

Using Omics to Understand Seasonal Weight Loss in Dairy Goats: An overview of Project GOATOMICS major achievements

André Martinho de Almeida

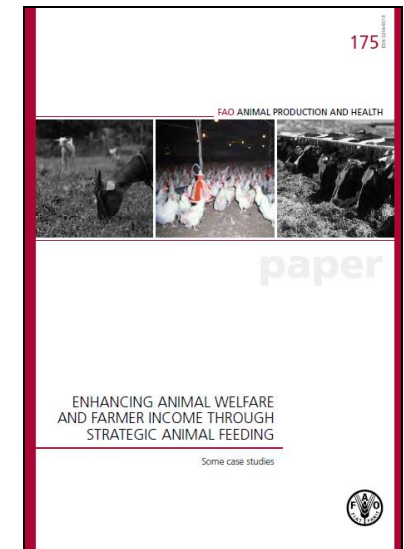
aalmeida@isa.ulisboa.pt

Thessaloniki, Greece

(2018)

Introduction

- Seasonal weight loss poses a serious limitation to animal production in Tropical and Mediterranean areas
- Due to poor quality of pastures in the dry season, animals may lose up to 30 % of their initial body weight – constraint to ruminant production sectors with special relevance to ruminant extensive production
- In contrast, rainy season pastures are usually considered of adequate quality and availability
- To control Seasonal Weight Loss, supplementation is often implemented during dry season – Unavailable in undeveloped countries and Expensive in developed countries



The Canary islands, Spain...



Dairy goats and the Canary Islands (Spain)

common ancestry



Palmera Breed

- adapted to rainy climate
- low tolerance to pasture scarcity



Majorera Breed

- adapted to arid climate
- high tolerance to pasture scarcity



Body live weight and milk production parameters in the *Majorera* and *Palmera* goat breeds from the Canary Islands: influence of weight loss

Joana R. Lérias · Lorenzo E. Hernández-Castellano · Antonio Morales-de-laNuez · Susana S. Araújo · Noemí Castro · Anastasio Argüello · Juan Capote · André M. Almeida

Establishment of the biochemical and endocrine blood profiles in the *Majorera* and *Palmera* dairy goat breeds: the effect of feed restriction

Joana R. Lérias^{1,2}, Raquel Peña³, Lorenzo E. Hernández-Castellano^{4,5}, Juan Capote⁶, Noemí Castro⁴, Anastasio Argüello⁴, Susana S. Araújo^{1,7,8}, Yolanda Saco³, Anna Bassols³ and André M. Almeida^{1,2,7,*}

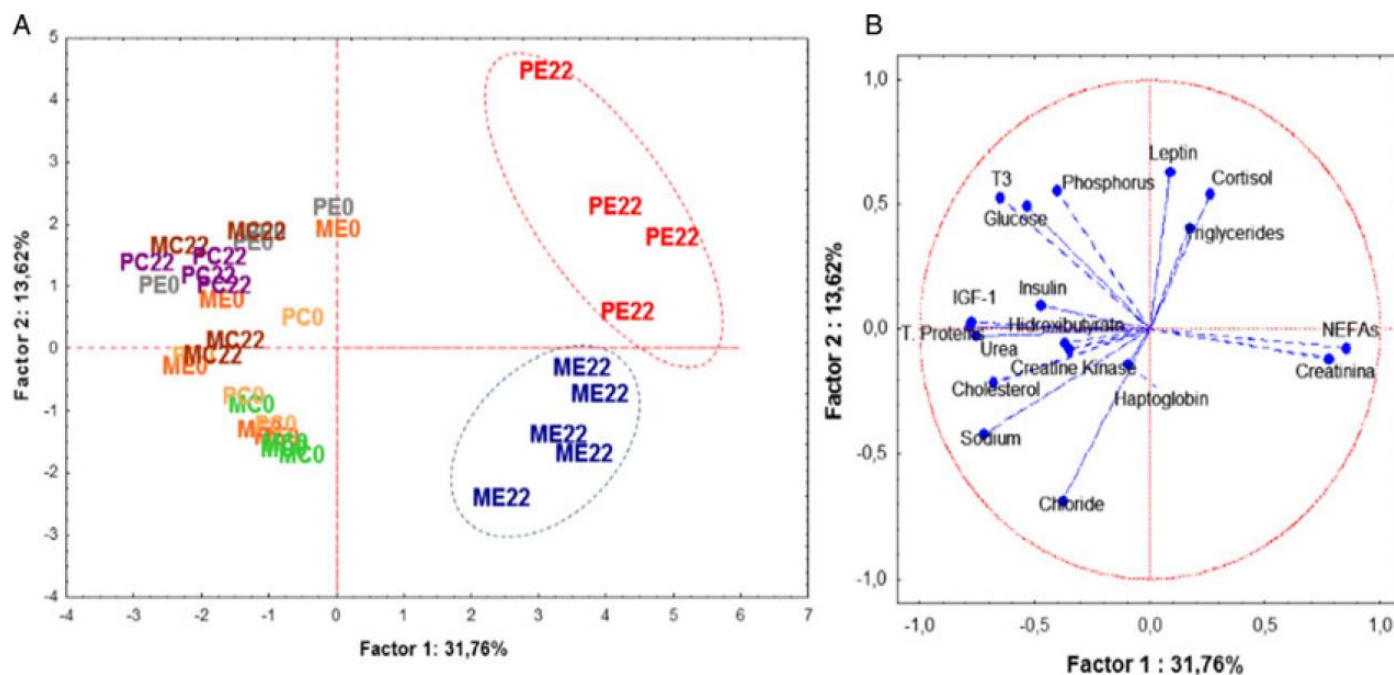


Fig. 1. PCA scatterplot of the *Majorera* and *Palmera* animals (control and underfed) at 0 and 22 d (A) and position of the variables projected in the plane as determined by the first two principal axes (B) (45.38% of the % explained variance). MC0 – *Majorera* Control, day 0; MC22 – *Majorera* control at day 22, ME0 – *Majorera* Underfed at day 0, ME22 – *Majorera* Underfed at day 22; PC0 – *Palmera* Control, day 0; PC22 – *Palmera* control at day 22, PE0 – *Palmera* Underfed at day 0 and PE22 – *Palmera* Underfed at day 22.

