# Nutritional Regulation of Milk Protein Synthesis

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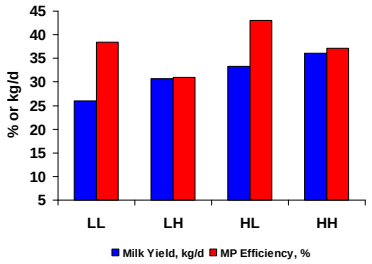
# Effects of Energy and MP Supply on Production and Protein Efficiency



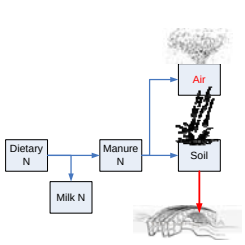

**Milk**
Protein *P*<.08
Energy *P*<.01
E x P *P*=.64

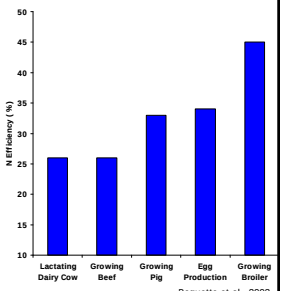
**Efficiency**
Energy *P*<.001
Protein *P*<.001
E x P *P*=.53

HH = 1.54 MCal/kg, 11.8% MP
HL = 1.54 MCal/kg, 9.5% MP
LH = 1.45 MCal/kg, 11.8% MP
LL = 1.45 MCal/kg, 9.5% MP

Rius et al., 2010, JDS

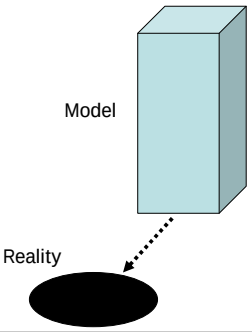
# N Conversion Efficiencies and Environmental Loading



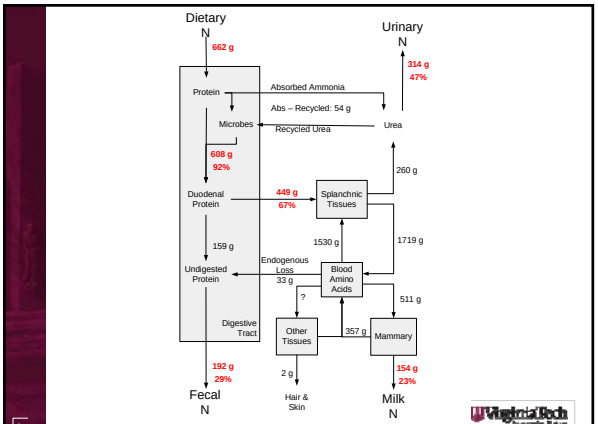




Bequette et al., 2003

# Is Our Bias Preventing Progress?

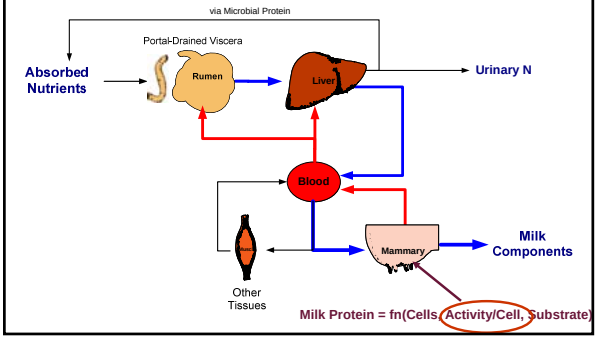



- Pigs can convert 85%of absorbed N to tissue when fed a perfect AA mix (Baker, 1996)
- Hypothesis: Inadequate knowledge of nutrient requirements in ruminants is the cause of poor efficiency.

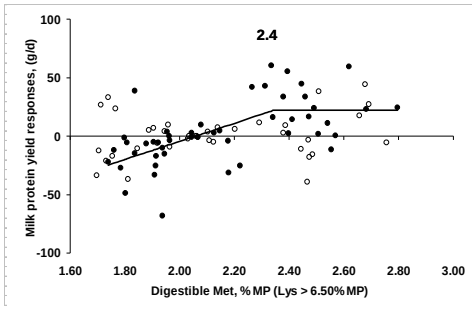



Adapted from Ipharraguerre et al., 2005

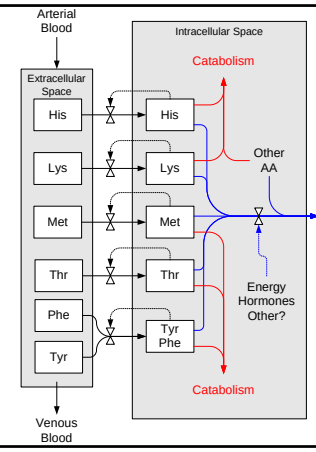
# Postabsorptive Delivery of AA to Mammary Tissue



