



➤ <sup>1</sup>H-NMR metabolomic study of Large White and Meishan pigs in late gestation: part 1- fetal placenta

J. Guibert, A. Imbert, N. Marty-Gasset, L. Gress, C. Cécile, L. Canario, Y. Billon, A. Bonnet, L. Liaubet and **CMD Bonnefont**



## ➤ Context: Influence of placental metabolism on piglet survival

Categories of perinatal mortality	Percentage
Death during farrowing	23.0%
Non- viable	21.4%
Early sepsis	17.6%
Mummified	11.0%
Crushing	7.83%
Starvation	5.58%
Other	13.59%
Total	100%

Pandolfi et al. 2017





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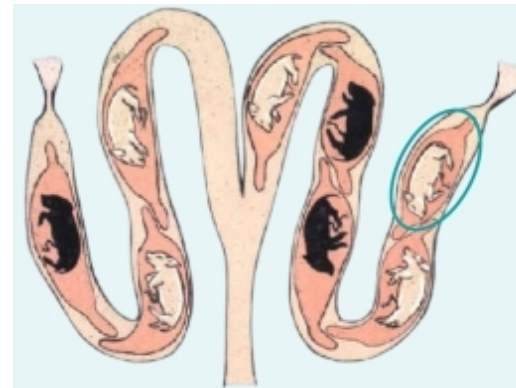
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Selection for lean tissue  
=> **piglets born in a less mature status**  
(Herpin et al. 2002, Panzardi et al. 2013)

**Limitation of the placental area** by the litter size  
=> smaller piglets  
=> and lower chance of survival  
(Rootwelt et al., 2013)

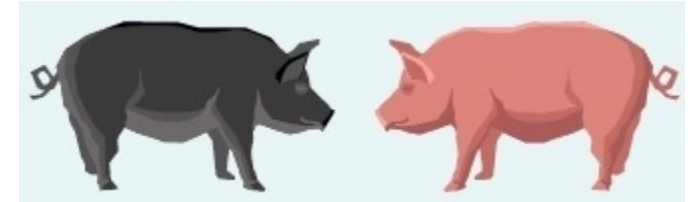
**Placental insufficiency** can impact survival from day 40 of pregnancy  
(Knight et al., 1977; Marsteller et al., 1997)





## > Influence of placental metabolism on piglet survival

Genetic breed influences piglet survival.  
In particular, piglets born from Meishan sows had a lower risk of stillbirth than piglets born from Large White sows  
(Canario et al. 2006)





## > Influence of placental metabolism on piglet survival

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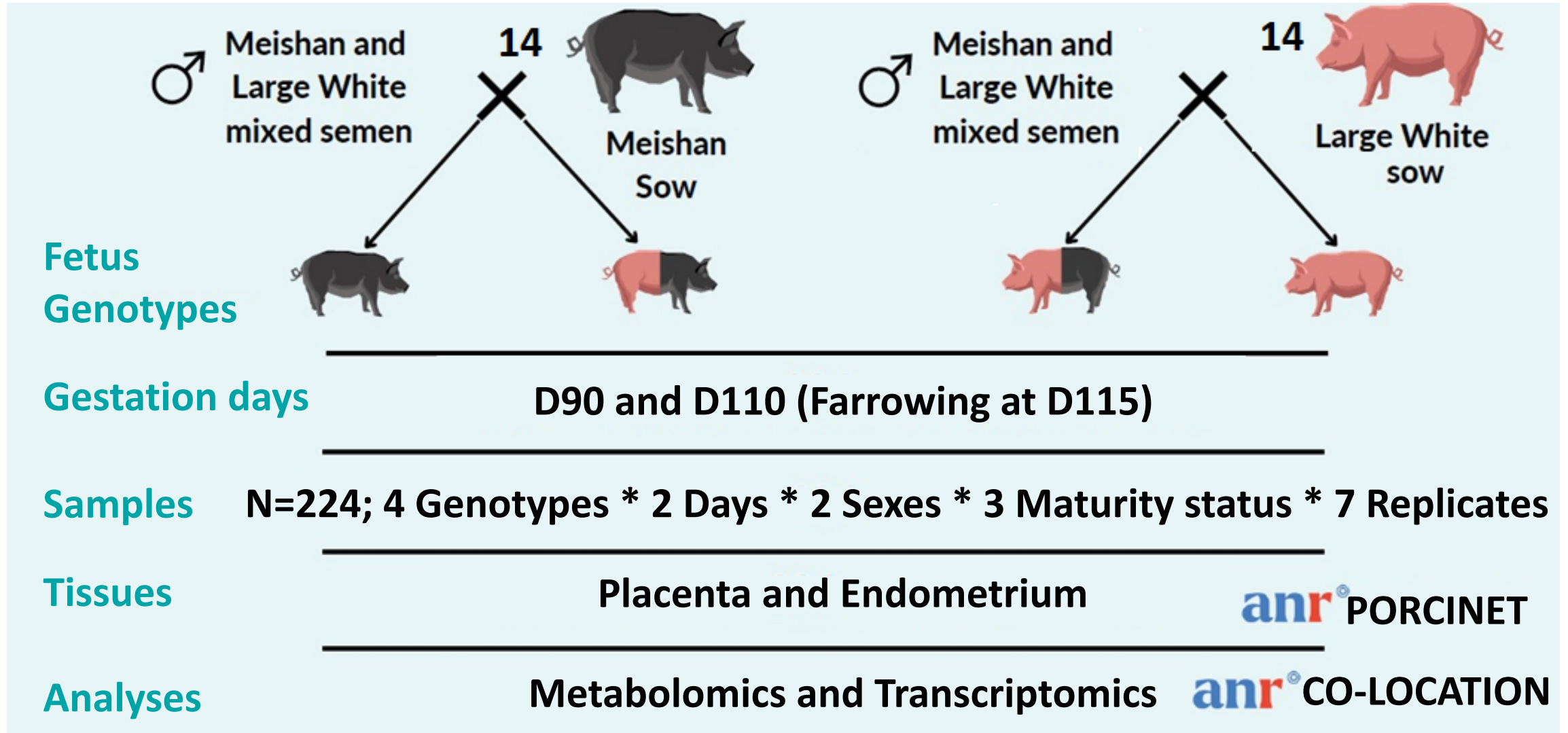