Experimental Design



Goats in lactation

Two breeds: Palmera(P) and Majorera(M)

Palmera: susceptible to weight loss

Majorera: tolerant to weight loss

Two groups per breed: Control(C) vs Underfed(E)

Underfed: 15% weight loss in 22 days;
80% decrease milk production

Control: slight increase in BW and same
level of milk production



Univ. Debrecen (Hu)

**Mammary
Gland
Biopsy**



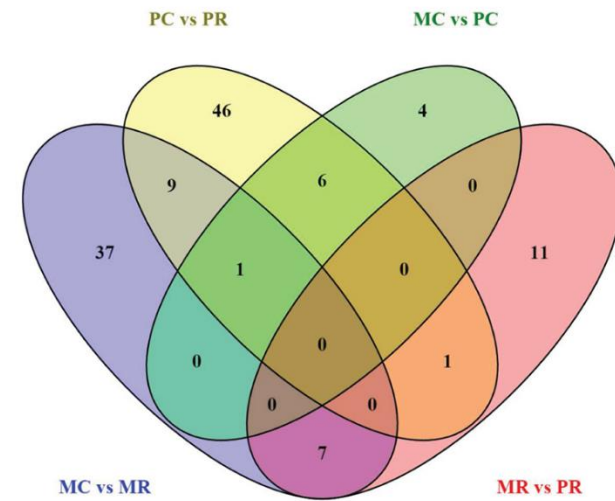
Metabolomics – NMR



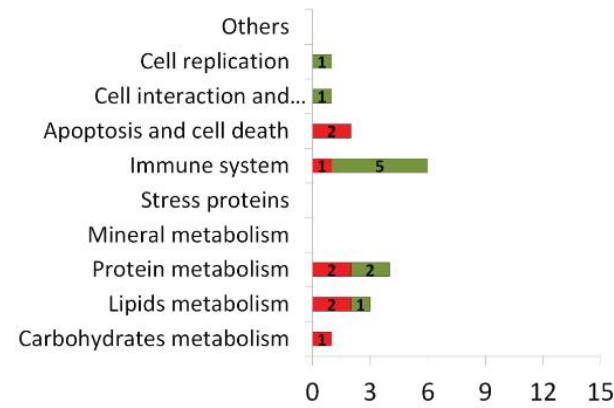
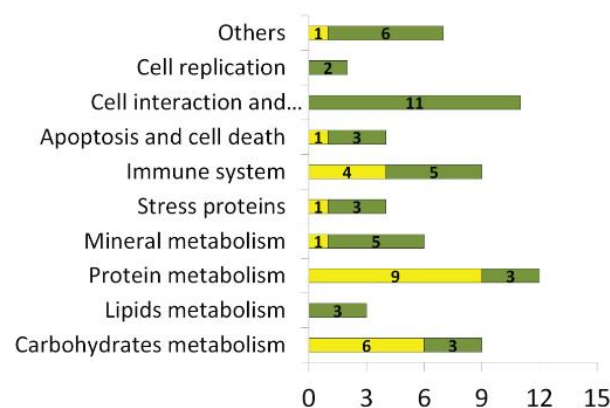
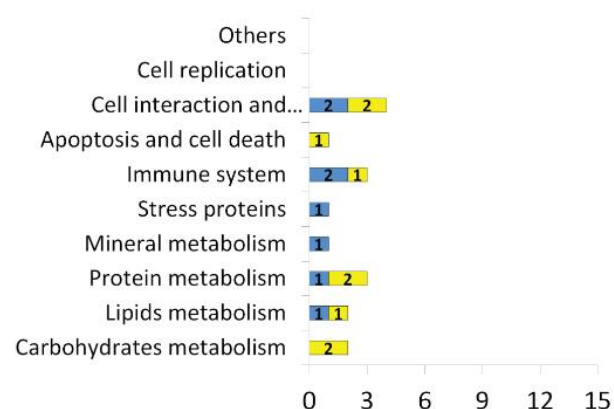
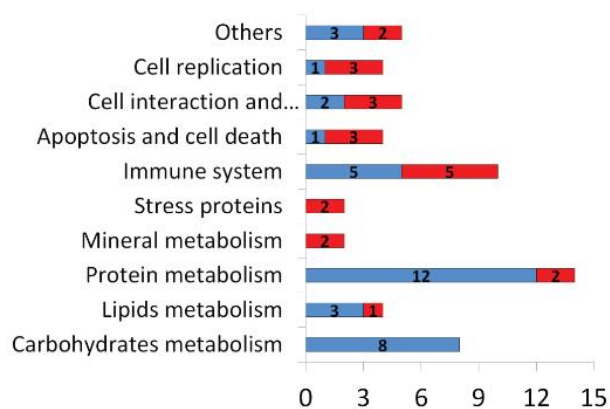
**Label free Proteomics, NGS Transcriptomics
& NMR Metabolomics Analysis**

Proteomics - Major Findings

- First Characterization of the goat mammary gland proteome → over 1000 proteins identified
- Goat entries and sheep and cattle homologues
- 96 proteins had differential expression (fold change > 1.98 and $P < 0.05$)
 - PC vs PE: 63 proteins
 - MC vs ME: 54 proteins
 - MC vs PC: 11 proteins
 - ME vs PE: 19 proteins
- Breed and Nutritional treatment affect significantly the mammary gland secretory tissue proteome



Major Findings



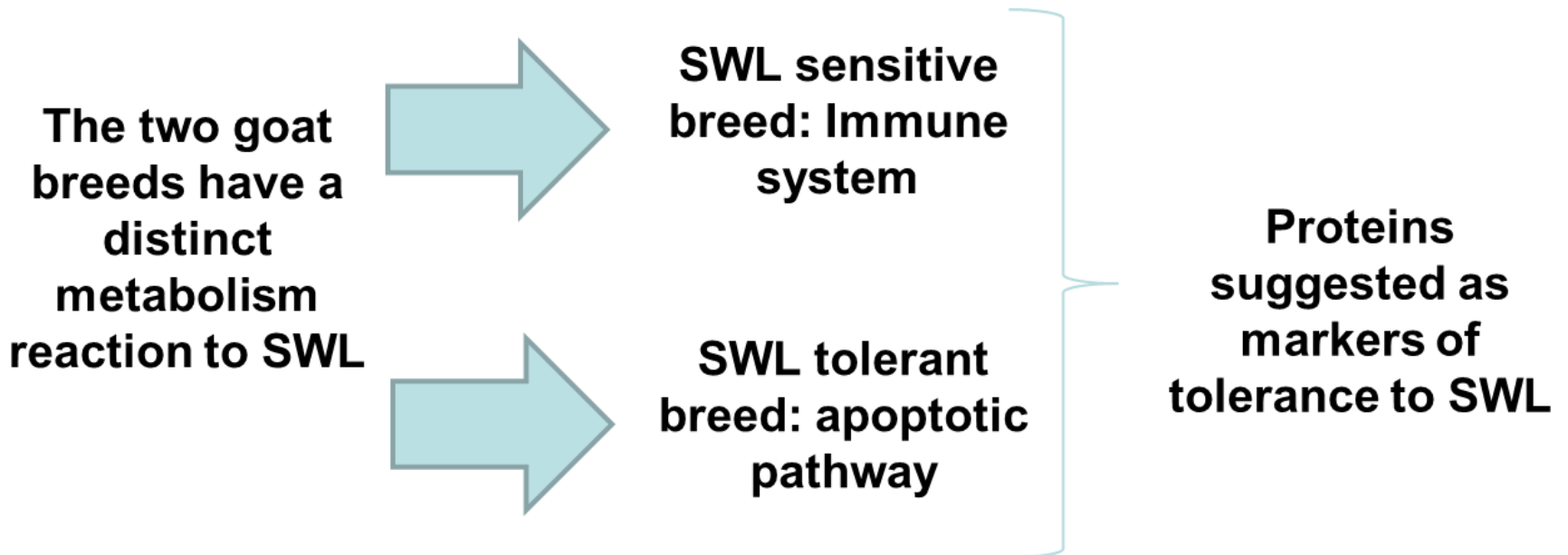
MC

ME

PC

PE

Proteomics: Conclusions



Mammary Gland: metabolite profiling (mmol/g tissue)