## Milk Protein Responses to Varying Digestible Methionine




NRC, 2001

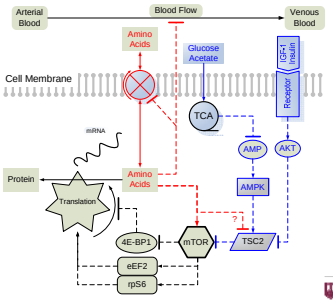



**Requirement Accuracy:**
Single Limiting Nutrient Concept is Inappropriate

- Single-limiting AA
  - 48% of Lit variation
  - 0% of variation when single EAA infused
- Multi-limiting AA
  - **64%** of Lit variation
  - ~50% of variation when single EAA infused
  - **Remaining error correlated with energy supply**

Hanigan et al., 2000

## Cell Signaling and mRNA Translation

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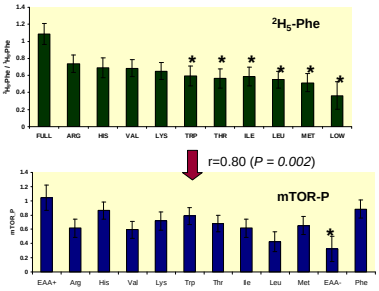
## Effects of Individual Amino Acid Deficiencies on Lactogenic Mammary Tissue Slices

**$^2$H-Phe Incorporation into casein**




**Deficiencies of multiple AA affect casein synthesis**

**Positive correlation between $^2H_5$-Phe and phospho-mTOR**

Appuhamy et al., 2009

* Differs (P<0.05) from +EAA

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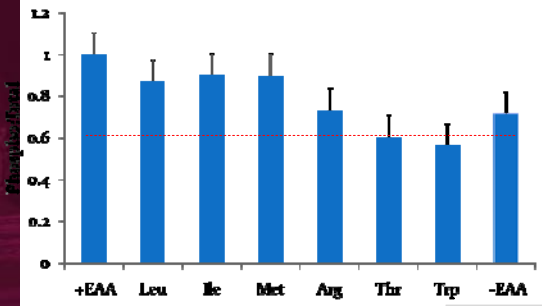
## Supplementation of EAA on Phosphorylation of mTOR




Appuhamy et al., 2009

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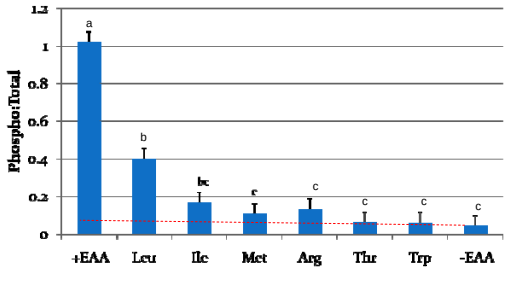
## Supplementation of EAA on Phosphorylation of rpS6




Appuhamy et al., 2009

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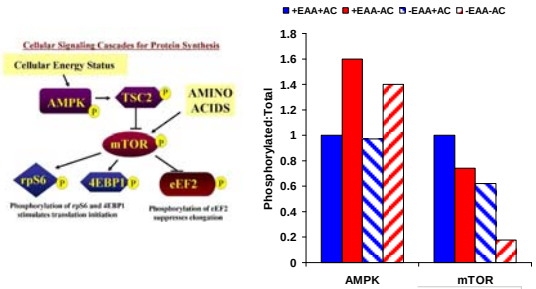
## Effects of Acetate and Essential Amino Acids on Mammary Cell Signaling In Vitro




Appuhamy et al., 2009

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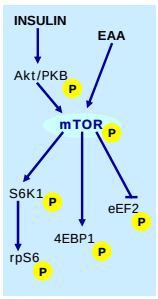
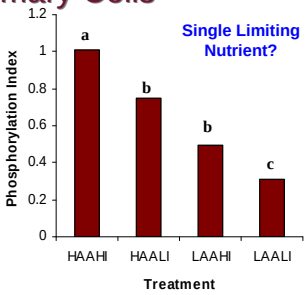
## Effects of Insulin and EAA on mTOR in Mammary Cells




Appuhamy et al., 2011

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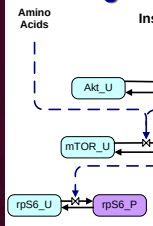
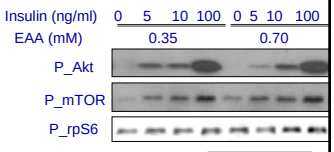
## Cell Signaling Model Parameterization




**Data**

- MAC-T cells
- 4 levels of EAA: 0, 0.35, 0.70, 3.50 mM
- 4 levels of insulin: 0, 5, 10, 100 ng/mL




