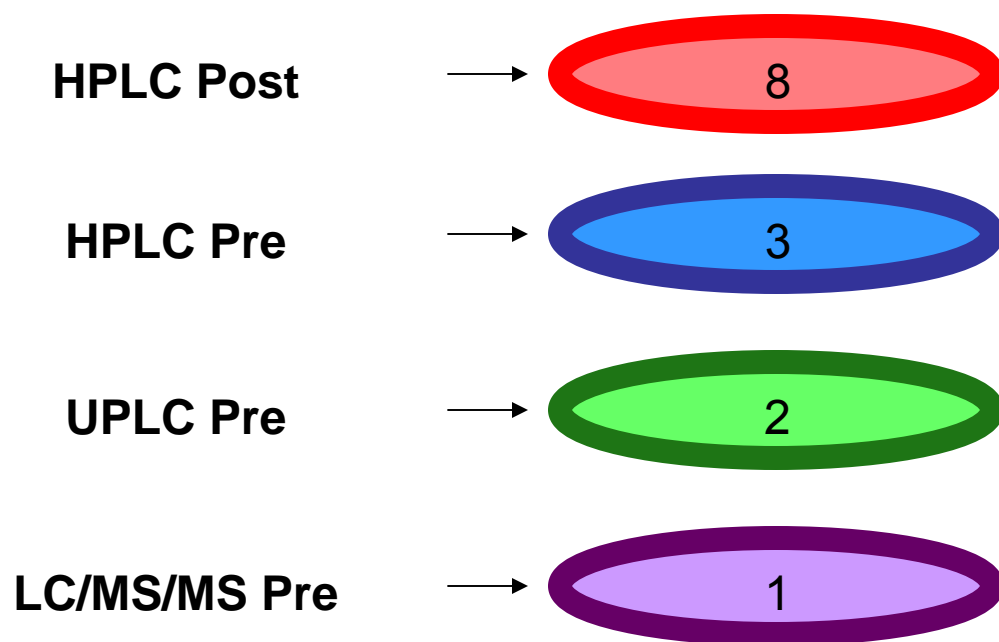
Precision (*within-batch and between-batch*)

## Participants

14 labs

# Instrument and Derivatization Utilized

- Lab A - **HPLC Post-Column**
- Lab B - **HPLC Post-Column**
- Lab C - **UPLC Pre-Column**
- Lab D - **HPLC Post-Column**
- Lab E - **HPLC Post-Column**
- Lab F - **HPLC Pre-Column**
- Lab G - **HPLC Pre-Column**
- Lab H - **HPLC Pre-Column**
- Lab I - **HPLC Post-Column**
- Lab J - **HPLC Post-Column**
- Lab K - **UPLC Pre-Column**
- Lab L - **LC/MS/MS Pre-Column**
- Lab M - **HPLC Post-Column**
- Lab N - **HPLC Post-Column**