Amino Acids Metabolism

	<i>Majorera</i>	<i>Palmera</i>
alanine	$1.0 \times 10^{-3} (\pm 1.9 \times 10^{-4})$	$9.2 \times 10^{-4} (\pm 4.9 \times 10^{-4})$
aspartate	$8.2 \times 10^{-4} (\pm 4.1 \times 10^{-4})$	$8.8 \times 10^{-4} (\pm 3.7 \times 10^{-4})$
creatine	$2.2 \times 10^{-3} (\pm 5.3 \times 10^{-4})$	$1.9 \times 10^{-3} (\pm 5.9 \times 10^{-4})$
glutamate	$7.2 \times 10^{-3} (\pm 1.8 \times 10^{-3})$	$6.3 \times 10^{-3} (\pm 2.9 \times 10^{-3})$
glutathione	$8.0 \times 10^{-4} (\pm 3.2 \times 10^{-4})$	$8.6 \times 10^{-4} (\pm 3.7 \times 10^{-4})$
glycine	$3.2 \times 10^{-3} (\pm 1.2 \times 10^{-3})$	$5.0 \times 10^{-3} (\pm 3.0 \times 10^{-3})$
inosine	$6.8 \times 10^{-4} (\pm 2.0 \times 10^{-4})$	$5.3 \times 10^{-4} (\pm 1.5 \times 10^{-4})$
taurine	$3.2 \times 10^{-3} (\pm 1.4 \times 10^{-3})$	$1.8 \times 10^{-3} (\pm 1.0 \times 10^{-3})$
3-methylhistidine	$1.6 \times 10^{-4} (\pm 4.3 \times 10^{-5})$	$1.7 \times 10^{-4} (\pm 6.7 \times 10^{-5})$
adenosylhomocysteine	$7.4 \times 10^{-5} (\pm 3.2 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 8.6 \times 10^{-5})$
anserine	$1.6 \times 10^{-4} (\pm 3.7 \times 10^{-5})$	$1.9 \times 10^{-4} (\pm 7.1 \times 10^{-5})$
benzoate	$7.7 \times 10^{-5} (\pm 4.7 \times 10^{-5})$	$6.7 \times 10^{-5} (\pm 5.5 \times 10^{-5})$
carnosine	$1.6 \times 10^{-4} (\pm 4.7 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 4.4 \times 10^{-5})$
creatine phosphphate	$2.0 \times 10^{-4} (\pm 1.9 \times 10^{-4})$	$1.0 \times 10^{-4} (\pm 3.6 \times 10^{-5})$
creatinine	$6.4 \times 10^{-5} (\pm 1.1 \times 10^{-5})$	$3.0 \times 10^{-5} (\pm 1.8 \times 10^{-5})$
isoleucine	$1.7 \times 10^{-4} (\pm 3.2 \times 10^{-5})$	$1.3 \times 10^{-4} (\pm 4.3 \times 10^{-5})$
leucine	$3.1 \times 10^{-4} (\pm 5.1 \times 10^{-5})$	$2.6 \times 10^{-4} (\pm 8.8 \times 10^{-5})$
phenylalanine	$1.1 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$9.0 \times 10^{-5} (\pm 2.6 \times 10^{-5})$
tyrosine	$1.4 \times 10^{-4} (\pm 2.2 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 4.2 \times 10^{-5})$
valine	$2.8 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$2.5 \times 10^{-4} (\pm 6.7 \times 10^{-5})$

Carbohydrates Metabolism

	<i>Majorera</i>	<i>Palmera</i>
glucose	$3.8 \times 10^{-3} (\pm 1.5 \times 10^{-3})$	$3.4 \times 10^{-3} (\pm 1.2 \times 10^{-3})$
lactase	$3.9 \times 10^{-3} (\pm 8.6 \times 10^{-4})$	$4.2 \times 10^{-3} (\pm 2.2 \times 10^{-3})$
lactose	$2.5 \times 10^{-2} (\pm 1.0 \times 10^{-2})$	$2.3 \times 10^{-2} (\pm 1.5 \times 10^{-2})$
UDP-glucose	$2.8 \times 10^{-4} (\pm 8.5 \times 10^{-5})$	$2.5 \times 10^{-4} (\pm 9.0 \times 10^{-5})$

Energy Metabolism

	<i>Majorera</i>	<i>Palmera</i>
AMP	$1.1 \times 10^{-3} (\pm 6.1 \times 10^{-4})$	$9.5 \times 10^{-4} (\pm 4.5 \times 10^{-4})$
IMP	$1.5 \times 10^{-4} (\pm 6.2 \times 10^{-5})$	$1.7 \times 10^{-4} (\pm 1.0 \times 10^{-4})$
NAD ⁺	$8.4 \times 10^{-5} (\pm 7.9 \times 10^{-5})$	$5.9 \times 10^{-5} (\pm 3.2 \times 10^{-5})$
ATP/ADP	$7.5 \times 10^{-5} (\pm 4.6 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 6.1 \times 10^{-5})$
NADP ⁺	$4.2 \times 10^{-5} (\pm 1.4 \times 10^{-5})$	$3.2 \times 10^{-5} (\pm 1.1 \times 10^{-5})$

Cofactors and Vitamins Metabolism

	<i>Majorera</i>	<i>Palmera</i>
betaine	$2.4 \times 10^{-3} (\pm 1.2 \times 10^{-3})$	$1.3 \times 10^{-3} (\pm 9.7 \times 10^{-4})$
carnitine	$6.8 \times 10^{-4} (\pm 1.2 \times 10^{-4})$	$4.9 \times 10^{-4} (\pm 1.8 \times 10^{-4})$
choline	$9.9 \times 10^{-4} (\pm 1.2 \times 10^{-4})$	$7.4 \times 10^{-4} (\pm 2.6 \times 10^{-4})$
glycerophosphocholine	$1.6 \times 10^{-3} (\pm 5.5 \times 10^{-4})$	$2.5 \times 10^{-3} (\pm 1.3 \times 10^{-3})$
myo-inositol	$1.5 \times 10^{-3} (\pm 5.5 \times 10^{-4})$	$1.6 \times 10^{-3} (\pm 6.2 \times 10^{-4})$
phosphocholine	$1.2 \times 10^{-3} (\pm 5.7 \times 10^{-4})$	$1.1 \times 10^{-3} (\pm 3.7 \times 10^{-4})$
acetyl-L-carnitine	$1.1 \times 10^{-4} (\pm 5.3 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 3.7 \times 10^{-5})$
methylmalonate	$5.9 \times 10^{-5} (\pm 7.3 \times 10^{-5})$	$5.7 \times 10^{-5} (\pm 6.8 \times 10^{-5})$
nicotinurate	$2.5 \times 10^{-4} (\pm 1.6 \times 10^{-4})$	$1.8 \times 10^{-4} (\pm 5.3 \times 10^{-5})$

