# Meta-Analysis of the Effects of Supplemental Rumen-Protected Lysine During the Lactating Period on Performance and Amino Acids Profiles in Dairy Cows


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Pictures by Bonnie Mohr http://www.bonniemohr.com/

# Introduction

- ✓ **Limiting Amino Acids:** Lysine and methionine are frequently the most limiting essential amino acids especially in corn or soybean-based diets of lactating dairy cows
- ✓ **Contribution:** Lysine contributes ~9% in milk protein, making it crucial to balance in dairy diets
- ✓ **Rumen-Protected Lysine (RPL):** Traditional sources of rumen undegradable protein like blood meal have variable lysine content due to processing, making RPL supplementation a viable alternative
- ✓ **Heat Damage to Lysine:** Lysine is highly susceptible to heat damage, reducing its digestibility in cottonseed meal, soybean meal, and whole soybeans
- ✓ **Experimental Variability:** Many studies on RPL supplementation have a limited number of experimental units, leading to variability and inconsistent results. Among all the studies that supplemented RPL in lactating dairy cows, only 30% of studies had 15 or more experimental units per treatment
- ✓ **Nutritional Guidelines:** The NRC (2001) recommended lysine and methionine at 7.2% and 2.4% of metabolizable protein, respectively, but updated estimates from NASEM (2021) suggest reassessing these values based on recent research
- ✓ **Need for Meta-analysis:** Robinson et al.'s 2010 meta-analysis included only 12 treatment means, highlighting the need for an updated analysis to incorporate the past 14 years of research on supplemental RPL and provide more comprehensive insights

Schwab (1995) Biotech. Anim. Feed. Anim. Feed. 115-145; NRC (2001); NASEM (2021)

# Hypotheses

- ✓ Supplementation of rumen-protected lysine (**RPL**) during the lactating period would improve postpartum performance in dairy cows

- ✓ Using mixed models meta-analysis would result in identification of an optimum concentration (%) of lysine in metabolizable protein (**MP**) to improve performance in dairy cows

# Objectives

- ✓ Determine the impact of supplemental lysine fed as RPL during the postpartum period on:
  - ✓ Productive performance
  - ✓ Blood amino acids profile

- ✓ Identify the optimum concentration of lysine as MP supplemented to lactating cows

- ✓ Determine if responses to metabolizable lysine as % of MP (**LYSMP**) depend on postpartum supplies of metabolizable methionine as % of MP (**METMP**)

# Inclusion Criteria

- ✓ Randomized experiments (RCBD or CRD) in which experimental unit (cow or pen) was clearly defined and supplementation of RPL started postpartum
- ✓ Experiments containing
  - ✓ Control, no supplemental RPL
  - ✓ Treated, one or more levels of lysine supplemented as RPL
- ✓ Concentration and intestinal digestibility of lysine in RPL source had to be reported, so the amount of metabolizable lysine could be calculated
- ✓ RPL as a component of TMR or top-dressed
- ✓ The ingredient composition of postpartum diets fed had to be reported
- ✓ Experiments had to report least squares means (LSM) and measure of error of individual treatments for the responses of interest