## > Objectives of the study:

- To study the metabolism of placenta with metabolomics analyses at the end of the gestation in both Large White and Meishan breeds
- To better understand the role of placenta in the determination of piglet maturity

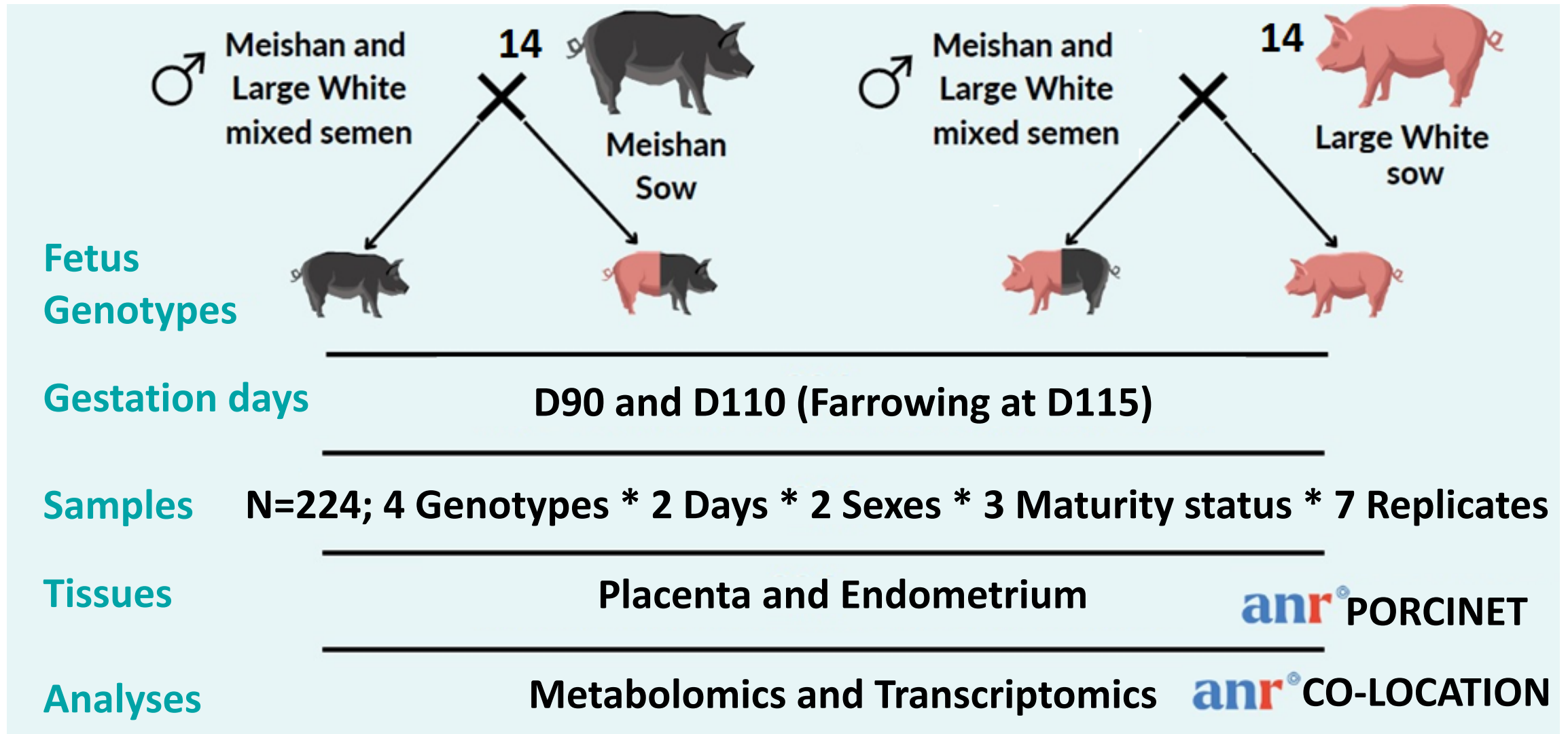


## ➤ Experimental design





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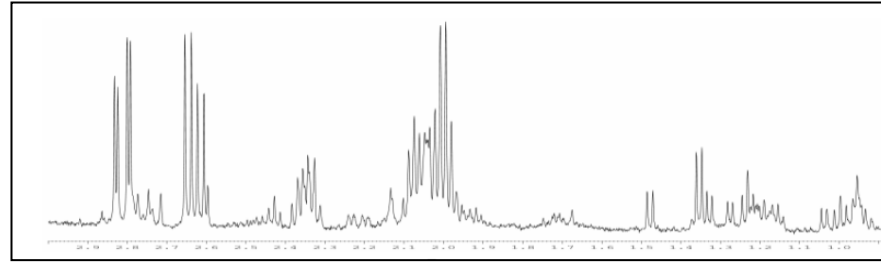


# ➤ <sup>1</sup>H-NMR acquisition and statistical analyses

Extraction of hydrophilic metabolites