Nucleotide Metabolism

	<i>Majorera</i>	<i>Palmera</i>
adenine	$3.1 \times 10^{-4} (\pm 4.2 \times 10^{-4})$	$1.9 \times 10^{-4} (\pm 1.2 \times 10^{-4})$
uridine	$1.5 \times 10^{-4} (\pm 1.0 \times 10^{-4})$	$1.2 \times 10^{-4} (\pm 2.7 \times 10^{-5})$
xanthine	$1.5 \times 10^{-4} (\pm 6.0 \times 10^{-5})$	$1.3 \times 10^{-4} (\pm 6.2 \times 10^{-5})$

TCA Cycle

	<i>Majorera</i>	<i>Palmera</i>
acetate	$1.4 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$2.3 \times 10^{-4} (\pm 1.1 \times 10^{-4})$
formate	$7.6 \times 10^{-4} (\pm 8.9 \times 10^{-4})$	$8.2 \times 10^{-4} (\pm 7.2 \times 10^{-4})$
fumarate	$1.3 \times 10^{-4} (\pm 3.5 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 5.4 \times 10^{-5})$
citrate	$1.1 \times 10^{-4} (\pm 7.2 \times 10^{-5})$	$2.0 \times 10^{-4} (\pm 2.6 \times 10^{-4})$
pyruvate	$2.0 \times 10^{-4} (\pm 8.1 \times 10^{-5})$	$1.6 \times 10^{-4} (\pm 1.3 \times 10^{-4})$
succinate	$1.5 \times 10^{-4} (\pm 7.5 \times 10^{-5})$	$2.0 \times 10^{-4} (\pm 1.0 \times 10^{-4})$

47 metabolites:

all common constituents
of mammary gland and
meat

Mammary Gland: metabolite profiling (mmol/g tissue)

Amino Acids Metabolism

	Majorera	Palmera
alanine	$1.0 \times 10^{-3} (\pm 1.9 \times 10^{-4})$	$9.2 \times 10^{-4} (\pm 4.9 \times 10^{-4})$
aspartate	$8.2 \times 10^{-4} (\pm 4.1 \times 10^{-4})$	$8.8 \times 10^{-4} (\pm 3.7 \times 10^{-4})$
creatine	$2.2 \times 10^{-3} (\pm 5.3 \times 10^{-4})$	$1.9 \times 10^{-3} (\pm 5.9 \times 10^{-4})$
glutamate	$7.2 \times 10^{-3} (\pm 1.8 \times 10^{-3})$	$6.3 \times 10^{-3} (\pm 2.9 \times 10^{-3})$
glutathione	$8.0 \times 10^{-4} (\pm 3.2 \times 10^{-4})$	$8.6 \times 10^{-4} (\pm 3.7 \times 10^{-4})$
glycine	$3.2 \times 10^{-3} (\pm 1.2 \times 10^{-3})$	$5.0 \times 10^{-3} (\pm 3.0 \times 10^{-3})$
inosine	$6.8 \times 10^{-4} (\pm 2.0 \times 10^{-4})$	$5.3 \times 10^{-4} (\pm 1.5 \times 10^{-4})$
taurine	$3.2 \times 10^{-3} (\pm 1.4 \times 10^{-3})$	$1.8 \times 10^{-3} (\pm 1.0 \times 10^{-3})$
3-methylhistidine	$1.6 \times 10^{-4} (\pm 4.3 \times 10^{-5})$	$1.7 \times 10^{-4} (\pm 6.7 \times 10^{-5})$
adenosylhomocysteine	$7.4 \times 10^{-5} (\pm 3.2 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 8.6 \times 10^{-5})$
anserine	$1.6 \times 10^{-4} (\pm 3.7 \times 10^{-5})$	$1.9 \times 10^{-4} (\pm 7.1 \times 10^{-5})$
benzoate	$7.7 \times 10^{-5} (\pm 4.7 \times 10^{-5})$	$6.7 \times 10^{-5} (\pm 5.5 \times 10^{-5})$
carnosine	$1.6 \times 10^{-4} (\pm 4.7 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 4.4 \times 10^{-5})$
creatine phosphphate	$2.0 \times 10^{-4} (\pm 1.9 \times 10^{-4})$	$1.0 \times 10^{-4} (\pm 3.6 \times 10^{-5})$
creatinine	$6.4 \times 10^{-5} (\pm 1.1 \times 10^{-5})$	$3.0 \times 10^{-5} (\pm 1.8 \times 10^{-5})$
isoleucine	$1.7 \times 10^{-4} (\pm 3.2 \times 10^{-5})$	$1.3 \times 10^{-4} (\pm 4.3 \times 10^{-5})$
leucine	$3.1 \times 10^{-4} (\pm 5.1 \times 10^{-5})$	$2.6 \times 10^{-4} (\pm 8.8 \times 10^{-5})$
phenylalanine	$1.1 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$9.0 \times 10^{-5} (\pm 2.6 \times 10^{-5})$
tyrosine	$1.4 \times 10^{-4} (\pm 2.2 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 4.2 \times 10^{-5})$
valine	$2.8 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$2.5 \times 10^{-4} (\pm 6.7 \times 10^{-5})$

Carbohydrates Metabolism

	Majorera	Palmera
glucose	$3.8 \times 10^{-3} (\pm 1.5 \times 10^{-3})$	$3.4 \times 10^{-3} (\pm 1.2 \times 10^{-3})$
lactase	$3.9 \times 10^{-3} (\pm 8.6 \times 10^{-4})$	$4.2 \times 10^{-3} (\pm 2.2 \times 10^{-3})$
lactose	$2.5 \times 10^{-2} (\pm 1.0 \times 10^{-2})$	$2.3 \times 10^{-2} (\pm 1.5 \times 10^{-2})$
UDP-glucose	$2.8 \times 10^{-4} (\pm 8.5 \times 10^{-5})$	$2.5 \times 10^{-4} (\pm 9.0 \times 10^{-5})$

Energy Metabolism

	Majorera	Palmera
AMP	$1.1 \times 10^{-3} (\pm 6.1 \times 10^{-4})$	$9.5 \times 10^{-4} (\pm 4.5 \times 10^{-4})$
IMP	$1.5 \times 10^{-4} (\pm 6.2 \times 10^{-5})$	$1.7 \times 10^{-4} (\pm 1.0 \times 10^{-4})$
NAD ⁺	$8.4 \times 10^{-5} (\pm 7.9 \times 10^{-5})$	$5.9 \times 10^{-5} (\pm 3.2 \times 10^{-5})$
ATP/ADP	$7.5 \times 10^{-5} (\pm 4.6 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 6.1 \times 10^{-5})$
NADP ⁺	$4.2 \times 10^{-5} (\pm 1.4 \times 10^{-5})$	$3.2 \times 10^{-5} (\pm 1.1 \times 10^{-5})$