# Literature Search

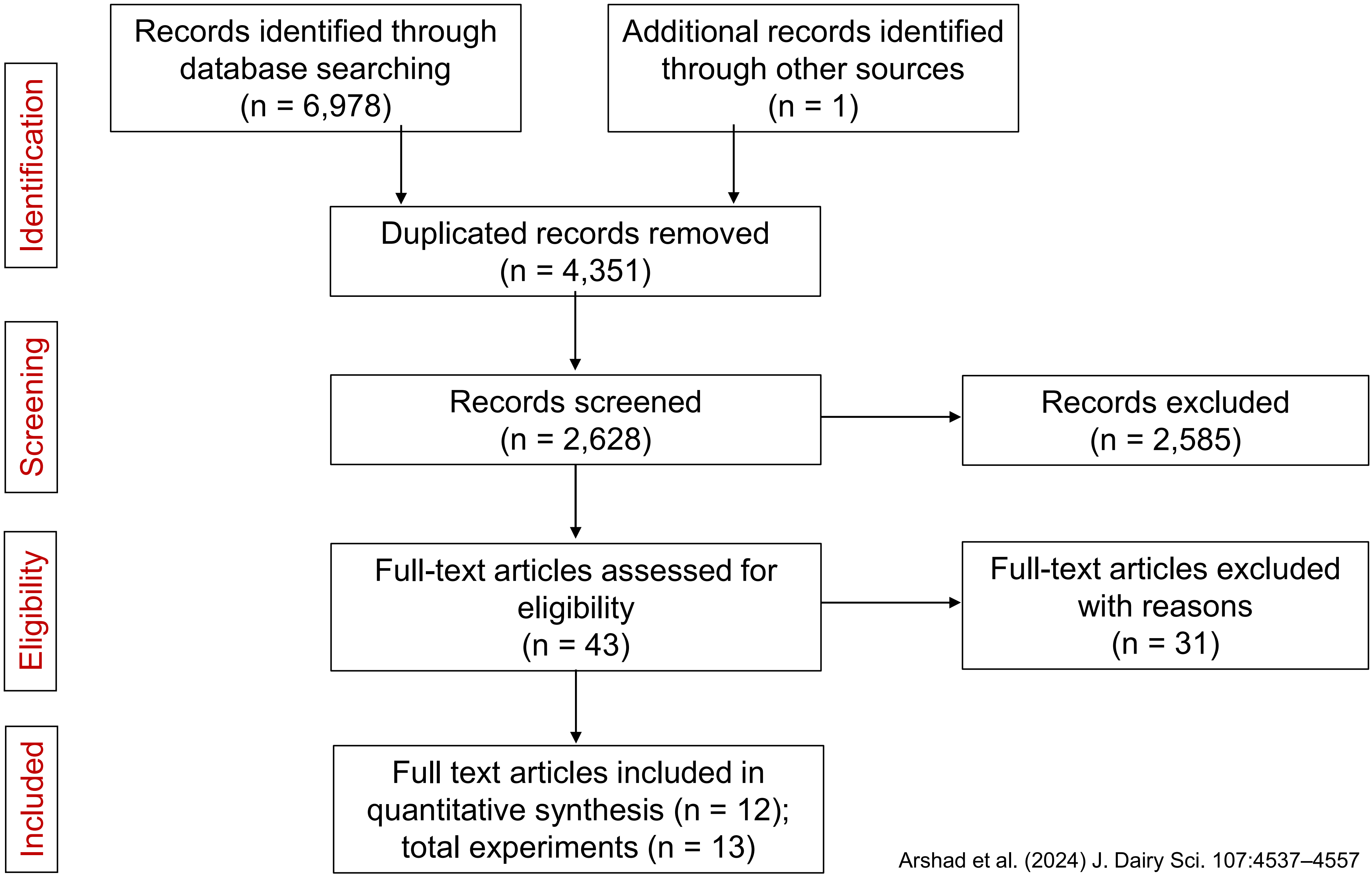
- ✓ Database searched
  - ✓ PubMed (US National Library of Medicine National Institutes of Health)
  - ✓ ISI Web of Science
  - ✓ Abstracts book of Journal of Animal Science
  - ✓ Abstracts book of Journal of Dairy Science

- ✓ Keywords
  - ✓ Lysine dairy cow
  - ✓ Lysine transition cow
  - ✓ Rumen-protected lysine cow
  - ✓ Rumen-protected lysine transition cow

The term *'transition'* was included in the key terms to capture studies applying factorial arrangements during the transition period. In this meta-analysis, we compared treatment groups with similar prepartum interventions that differed postpartum (with or without supplementation of RPL)

- ✓ Additional requests were made to individual authors of manuscripts to identify data that might have been collected, but not reported in the published paper

# Prisma Flow Diagram

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# Meta-Analysis of Published Literature

- ✓ 12 manuscripts with 13 randomized experiments
  - ✓ 40 treatment means and 594 parous lactating cows

## Data collected

- ✓ **Predictors**
  - ✓ Breed (Holstein or other)
  - ✓ Design of the experiment
  - ✓ Ingredient and nutrient composition of postpartum diets
  - ✓ Rumen-protected lysine product fed
  - ✓ Rumen-protected methionine product fed
  - ✓ Amount of metabolizable lysine and methionine fed
  - ✓ Duration of feeding RPL in the postpartum period
- ✓ **Responses of interest:** LSM and measures of error (SED/SD/SEM)
  - ✓ DM intake
  - ✓ BW and BW change
  - ✓ Productive performance
  - ✓ Feed efficiency
  - ✓ Concentrations of amino acids in blood

# Mathematical Approaches

- ✓ Classic mixed effects meta-analysis and meta-regression to determine the effect of supplementing RPL postpartum
  - ✓ METAN and METAREG procedures of STATA release 14
  - ✓ Weighted according to SD and number of experimental units to account for the precision of each experiment

- ✓ **RPL as a categorical variable:**
  - ✓ **Fixed effects:** RPL (not supplemented vs. supplemented), and METMP

- ✓ Mixed effects meta-analysis to determine the optimum amount of LYSMP fed
  - ✓ MIXED procedures of SAS ver. 9.4
  - ✓ Weighted by the inverse of SEM squared ($1/SEM^2$) to account for the precision of each experiment

- ✓ **LYSMP as a continuous variable:**
  - ✓ **Fixed effects:** LYSMP, LYSMP x LYSMP, METMP, and LYSMP x METMP

