





#### Correlation between growth curve parameters and plasma metabolites in replacement dairy heifers

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### Introduction

#### Rearing period of dairy heifers: nutritional status and growth

- <u>Goals</u>: rate of growth, sexual maturity  $\rightarrow$  13 mo  $\Rightarrow$  conceive  $\rightarrow$  15 mo
- <u>Problems</u>: sub-optimal nutrition, delayed puberty; <u>skeletal</u> <u>growth</u>  $\rightarrow$  increases the risk of dystocia Brickell *et al.*, (2009)
- Plasma metabolites in growing dairy heifers: nutritional status and prepubertal development
- metabolic changes as signals for pubertal development (Gasser et al., 2006)
- Brickell et al. (2009)  $\rightarrow$  plasma IGF-I, insulin, glucose, urea
- related to growth
- affected by energy and protein intake

Other plasma metabolites: minerals, enzymes, proteins?

#### Heifer metabolism and long-term effects

Negative correlation prepubertal - postcalving plasma [glucose]  $\rightarrow$  link between metabolic profile of the growing heifer and that at the beginning of the first lactation? (Taylor et al., 2004)



### Introduction

#### AIM

- to investigate on the relationships among growth curve, metabolic profile during growth, and metabolic profile just before 1<sup>st</sup> calving
- to explore the possibility of new statistical approaches, based on Principal Component Analysis (PCA), to describe the relationships between metabolism and growth of dairy heifers

### **Material and Methods**

#### Animals, treatments, and controls

- 60 Italian Friesian heifers, 2 experimental feeding groups: moderate (0.70 kg/d; M, n=40) vs. high (0.90 kg/d; H, n=20) ADG from the 5<sup>th</sup> to the 15<sup>th</sup> mo of age
- Diets were calculated according to NRC (2001)

#### **Body measurements**

 Every 28 d: BW, BCS (ADAS), wither height (WH), hip height (HH), body length (BL), heart girth (HG)



### **Material and Methods**

**Blood sampling and analysis** 

- Blood samples at 9 and 15 mo of age, and 2 wk prepatum
- Determined parameters:
  - blood: haematocrit
  - plasma: glucose, urea, total cholesterol, Ca, inorganic
     P, Mg, Na, K, Cl, total protein, albumin, total bilirubin,
     Zn, alkaline phosphatase (AP), aspartate
     aminotransferase (AST), and γ-glutamyltransferase (γ-GT), ceruloplasmin, and haptoglobin
  - plasma (prepartum only): triglycerides, NEFA, BHBA, creatinine, and LDH

### **Material and Methods**

Statistical analysis

- BW, WH, HH, BL, and HG  $\rightarrow$  Laird's form (1966) of the Gompertz's curve: W = W<sub>0</sub> EXP { b<sub>1</sub> [ 1 EXP (-b<sub>2</sub> t )]}
- W = BW (kg) at time t (d),  $W_0$  = BW at time 0, t = time (d),  $b_1$  = initial specific growth rate, and  $b_2$  = maturation rate
- (see next slide for a better understanding of the effect of  $b_1$  and  $b_2$  on the curve)
- $b_1 \text{ and } b_2 \rightarrow ANOVA$
- metabolic profile  $\rightarrow$  ANOVA
- PCA on data from blood at 9 and 15 mo
- Pearson's correlation coefficients between b<sub>1</sub>, b<sub>2</sub>, plasma metabolite during growth and before 1<sup>st</sup> calving, PC1 individual scores

### **Results and Discussion**

Differences from dietary treatment (Abeni et al., 2006)

- actual ADG: 0.77 kg/d (M) vs. 0.90 kg/d (H); BW at 15 mo 359 vs. 406 kg respectively
- differences in  $b_1$  HG (P = 0.03); no difference in  $b_2$
- higher plasma urea, albumin, at 9 mo, and γ-GT at both ages, in H heifers (Abeni et al., 2006)
- range of variation within reference values (see next slides) for the considered metabolites

Range of values at <u>9 mo</u> of age compared to the reference range (mean = 0; lower limit = -50%; upper limit = +50%) of dry cow



Range of values at <u>15 mo</u> of age compared to the reference range (mean = 0; lower limit = -50%; upper limit = +50%) of dry cow



Summary of Pearson's correlation coefficients (P < 0.001) for the associations between metabolic parameters measured at 9 and 15 months of age and just before 1<sup>st</sup> calving with growth curve parameters of BW, WH, HH, BL, and HG.