Cofactors and Vitamins Metabolism

	Majorera	Palmera
betaine	$2.4 \times 10^{-3} (\pm 1.2 \times 10^{-3})$	$1.3 \times 10^{-3} (\pm 9.7 \times 10^{-4})$
carnitine	$6.8 \times 10^{-4} (\pm 1.2 \times 10^{-4})$	$4.9 \times 10^{-4} (\pm 1.8 \times 10^{-4})$
choline	$9.9 \times 10^{-4} (\pm 1.2 \times 10^{-4})$	$7.4 \times 10^{-4} (\pm 2.6 \times 10^{-4})$
glycerophosphocholine	$1.6 \times 10^{-3} (\pm 5.5 \times 10^{-4})$	$2.5 \times 10^{-3} (\pm 1.3 \times 10^{-3})$
myo-inositol	$1.5 \times 10^{-3} (\pm 5.5 \times 10^{-4})$	$1.6 \times 10^{-3} (\pm 6.2 \times 10^{-4})$
phosphocholine	$1.2 \times 10^{-3} (\pm 5.7 \times 10^{-4})$	$1.1 \times 10^{-3} (\pm 3.7 \times 10^{-4})$
acetyl-L-carnitine	$1.1 \times 10^{-4} (\pm 5.3 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 3.7 \times 10^{-5})$
methylmalonate	$5.9 \times 10^{-5} (\pm 7.3 \times 10^{-5})$	$5.7 \times 10^{-5} (\pm 6.8 \times 10^{-5})$
nicotinurate	$2.5 \times 10^{-4} (\pm 1.6 \times 10^{-4})$	$1.8 \times 10^{-4} (\pm 5.3 \times 10^{-5})$

Nucleotide Metabolism

	Majorera	Palmera
adenine	$3.1 \times 10^{-4} (\pm 4.2 \times 10^{-4})$	$1.9 \times 10^{-4} (\pm 1.2 \times 10^{-4})$
uridine	$1.5 \times 10^{-4} (\pm 1.0 \times 10^{-4})$	$1.2 \times 10^{-4} (\pm 2.7 \times 10^{-5})$
xanthine	$1.5 \times 10^{-4} (\pm 6.0 \times 10^{-5})$	$1.3 \times 10^{-4} (\pm 6.2 \times 10^{-5})$

TCA Cycle

	Majorera	Palmera
acetate	$1.4 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$2.3 \times 10^{-4} (\pm 1.1 \times 10^{-4})$
formate	$7.6 \times 10^{-4} (\pm 8.9 \times 10^{-4})$	$8.2 \times 10^{-4} (\pm 7.2 \times 10^{-4})$
fumarate	$1.3 \times 10^{-4} (\pm 3.5 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 5.4 \times 10^{-5})$
citrate	$1.1 \times 10^{-4} (\pm 7.2 \times 10^{-5})$	$2.0 \times 10^{-4} (\pm 2.6 \times 10^{-4})$
pyruvate	$2.0 \times 10^{-4} (\pm 8.1 \times 10^{-5})$	$1.6 \times 10^{-4} (\pm 1.3 \times 10^{-4})$
succinate	$1.5 \times 10^{-4} (\pm 7.5 \times 10^{-5})$	$2.0 \times 10^{-4} (\pm 1.0 \times 10^{-4})$

Lactose

$\sim 2 \times 10^{-2}$ mmol/g tissue

Mammary Gland: metabolite profiling (mmol/g tissue)

Amino Acids Metabolism

	<i>Majorera</i>	<i>Palmera</i>
alanine	$1.0 \times 10^{-3} (\pm 1.9 \times 10^{-4})$	$9.2 \times 10^{-4} (\pm 4.9 \times 10^{-4})$
aspartate	$8.2 \times 10^{-4} (\pm 4.1 \times 10^{-4})$	$8.8 \times 10^{-4} (\pm 3.7 \times 10^{-4})$
creatine	$2.2 \times 10^{-3} (\pm 5.3 \times 10^{-4})$	$1.9 \times 10^{-3} (\pm 5.9 \times 10^{-4})$
glutamate	$7.2 \times 10^{-3} (\pm 1.8 \times 10^{-3})$	$6.3 \times 10^{-3} (\pm 2.9 \times 10^{-3})$
glutathione	$8.0 \times 10^{-4} (\pm 3.2 \times 10^{-4})$	$8.6 \times 10^{-4} (\pm 3.7 \times 10^{-4})$
glycine	$3.2 \times 10^{-3} (\pm 1.2 \times 10^{-3})$	$5.0 \times 10^{-3} (\pm 3.0 \times 10^{-3})$
inosine	$6.8 \times 10^{-4} (\pm 2.0 \times 10^{-4})$	$5.3 \times 10^{-4} (\pm 1.5 \times 10^{-4})$
taurine	$3.2 \times 10^{-3} (\pm 1.4 \times 10^{-3})$	$1.8 \times 10^{-3} (\pm 1.0 \times 10^{-3})$
3-methylhistidine	$1.6 \times 10^{-4} (\pm 4.3 \times 10^{-5})$	$1.7 \times 10^{-4} (\pm 6.7 \times 10^{-5})$
adenosylhomocysteine	$7.4 \times 10^{-5} (\pm 3.2 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 8.6 \times 10^{-5})$
anserine	$1.6 \times 10^{-4} (\pm 3.7 \times 10^{-5})$	$1.9 \times 10^{-4} (\pm 7.1 \times 10^{-5})$
benzoate	$7.7 \times 10^{-5} (\pm 4.7 \times 10^{-5})$	$6.7 \times 10^{-5} (\pm 5.5 \times 10^{-5})$
carnosine	$1.6 \times 10^{-4} (\pm 4.7 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 4.4 \times 10^{-5})$
creatine phosphphate	$2.0 \times 10^{-4} (\pm 1.9 \times 10^{-4})$	$1.0 \times 10^{-4} (\pm 3.6 \times 10^{-5})$
creatinine	$6.4 \times 10^{-5} (\pm 1.1 \times 10^{-5})$	$3.0 \times 10^{-5} (\pm 1.8 \times 10^{-5})$
isoleucine	$1.7 \times 10^{-4} (\pm 3.2 \times 10^{-5})$	$1.3 \times 10^{-4} (\pm 4.3 \times 10^{-5})$
leucine	$3.1 \times 10^{-4} (\pm 5.1 \times 10^{-5})$	$2.6 \times 10^{-4} (\pm 8.8 \times 10^{-5})$
phenylalanine	$1.1 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$9.0 \times 10^{-5} (\pm 2.6 \times 10^{-5})$
tyrosine	$1.4 \times 10^{-4} (\pm 2.2 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 4.2 \times 10^{-5})$
valine	$2.8 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$2.5 \times 10^{-4} (\pm 6.7 \times 10^{-5})$