- ✓ All models included the random effect of experiment

## Descriptive statistics of nutrient intake and estimated duodenal flow based on NASEM (2021)[1]

| Item | Control (n = 17) | | | RPL (n = 23) | | |
|---|---|---|---|---|---|---|
| | Mean ± SD | Minimum | Maximum | Mean ± SD | Minimum | Maximum |
| Content, DM basis | | | | | | |
| $NE_L$, Mcal/kg | 1.71 ± 0.07 | 1.60 | 1.80 | 1.72 ± 0.06 | 1.60 | 1.80 |
| CP, % | 16.5 ± 1.2 | 14.3 | 18.3 | 16.7 ± 1.2 | 14.5 | 18.5 |
| MP, % | 9.74 ± 0.78 | 8.20 | 11.7 | 9.86 ± 0.80 | 8.23 | 11.7 |
| Intake | | | | | | |
| DMI, kg/d | 21.6 ± 3.6 | 16.3 | 27.7 | 22.3 ± 3.2 | 16.3 | 28.1 |
| $NE_L$, Mcal/d | 35.5 ± 5.6 | 28.5 | 46.6 | 37.0 ± 5.6 | 28.2 | 46.1 |
| CP, g/d | 3,411 ± 474 | 2,624 | 4,147 | 3,576 ± 479 | 2,836 | 4,320 |
| Metabolizable, g/d | | | | | | |
| Protein, g/d | 2,008 ± 250 | 1,649 | 2,452 | 2,111 ± 259 | 1,646 | 2,529 |
| Lysine as RPL,[2] g/d | 0 | --- | --- | 19.3 ± 10.3 | 5.10 | 40.6 |
| Lysine, g/d | 140 ± 18.8 | 106 | 167 | 160 ± 22 | 120 | 195 |
| LYSMP,[3] %MP | 7.02 ± 0.87 | 4.66 | 7.86 | 7.64 ± 0.96 | 5.10 | 9.25 |
| METMP,[4] % MP | 2.44 ± 0.27 | 1.89 | 2.85 | 2.41 ± 0.28 | 1.82 | 2.91 |

[1]Predicted contents of nutrients in the diets offered and intake of nutrients were calculated using the ingredient composition of each diet with NASEM Dairy 8 (NASEM (2021) and adjusted to the observed postpartum DMI.

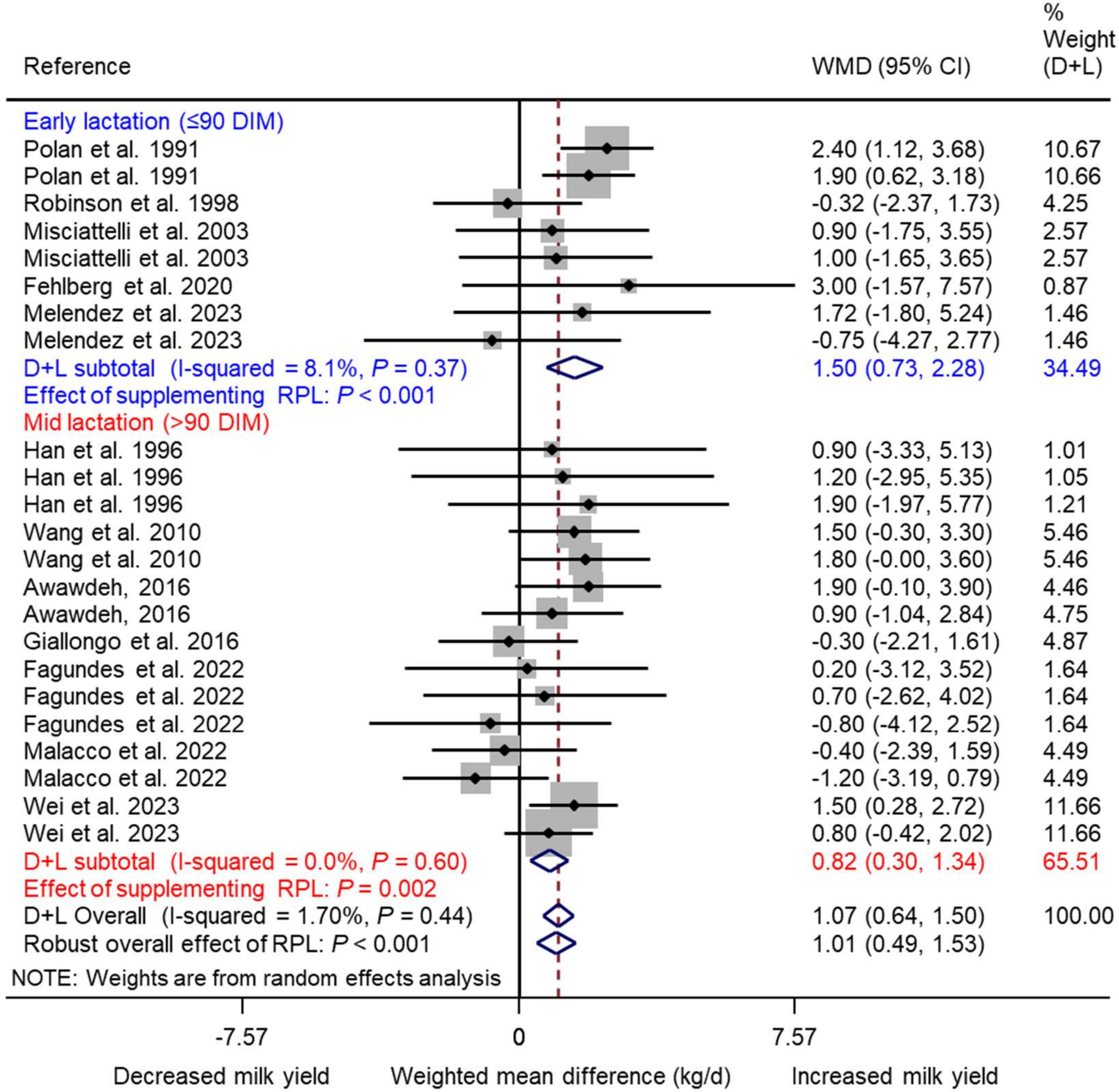
[2]Lys as RPL = intake of metabolizable Lys only from RPL product fed as top-dress or incorporated into the TMR. Intake of metabolizable Lys was calculated based on the concentration of Lys and intestinal digestibility of each RPL product reported in the respective papers.