Appuhamy et al., 2010

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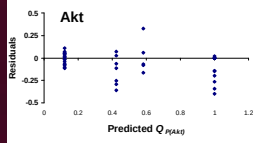
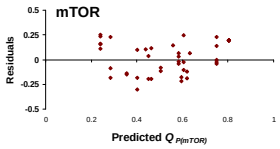
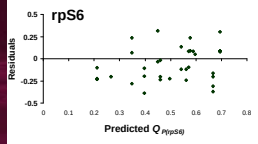
## Cell Signaling Model Results




| Protein | RMSPE | Mean bias | Slope bias | Dispersion |
|---|---|---|---|---|
| | | % | | |
| Akt | 35.3 | 16.9 | 7.52 | 75.6 |
| mTOR | 30.2 | 9.80 | 0.06 | 90.1 |
| rpS6 | 30.0 | 10.5 | 9.20 | 80.3 |

Appuhamy et al., 2010

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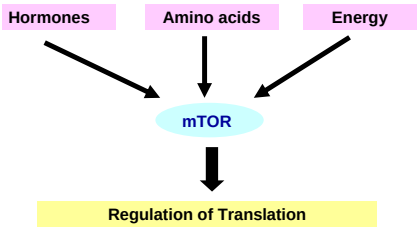
## Regulation of mRNA Translation




**mTOR: Mammalian target of rapamycin**

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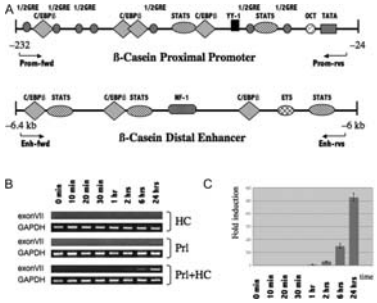
## Nutritional Regulation of Milk Protein Gene Expression?




- GRE – glucocorticoid
- C/EBP $\beta$ - **GH**, IL-6
- STAT5 – Prolactin, **GH**, Cytokines
- ETS – **Insulin, GH**
- NF-1 – TNF, IL-1

from Kabotyanski et al., 2006

Derived from www.genego.com, Thomson Reuters

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## Insulin Responses

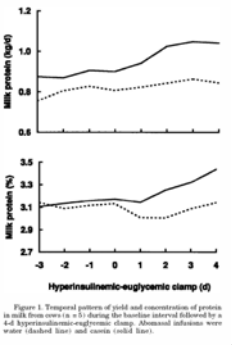
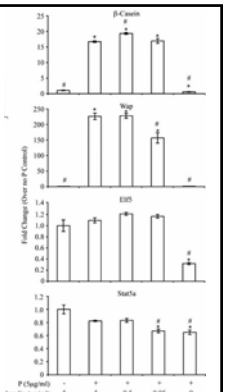



Figure 1. Temporal pattern of yield and concentration of protein in milk from cows (n = 5) during the baseline interval followed by a 4-d hyperinsulinemic-euglycemic clamp. Abomasal infusions were water (dashed line) and casein (solid line).

from Griinari et al., 1997




from Menziesi et al., 2010

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## Translation Conclusions

- **Signaling pathways interact to integrate nutritional signals**
  - Insulin/(IGF-1)
  - Acetate/glucose (AMP)
  - Amino acids

  } Energy Supply and Composition

- **We need to discard our current AA requirement framework or drastically revise it**
  - Postabsorptive AA efficiency is variable
  - Energy, AA mix, and hormonal state partially dictate AA efficiency

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## Gene Expression Conclusions

- Clearly there is milk protein gene regulation!
  - Lactating vs non-lactating well explored
- Evidence for nutritional control: GH/IGF-1 and Insulin
  - Missing dose/response data to assess lactogenic role
  - Amino acid role?
  - Glucose, acetate, and fatty acids?
- RM Akers, 2006
  - "there is an exciting, and bewildering universe of growth factors, transcription factors, receptors, intracellular signaling intermediates, and extracellular molecules that must ultimately interact to determine the size of the mature udder and the functional capacity of mammary gland in the lactating cow".

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## Acknowledgements


Travel Support
- Rednex Project

Research Funding
- Balchem
- USDA/CSREES under Regional Research Project NC-1040
- Land O' Lakes/Purina Feed
- Venture Milling
- Virginia Agricultural Council
- Virginia State Dairymen's Association

Students
- Ranga Appuhamy
- Agustin Rius
- Deepthi Nayananjalie
- Sebastian Arriola
- Ashley Bell

Collaborators
- Jim France, U. of Guelph
- Danijela Kirovsky, U. of Belgrade
- Jeffery Escobar, VT
- Mike Akers, VT
- Mike McGilliard, VT
- Ondrej Becvar, VT


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