Carbohydrates Metabolism

	<i>Majorera</i>	<i>Palmera</i>
glucose	$3.8 \times 10^{-3} (\pm 1.5 \times 10^{-3})$	$3.4 \times 10^{-3} (\pm 1.2 \times 10^{-3})$
lactase	$3.9 \times 10^{-3} (\pm 8.6 \times 10^{-4})$	$4.2 \times 10^{-3} (\pm 2.2 \times 10^{-3})$
lactose	$2.5 \times 10^{-2} (\pm 1.0 \times 10^{-2})$	$2.3 \times 10^{-2} (\pm 1.5 \times 10^{-2})$
UDP-glucose	$2.8 \times 10^{-4} (\pm 8.5 \times 10^{-5})$	$2.5 \times 10^{-4} (\pm 9.0 \times 10^{-5})$

Energy Metabolism

	<i>Majorera</i>	<i>Palmera</i>
AMP	$1.1 \times 10^{-3} (\pm 6.1 \times 10^{-4})$	$9.5 \times 10^{-4} (\pm 4.5 \times 10^{-4})$
IMP	$1.5 \times 10^{-4} (\pm 6.2 \times 10^{-5})$	$1.7 \times 10^{-4} (\pm 1.0 \times 10^{-4})$
NAD ⁺	$8.4 \times 10^{-5} (\pm 7.9 \times 10^{-5})$	$5.9 \times 10^{-5} (\pm 3.2 \times 10^{-5})$
ATP/ADP	$7.5 \times 10^{-5} (\pm 4.6 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 6.1 \times 10^{-5})$
NADP ⁺	$4.2 \times 10^{-5} (\pm 1.4 \times 10^{-5})$	$3.2 \times 10^{-5} (\pm 1.1 \times 10^{-5})$

Cofactors and Vitamins Metabolism

	<i>Majorera</i>	<i>Palmera</i>
betaine	$2.4 \times 10^{-3} (\pm 1.2 \times 10^{-3})$	$1.3 \times 10^{-3} (\pm 9.7 \times 10^{-4})$
carnitine	$6.8 \times 10^{-4} (\pm 1.2 \times 10^{-4})$	$4.9 \times 10^{-4} (\pm 1.8 \times 10^{-4})$
choline	$9.9 \times 10^{-4} (\pm 1.2 \times 10^{-4})$	$7.4 \times 10^{-4} (\pm 2.6 \times 10^{-4})$
glycerophosphocholine	$1.6 \times 10^{-3} (\pm 5.5 \times 10^{-4})$	$2.5 \times 10^{-3} (\pm 1.3 \times 10^{-3})$
myo-inositol	$1.5 \times 10^{-3} (\pm 5.5 \times 10^{-4})$	$1.6 \times 10^{-3} (\pm 6.2 \times 10^{-4})$
phosphocholine	$1.2 \times 10^{-3} (\pm 5.7 \times 10^{-4})$	$1.1 \times 10^{-3} (\pm 3.7 \times 10^{-4})$
acetyl-L-carnitine	$1.1 \times 10^{-4} (\pm 5.3 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 3.7 \times 10^{-5})$
methylmalonate	$5.9 \times 10^{-5} (\pm 7.3 \times 10^{-5})$	$5.7 \times 10^{-5} (\pm 6.8 \times 10^{-5})$
nicotinurate	$2.5 \times 10^{-4} (\pm 1.6 \times 10^{-4})$	$1.8 \times 10^{-4} (\pm 5.3 \times 10^{-5})$

Nucleotide Metabolism

	<i>Majorera</i>	<i>Palmera</i>
adenine	$3.1 \times 10^{-4} (\pm 4.2 \times 10^{-4})$	$1.9 \times 10^{-4} (\pm 1.2 \times 10^{-4})$
uridine	$1.5 \times 10^{-4} (\pm 1.0 \times 10^{-4})$	$1.2 \times 10^{-4} (\pm 2.7 \times 10^{-5})$
xanthine	$1.5 \times 10^{-4} (\pm 6.0 \times 10^{-5})$	$1.3 \times 10^{-4} (\pm 6.2 \times 10^{-5})$

TCA Cycle

	<i>Majorera</i>	<i>Palmera</i>
acetate	$1.4 \times 10^{-4} (\pm 3.9 \times 10^{-5})$	$2.3 \times 10^{-4} (\pm 1.1 \times 10^{-4})$
formate	$7.6 \times 10^{-4} (\pm 8.9 \times 10^{-4})$	$8.2 \times 10^{-4} (\pm 7.2 \times 10^{-4})$
fumarate	$1.3 \times 10^{-4} (\pm 3.5 \times 10^{-5})$	$1.1 \times 10^{-4} (\pm 5.4 \times 10^{-5})$
citrate	$1.1 \times 10^{-4} (\pm 7.2 \times 10^{-5})$	$2.0 \times 10^{-4} (\pm 2.6 \times 10^{-4})$
pyruvate	$2.0 \times 10^{-4} (\pm 8.1 \times 10^{-5})$	$1.6 \times 10^{-4} (\pm 1.3 \times 10^{-4})$
succinate	$1.5 \times 10^{-4} (\pm 7.5 \times 10^{-5})$	$2.0 \times 10^{-4} (\pm 1.0 \times 10^{-4})$

Lactose

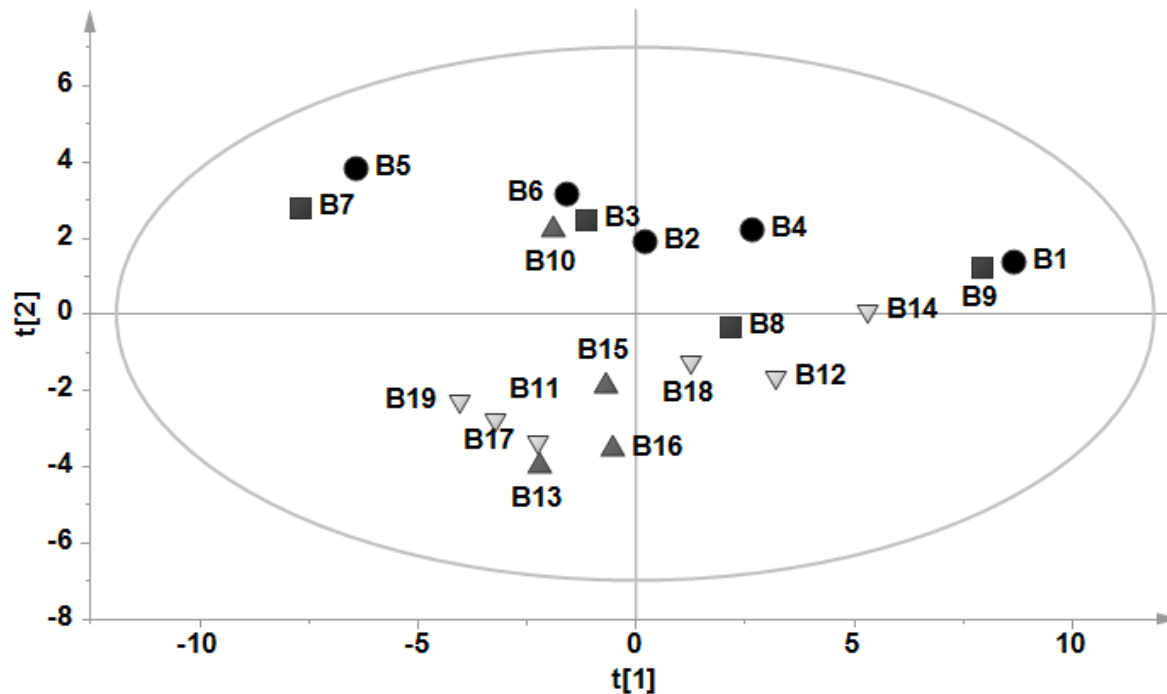
$\sim 2 \times 10^{-2}$ mmol/g tissue

NADP⁺

$\sim 4 \times 10^{-5}$ mmol/g tissue

Mammary Gland: multivariate analysis

Principal Component Analysis (PCA)