[3]LYSMP = metabolizable lysine as % of MP calculated using NASEM Dairy 8 (NASEM (2021) according to dietary ingredients and DMI of each treatment mean.

[4]METMP = metabolizable methionine as % of MP calculated using NASEM Dairy 8 (NASEM (2021) according to dietary ingredients and DMI of each treatment mean.

Arshad et al. (2024) J. Dairy Sci. 107:4537–4557

# Effect of RPL on Milk Yield: Lactation Stage

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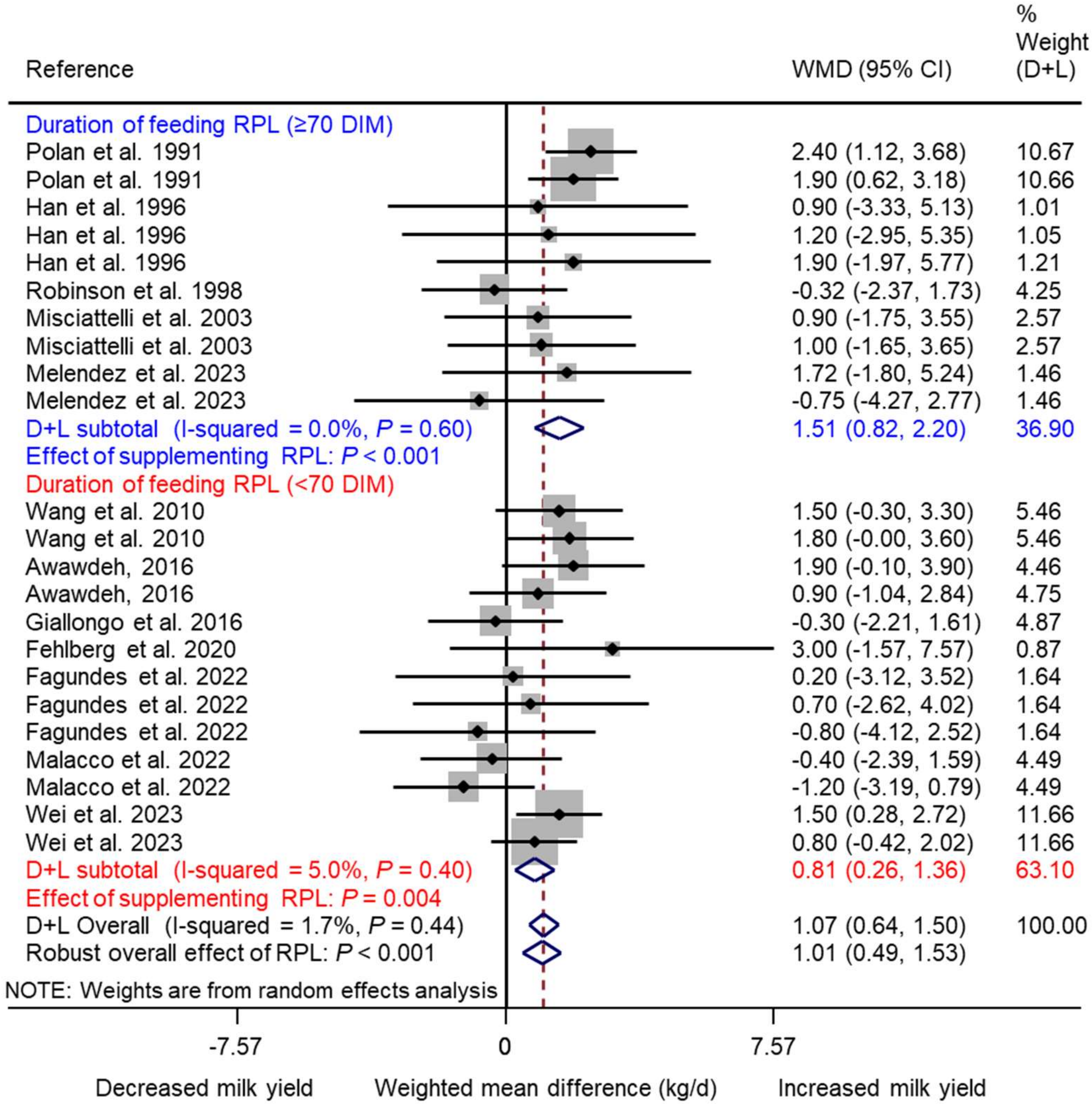
# Effect of RPL on Milk Yield: Feeding Duration