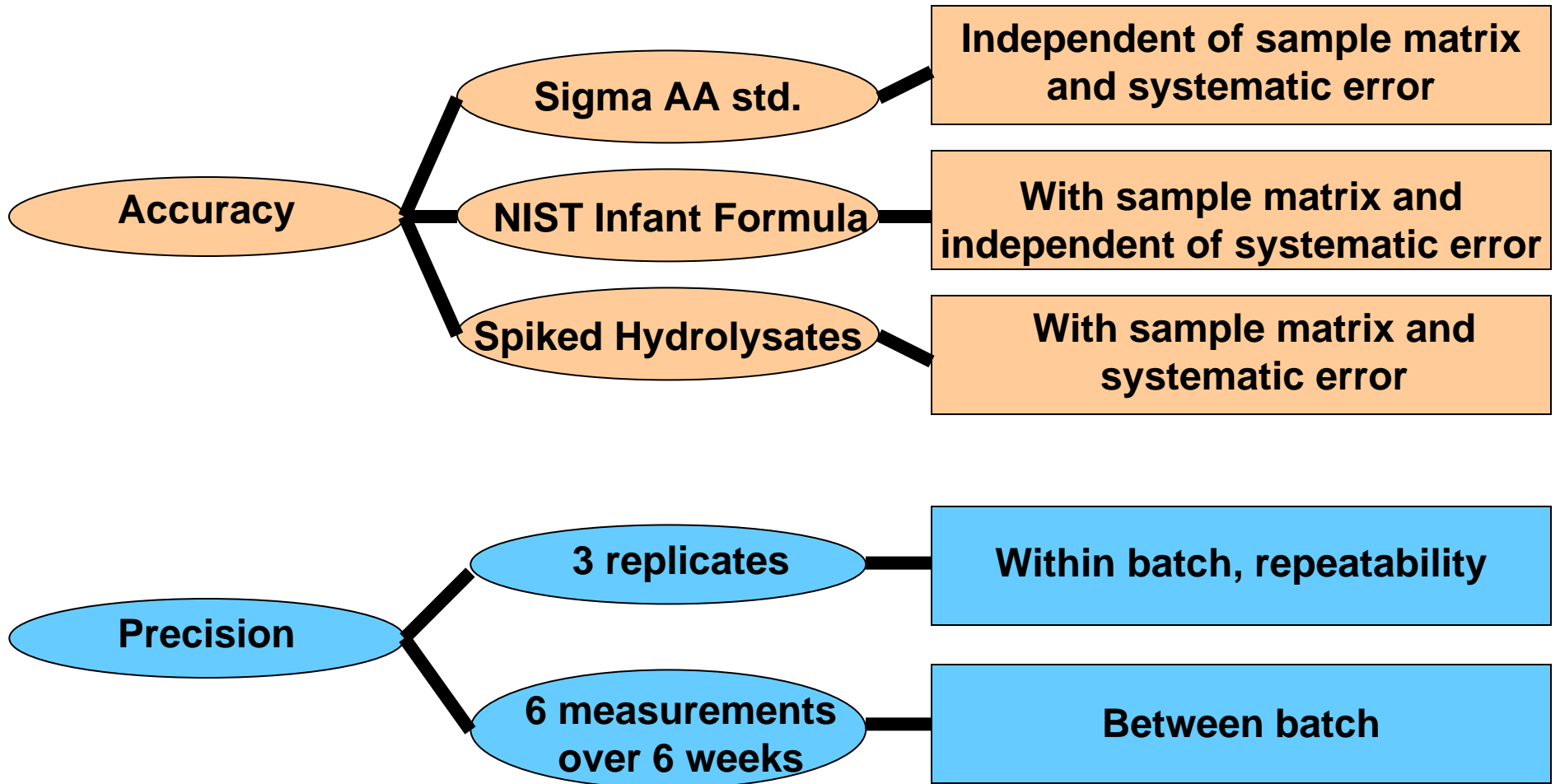


# Sample Preparation

Samples prepared using AOAC method for acid hydrolysis (AOAC 994.12) from:

- Soybean meal
- DDGS provided by the AAFCO Check Sample program
- Swine starter provided by the AAFCO Check Sample program
- Poultry feed provided by the AAFCO Check Sample program
- Infant/adult nutrition formula (NIST SRM 1849)