PCA:

- unsupervised analysis
- cluster the samples (scores)
- identify metabolites responsible for the group clustering (loadings)

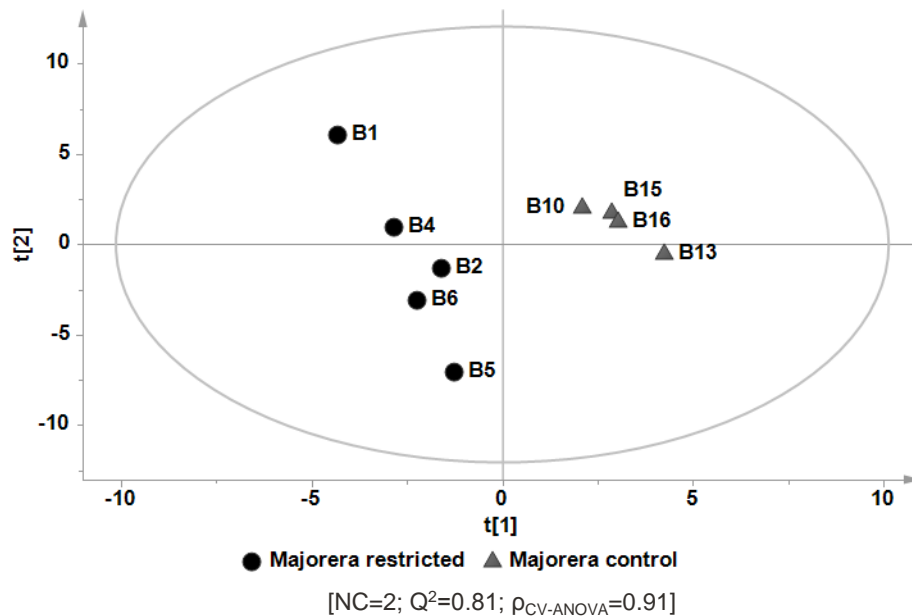
- PCA is not capable of separating the 4 groups.

[NC=3]

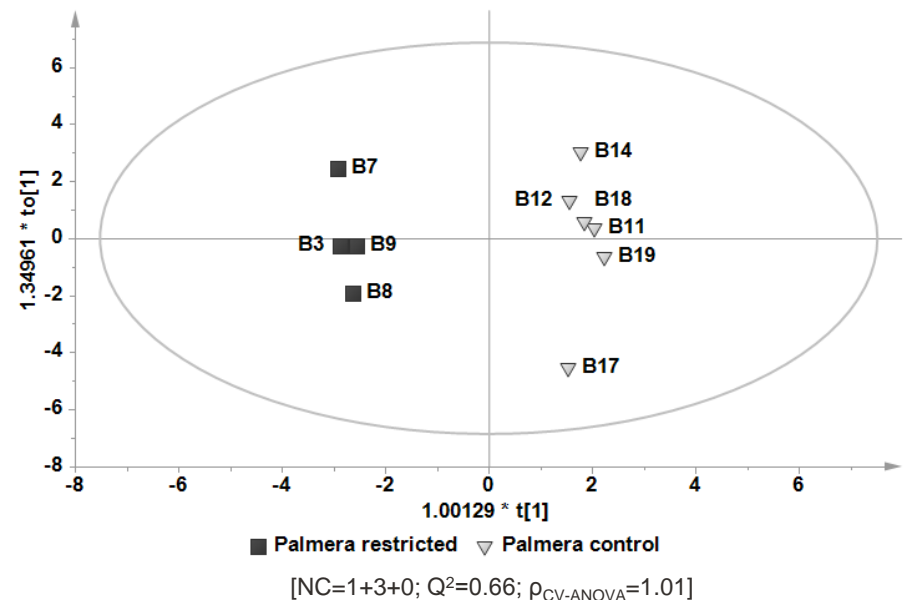
Mammary Gland: multivariate analysis

Partial Least Squares Discriminant Analysis (PLS) & Orthogonal Partial Least Squares Discriminant Analysis (OPLS)

PLS *Majorera* breed



OPLS *Palmera* breed



- PLS/OPLS analysis was necessary to differentiate between control and restricted groups.

PLS / OPLS:

- supervised analysis
- evaluate repeated variables between groups
- highlight differences between groups