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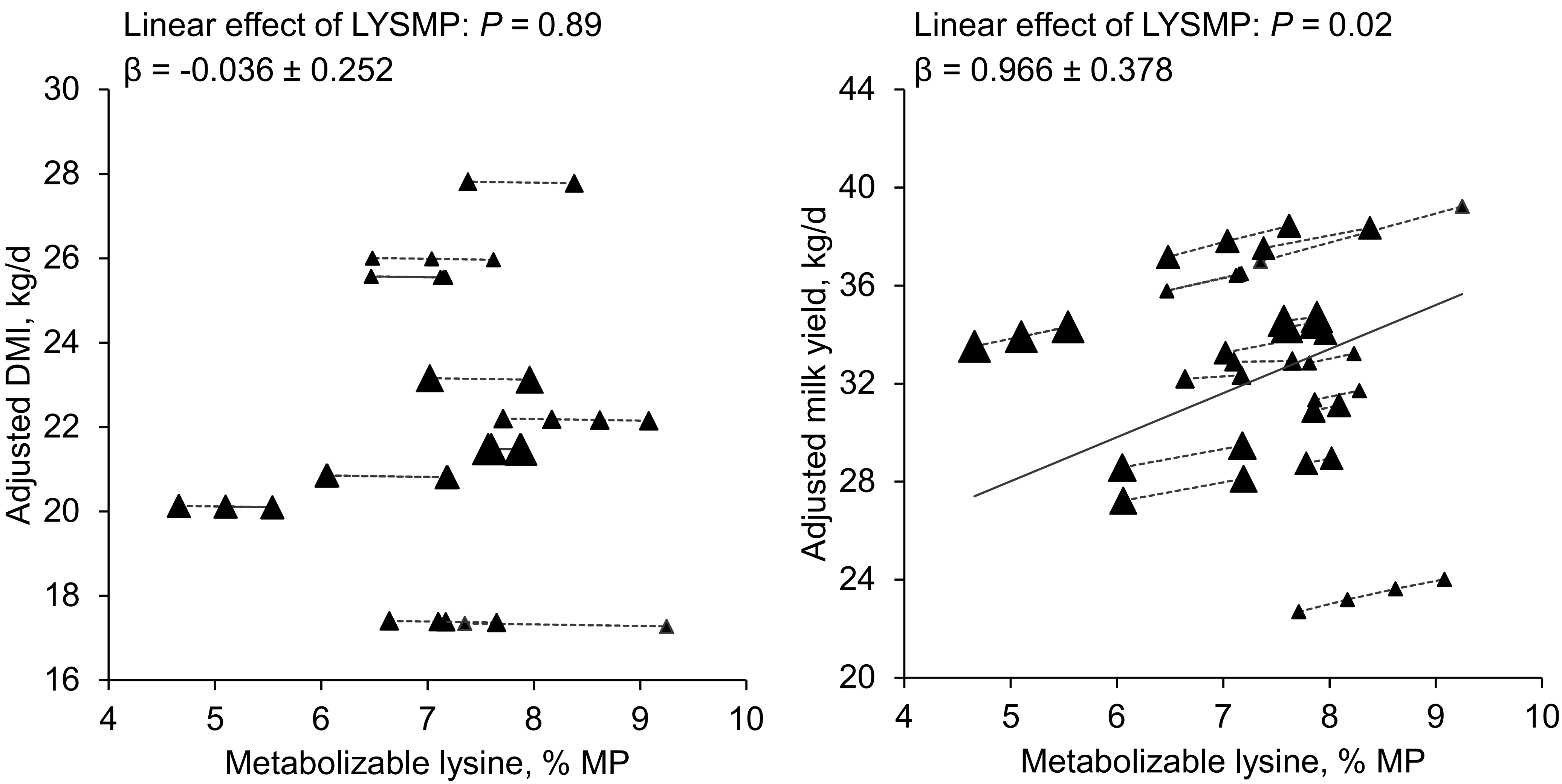
# DM Intake and Milk Yield