# AOCS AA Experimental Design





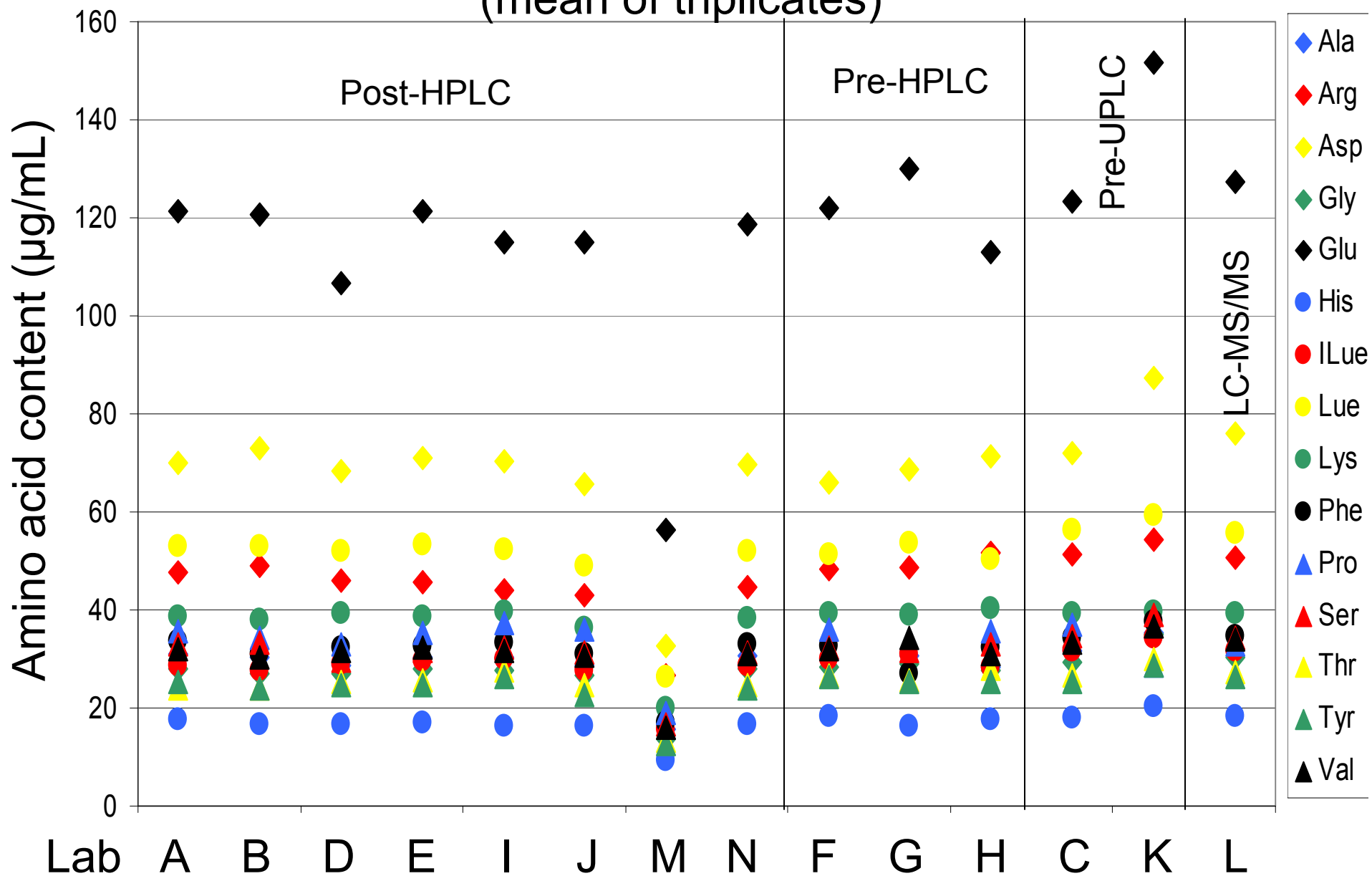