| Age at measurement | Variable      | Growth curve   |                |                |                |                |                |
|--------------------|---------------|----------------|----------------|----------------|----------------|----------------|----------------|
|                    | -             | BW             |                | HH             |                | HG             |                |
|                    |               | b <sub>1</sub> | b <sub>2</sub> | b <sub>1</sub> | b <sub>2</sub> | b <sub>1</sub> | b <sub>2</sub> |
|                    |               |                |                |                |                |                |                |
| 9 months           | Urea          |                |                | 0.387          | -0.457         | 0.428          | -0.513         |
|                    | Ceruloplasmin |                |                | 0.362          | -0.518         |                |                |
| 15 months          | Glucose       | 0.619          | -0.700         |                |                |                |                |
|                    | Ceruloplasmin | 0.412          | -0.413         | 0.467          | -0.465         |                |                |
| Prepartum          | Creatinine    |                |                | 0.497          | -0.486         |                |                |
|                    |               |                |                |                |                |                |                |
|                    |               |                |                |                |                |                |                |

- Plasma urea at 9 mo and HG curve agree with Brickell *et* al. (2009) ⇒ development of gastro-intestinal system?
- Results on plasma glucose at 15 mo agree with those of Brickell *et al.* (2009)
- Plasma ceruloplasmin at 9 and 15 mo ⇒ not only a signal of inflammatory process?
- Prepartum creatinine and HH growth curve ⇒ higher heifers with greater mass of skeletal muscles?

Figure 1 - Loading plot, describing the relationships among plasma metabolites derived from a principal component analysis based on blood sampling at 9 and 15 months of age in dairy heifers (n = 60); Eigenvalues: PC1=7.51 (18.8% of tot. var.); PC2=4.41 (11.0% of tot. var.).



# 2. Principal Components Analysis (PCA) with metabolic profile of growing heifers

Summary of Pearson's correlation coefficients for the associations between PC1 individual scores and growth curve parameters of BW, WH, HH, BL, and HG.

|   |                       |                | Growth                | curve          |                |                |  |
|---|-----------------------|----------------|-----------------------|----------------|----------------|----------------|--|
|   | BW                    |                | HH                    |                | HG             |                |  |
|   | <b>b</b> <sub>1</sub> | b <sub>2</sub> | <b>b</b> <sub>1</sub> | b <sub>2</sub> | b <sub>1</sub> | b <sub>2</sub> |  |
|   |                       |                |                       |                |                |                |  |
| r | 0.38                  | -0.48          | 0.34                  | -0.57          | 0.39           | -0.53          |  |
|   |                       |                |                       |                |                |                |  |
| Р | 0.004                 | 0.0001         | 0.008                 | < 0.0001       | 0.002          | < 0.0001       |  |
|   |                       |                |                       |                |                |                |  |
|   |                       |                |                       |                |                |                |  |

## 2. Principal Components Analysis (PCA) with metabolic profile of growing heifers

- PC1 correlated with growth measures
- PC2 related with age at sampling
- PC1 mainly composed by albumin, globulin, and their sum at both ages, plasma glucose at 15 mo, and macrominerals at 15 mo
- Relative scarcity of literature on macrominerals in growing dairy heifers, also considering changes in growth potential related to changes in genetic merit for milk production

# 3. Correlations between metabolic profile of growing and precalving heifers

• Plasma cholesterol at 15 mo and just before calving (r=0.542; P<0.0001)  $\Rightarrow$  does it related to genetic basis?

 Plasma glucose at 15 mo and precalving plasma albumin (r=-0.447; P=0.0001) ⇒ higher glucose and insulin resistance (?); relations with inflammation susceptibility (?)

# 4. Correlations between metabolic parameters of precalving heifers

|                           | NEFA (Log <sub>10</sub> ) | BHBA (Log <sub>10</sub> ) |
|---------------------------|---------------------------|---------------------------|
| Cucp                      | 0.425***                  | 0.551***                  |
| AST                       | 0.394***                  | 0.655***                  |
| Bilirubin                 | 0.771***                  | 0.672***                  |
| NEFA (Log <sub>10</sub> ) | -                         | 0.628***                  |
| BHBA (Log <sub>10</sub> ) | 0.628***                  | -                         |
| Creatinine                | 0.465***                  | 0.374**                   |
| LDH                       | 0.439***                  | 0.692***                  |
|                           |                           |                           |

Bilirubin as signal of digestive or hepatic problems that may lead to early body fat mobilization?
LDH, and AST ⇒

signals of hepatic activity/problems? Plasma creatinine as signal of body proteins mobilization, jointly with body fat (↑ NEFA)?



- There are many relationships between growth curves and metabolic profile of growing heifers, specially for the aspects related with energy and protein metabolism
- A combined approach with PCA and growth curves allows an integrated view of the heifer metabolism during growth
- Is there a role for the mineral component of heifer nutrition that has to be optimized?

#### Thank you for your attention