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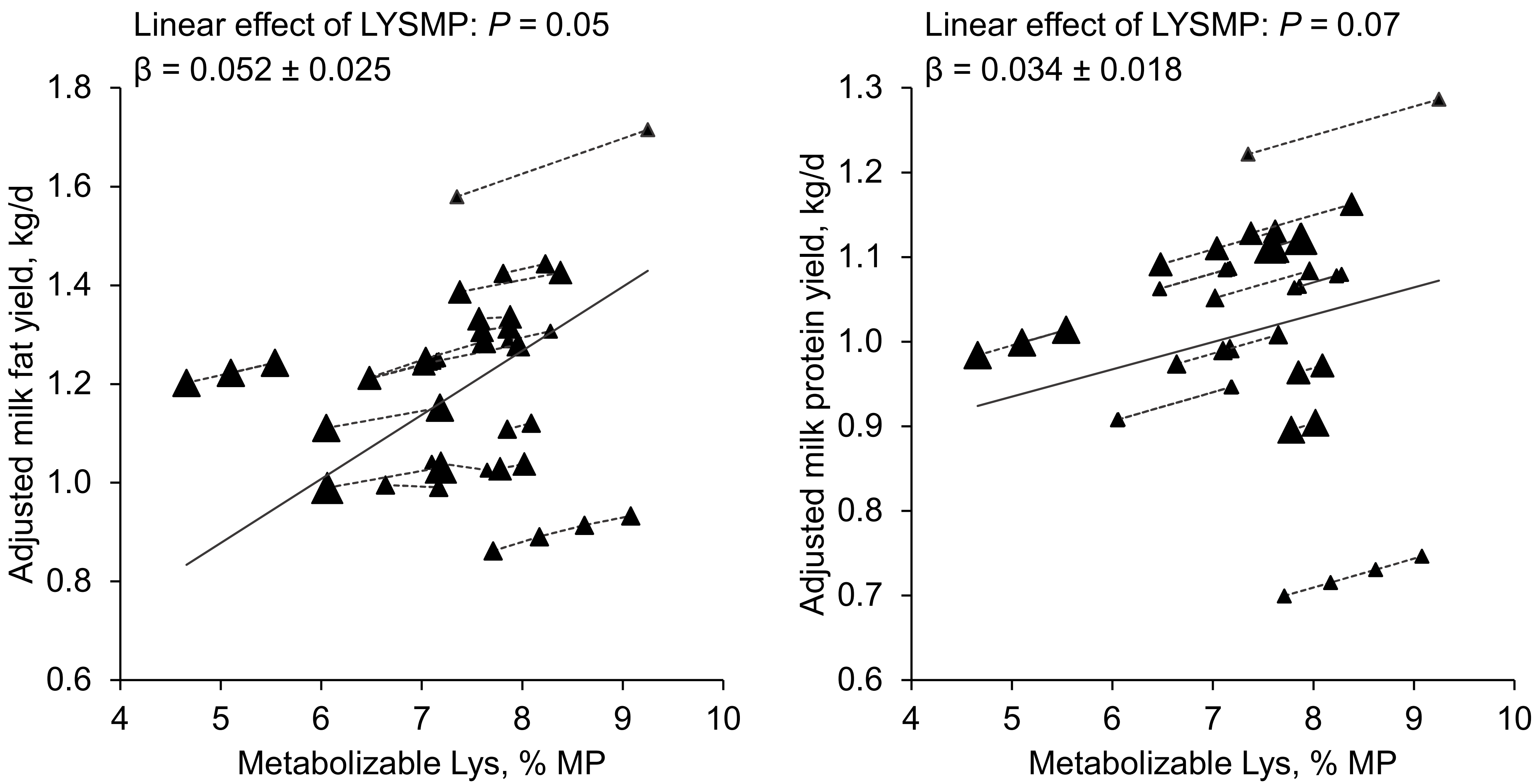
# Yields of Fat and Protein in Milk