Metabolomics: Conclusions

Majorera breed

• Acetate *

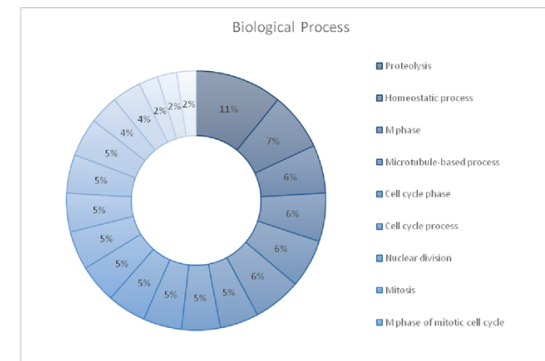
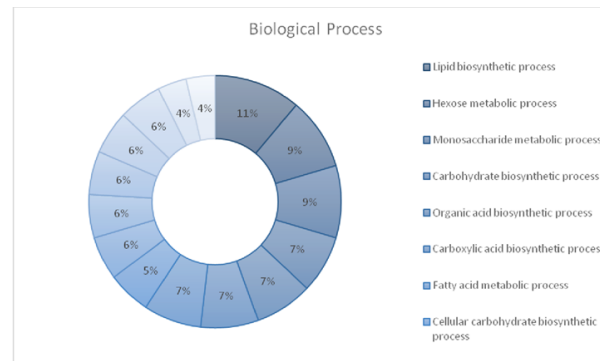
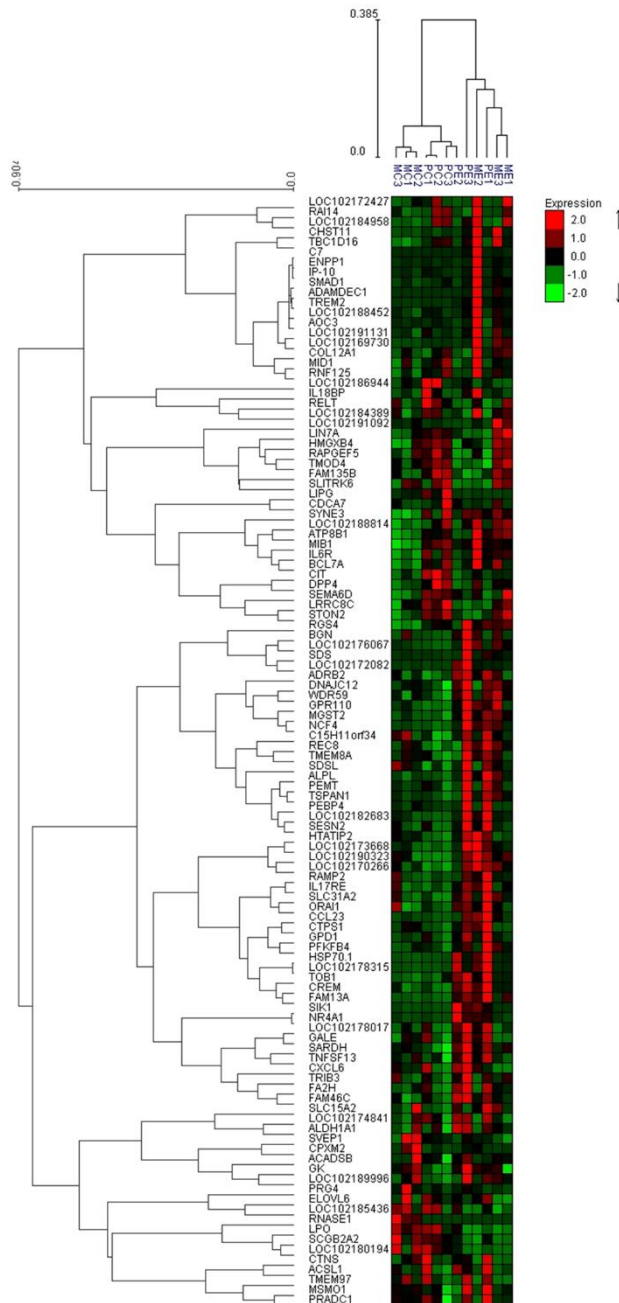
• IMP
• Phosphocholine

- Variations are consistent between breeds
- Differences could be related to:
 - ✓ metabolism adaptation to the low-energy diet
 - ✓ microflora composition *

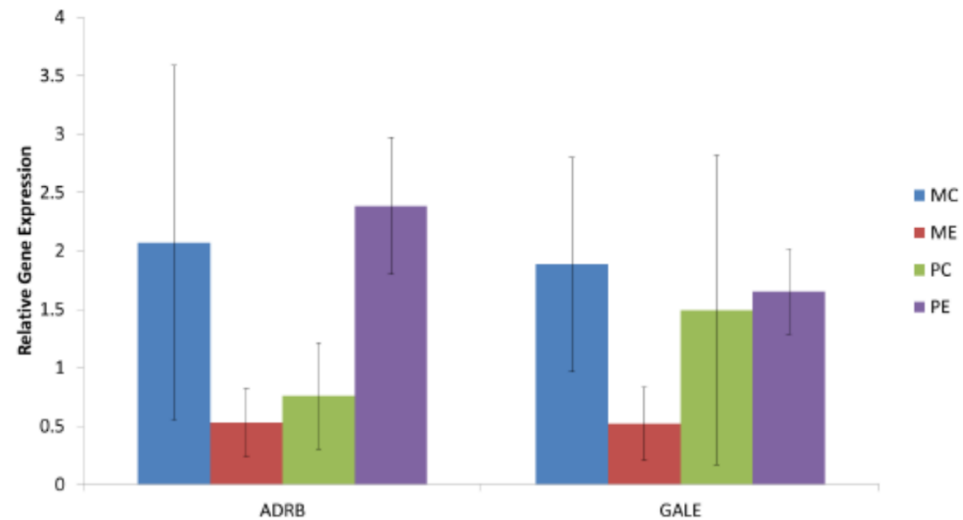
Palmera breed

• IMP
• ATP/ADP/AMP
• Phosphocholine

Transcriptome analysis



Reprogramming of genes expression occurs as result of the stress imposed, irrespective to the breed studied



Validation of expression profiles through Real time quantitative PCR

Transcriptomics Analysis: Major Conclusions

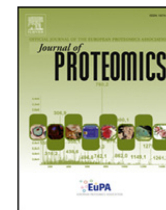
- Reprogramming of gene expression as a result of the stress imposed
- Different behavior of both breeds in response to SWL
- Numerous Biological processes affected in both breeds (Proteolysis, Fatty Acid Synthesis, etc.)
- Release of new genomic resources and shed light into the genes and regulatory networks underlying milk production under nutritional stress



Contents lists available at ScienceDirect

Journal of Proteomics

journal homepage: www.elsevier.com/locate/jprot



The goat (*Capra hircus*) mammary gland secretory tissue proteome as influenced by weight loss: A study using label free proteomics



Lorenzo E. Hernández-Castellano ^a, Ana M. Ferreira ^{b,c}, Paolo Nanni ^d, Jonas Grossmann ^d, Anastasio Argüello ^e, Juan Capote ^f, Guohong Cai ^g, John Lippolis ^g, Noemí Castro ^e, Andre M. de Almeida ^{b,h,i,*}

Molecular
BioSystems



PAPER



Cite this: *Mol. Biosyst.*, 2016,
12, 2094

NMR-metabolomics profiling of mammary gland secretory tissue and milk serum in two goat breeds with different levels of tolerance to seasonal weight loss†

Mariana Palma,^a Lorenzo E. Hernández-Castellano,^b Noemí Castro,^c Anastasio Argüello,^c Juan Capote,^d Manolis Matzapetakis^{‡a} and André Martinho de Almeida^{‡§*aef}

Acknowledgements

ITQB/UNL & IBET (Portugal)

José Salvado

Mariana Palma

Ana FJoana Lerias

erreira

Manolis Matzapetakis

Susana Araujo



Un. Barcelona (Spain) **UAB**

Anna Bassols

Raquel Pato

Universitat Autònoma
de Barcelona

CIIMAR (Portugal)

Graziano Cugno

Alexandre Campos



Functional Genomics

Center Zurich

(Switzerland)

Paolo Nanni

Jonas Grossman

Claudia Fortes



Universität
Zürich^{UZH}

ICIA & ULPGC (Spain)

Juan Capote

Lorenzo Hernandez-Castellano

Antonio Morales

Noemi Castro

Tacho Arguello



USDA (Ames, IA, USA)

John Lippolis

Guonh-Hong Cai



Fundação para a Ciência e a Tecnologia
MINISTÉRIO DA EDUCAÇÃO E CIÊNCIA



- PhD Grant SFRH/BD/85391/2012
- Project PTDC/CVT/116499/2010