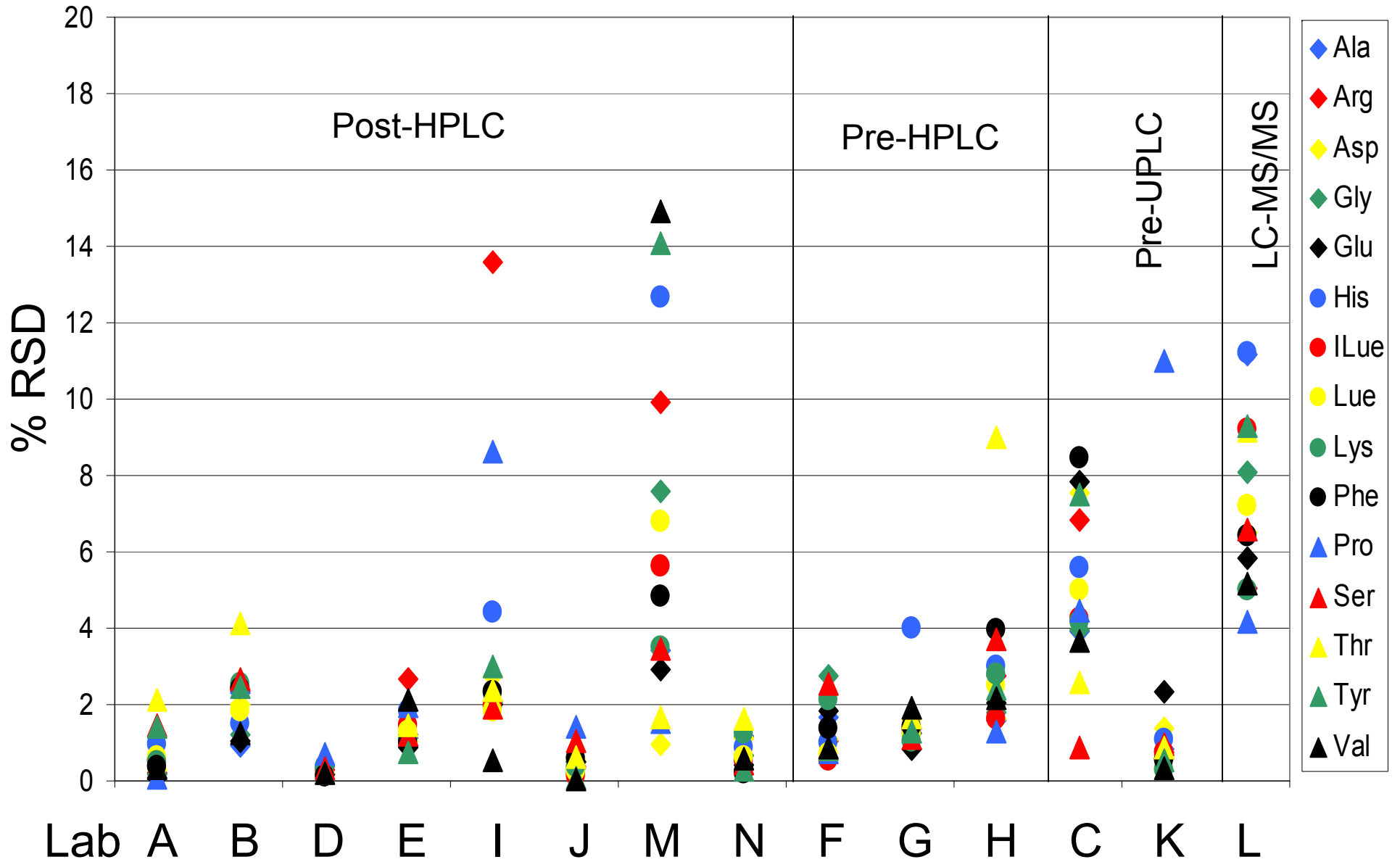
# Comparison of Soybean hydrolysate

(mean of triplicates)