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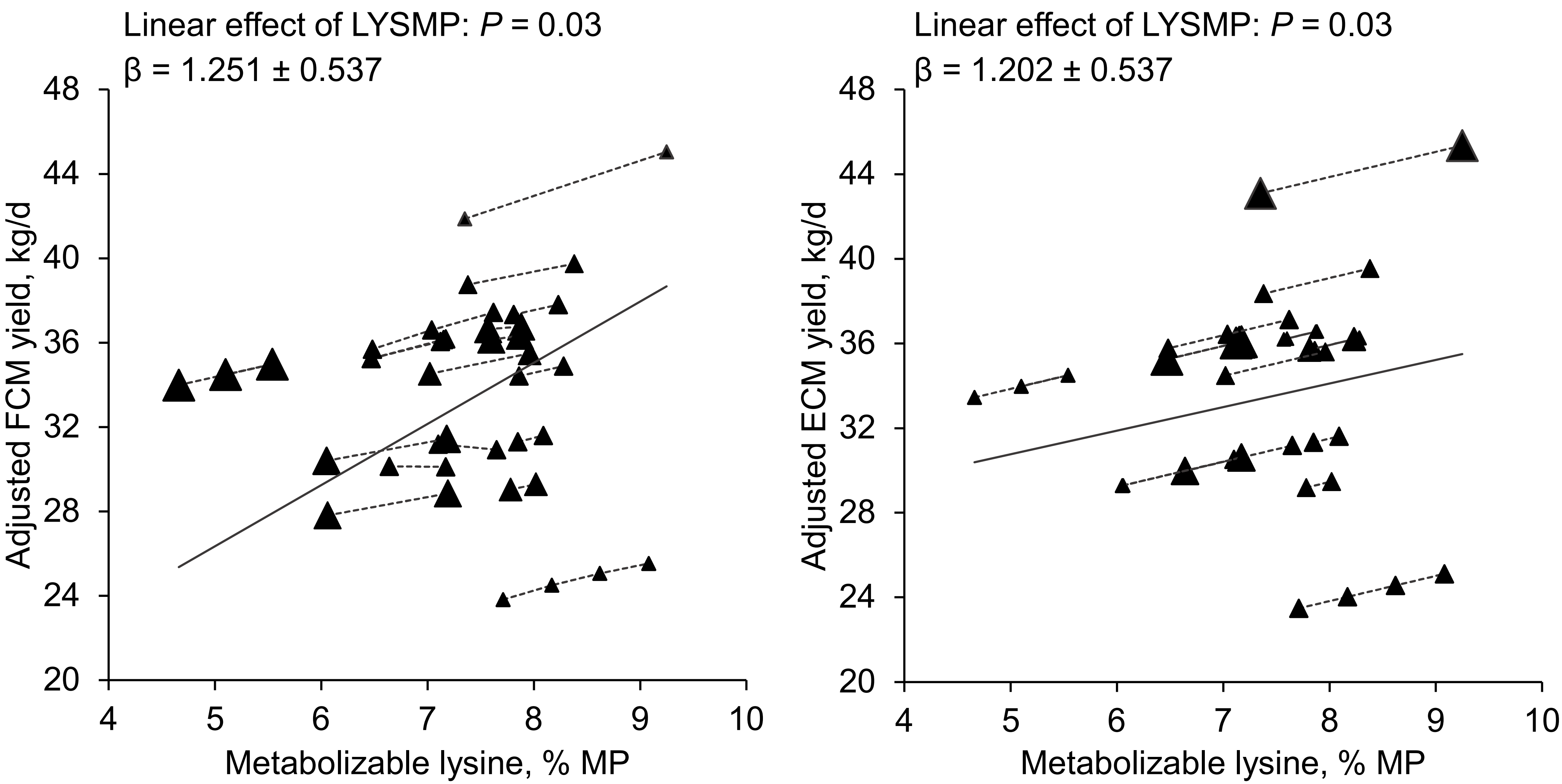
# Yields of Fat-Corrected and Energy-Corrected Milk

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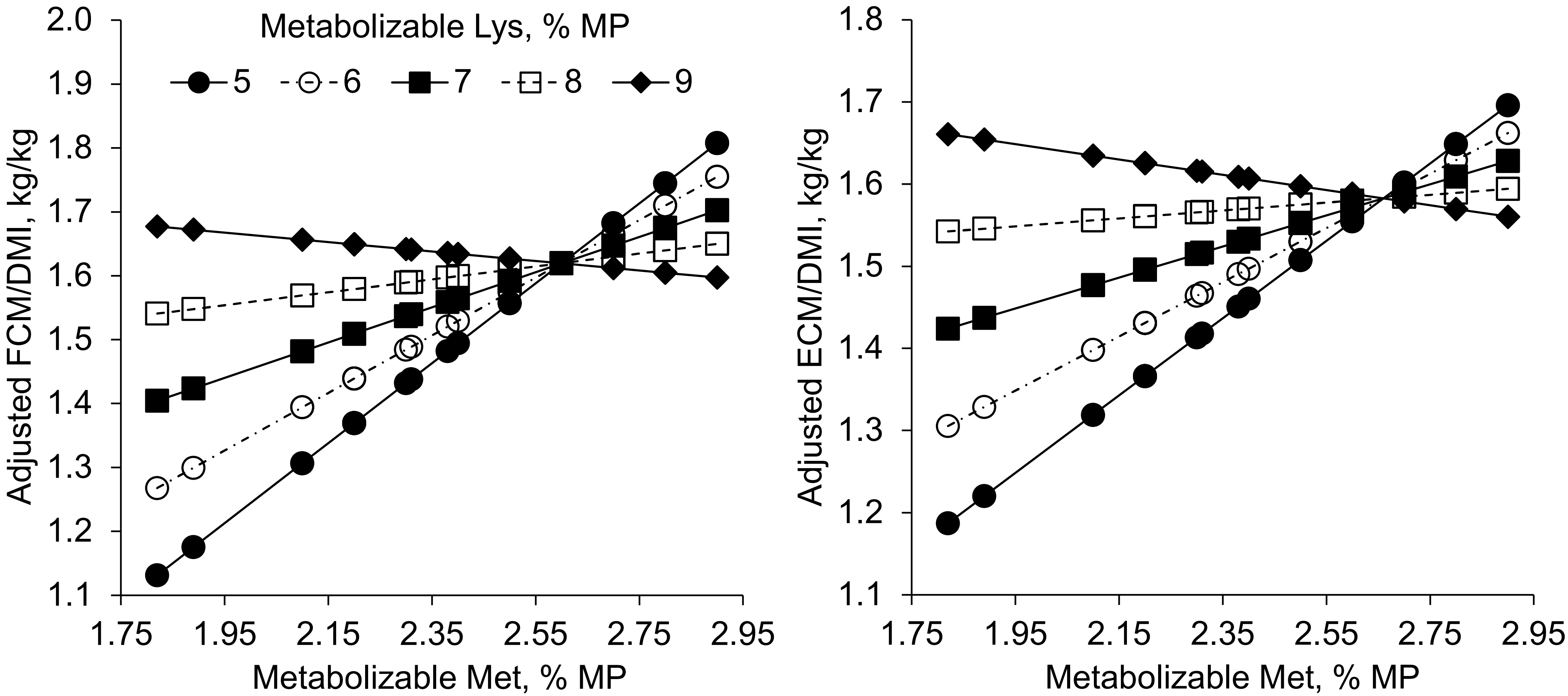
# Interactions Between Metabolizable Lysine and Postpartum Dietary Methionine

LYSMP of 7.40 %; METMP of 1.82 % = 1.46 kg/kg

LYSMP of 7.40 %; METMP of 2.91 % = 1.68 kg/kg

LYSMP of 7.40 %; METMP of 1.82 % = 1.47 kg/kg

LYSMP of 7.40 %; METMP of 2.91 % = 1.62 kg/kg




Arshad et al. (2024) J. Dairy Sci. 107:4537–4557

**Estimated LSM and respective SEM for concentrations of blood concentrations of AA when lactating cows are fed postpartum diets differing in metabolizable Lys as percentage of MP (6.5% or 8.5%)[1]**

| | | LYSMP,[2] % MP | | *P*-value[3] | |
|---|---|---|---|---|---|
| Item, μ*M* | Means (Exp.),[4] n | 6.5 | 8.5 | Linear | Quadratic |
| Arginine | 22 (07) | 62.9 ± 4.9 | 68.8 ± 5.1 | 0.03 | 0.06 |
| Histidine | 22 (07) | 69.2 ± 21.8 | 59.9 ± 22.2 | 0.17 | --- |
| Lysine | 22 (07) | 64.1 ± 4.7 | 80.7 ± 6.1 | 0.02 | --- |
| Methionine | 22 (07) | 34.3 ± 3.8 | 29.7 ± 4.1 | 0.03 | --- |
| Phenylalanine | 22 (07) | 108 ± 18 | 103 ± 19 | 0.46 | --- |
| Total essential AA* | 22 (07) | 871 ± 56 | 854 ± 56 | 0.02 | --- |
| Non-essential AA | | | | | |
| Alanine | 19 (06) | 239 ± 16 | 223 ± 16 | 0.01 | --- |
| Glycine | 19 (06) | 291 ± 37 | 290 ± 37 | 0.93 | --- |
| Serine | 19 (06) | 77.4 ± 15.3 | 75.2 ± 15.3 | 0.45 | --- |

[1]Results from the final multivariable models when lactating cows are fed diets differing in the content of LYSMP (6.5% or 8.5%).
[2]LYSMP = metabolizable Lys as percentage of MP calculated using NASEM Dairy 8 (NASEM (2021) according to dietary ingredients and DMI of each treatment mean.
[3]Linear = linear effect of LYSMP; quadratic = quadratic effect of LYSMP. Cells with dashes indicate that the final multivariable model did not include the quadratic effect of LYSMP because *P* > 0.10.
*Represents an interaction (P ≤ 0.04) between LYSMP and METMP.
[4] Represents the number of treatment means (and experiments) contributed data in the statistical analysis.

Arshad et al. (2024) J. Dairy Sci. 107:4537–4557

# Conclusions

- ✓ Supplementing lysine as RPL during the postpartum period
  - ✓ Increased the blood concentrations of lysine
  - ✓ When fed in early lactation (≤90 DIM) or for an extended duration (≥70 DIM) resulted in 1.51 kg/d more milk compared to non-supplemental cows
- ✓ Increasing LYSMP from 6.5% to 8.5%
  - ✓ Linearly increased yields of milk, FCM, ECM, and milk fat by 1.8, 2.5, 2.4, and 0.10 kg/d, respectively, without a concurrent increase in DMI.
  - ✓ Feed efficiency response was influenced by postpartum metabolizable methionine
- ✓ The optimum amount of supplemental RPL was not identified because the increments in productive performance were maximized up to 9.25% of LYSMP in multiparous postpartum cows

# Limitations or Future Directions

- ✓ Data analyzed included only parous cows
  - ✓ Nulliparous represent 30 to 35% of the transition cows on a farm
  - ✓ We really do not know the response of nulliparous to supplemental RPL
- ✓ Information on effects of supplementation of RPL on health events is not available in the literature. Preliminary work from scientific community has shown potential benefits of RPL on immune functions in dairy cows
- ✓ Large dose-titration experiments are needed to understand the impact of LYSMP beyond 8.5 or 9.25 % in the diets of lactating cows

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# Questions/Comments


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