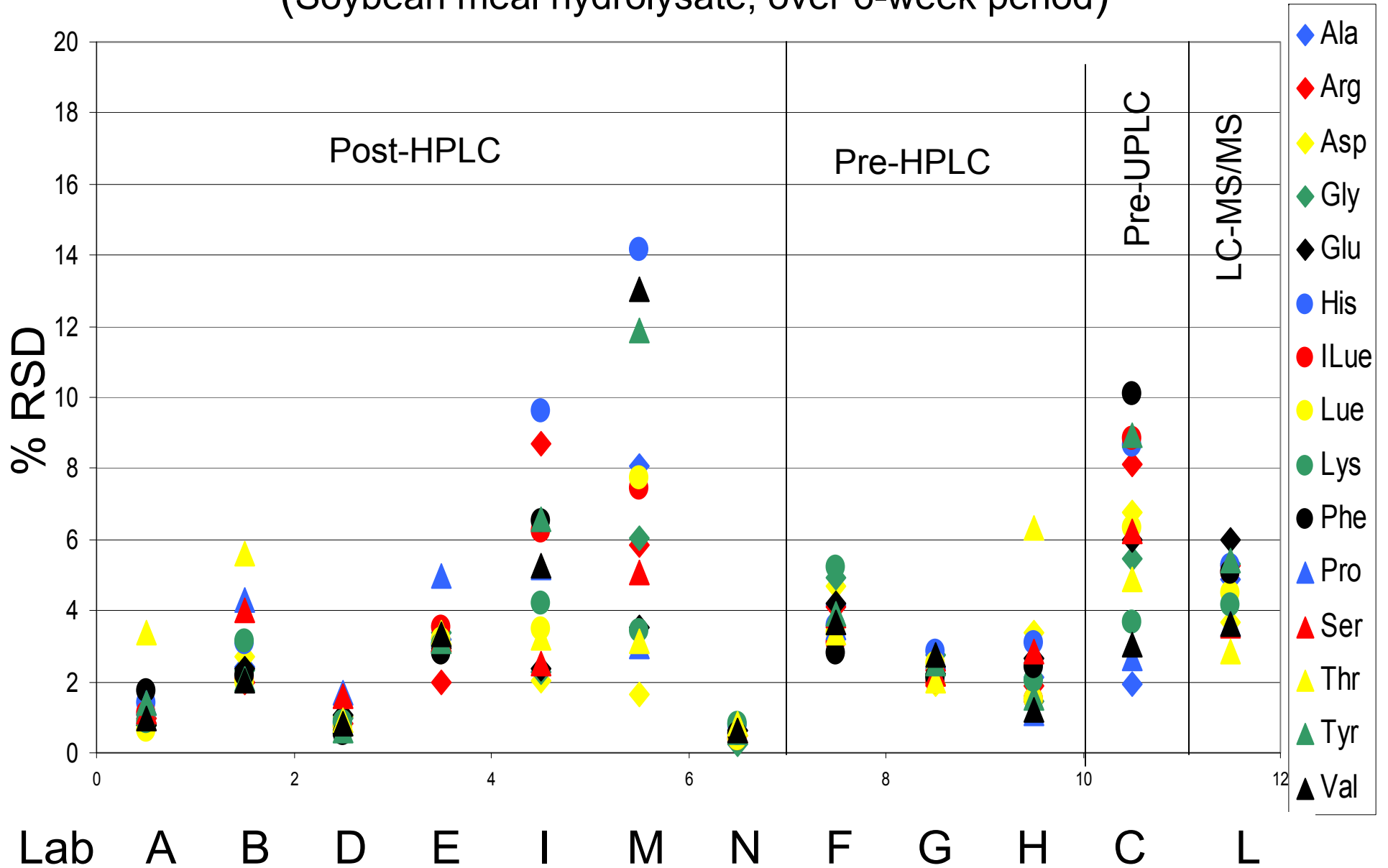
# Precision (RSD, %)

(Based on soybean meal hydrolysate triplicate)



# Precision (RSD, %)

(Soybean meal hydrolysate, over 6-week period)



# Comments

Significance of error in the data:

- Error between labs may be greater than the error between methods
- Masks the error between methods

All labs should include a reference material in every batch of samples, such as NIST SRM

Use of an internal standard should be included in the sample pretreatment phase, after hydrolysis.

# Phase II Study

08/2010 - 03/2011

Goal of study can be described in two parts:

1. Determine the major differences between the various hydrolysis procedures used by each lab participant that affect accuracy and repeatability.
2. Make recommendations and improvements to existing hydrolysis procedures through the cumulative data collected from all study participants.

# Experimental Design

- Each lab participant will receive 5 animal feed samples (including NIST SRM) and hydrolyze each sample once a week for three weeks
- All the hydrolysate samples will be collected and tested for 15 amino acids by one participating lab in the U.S.

People interested in participating should contact

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