NMR 600 MHz  
Bruker Avance III HD  
(MeTaToul-AXIOM)

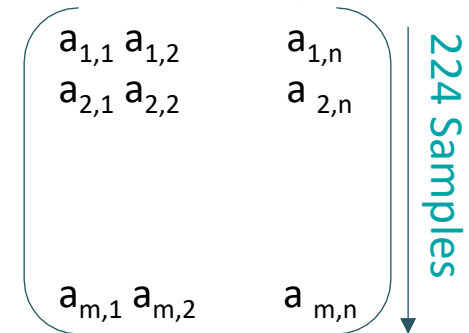


<sup>1</sup>H-NMR spectra



ASICS package  
Lefort et al. 2019

48 Metabolites



48 identified, filtered and selected metabolites

Analyses  
PCA, PLS-DA



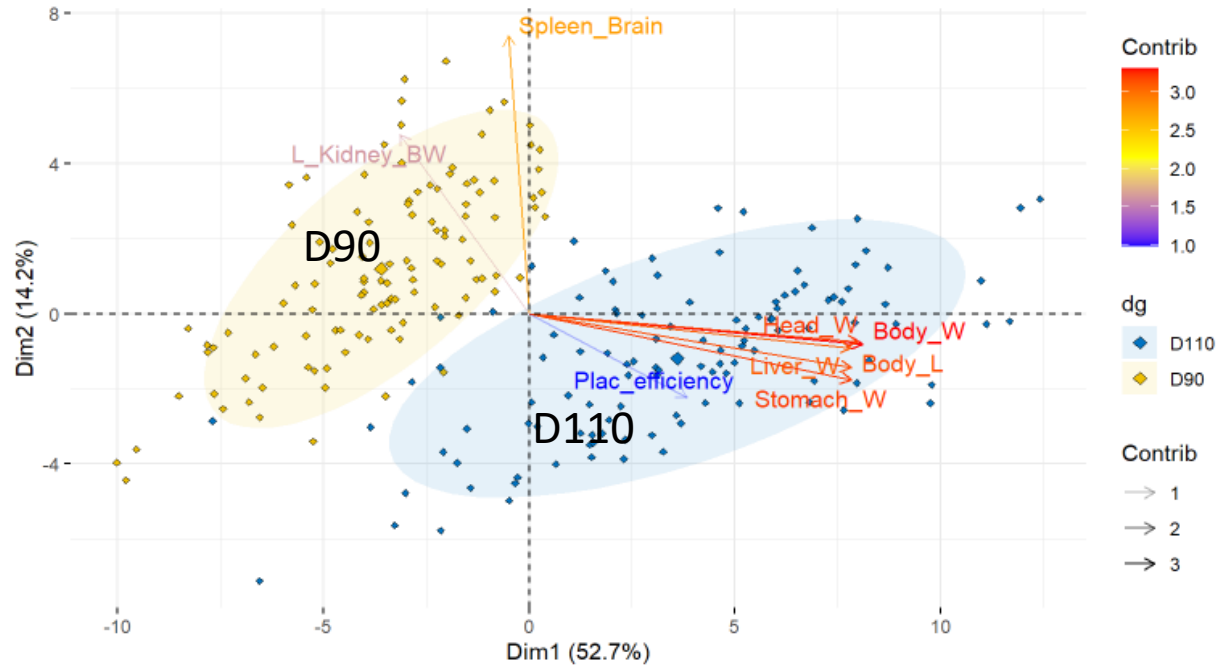


## ➤ Evolution of the fetus phenotypes at the end of gestation

Strong separation between D90 and D110

Strong separation between LW and MS at D90 and D110

Data: Fetus phenotypes,  
Analysis: PCA  
Bi-plot



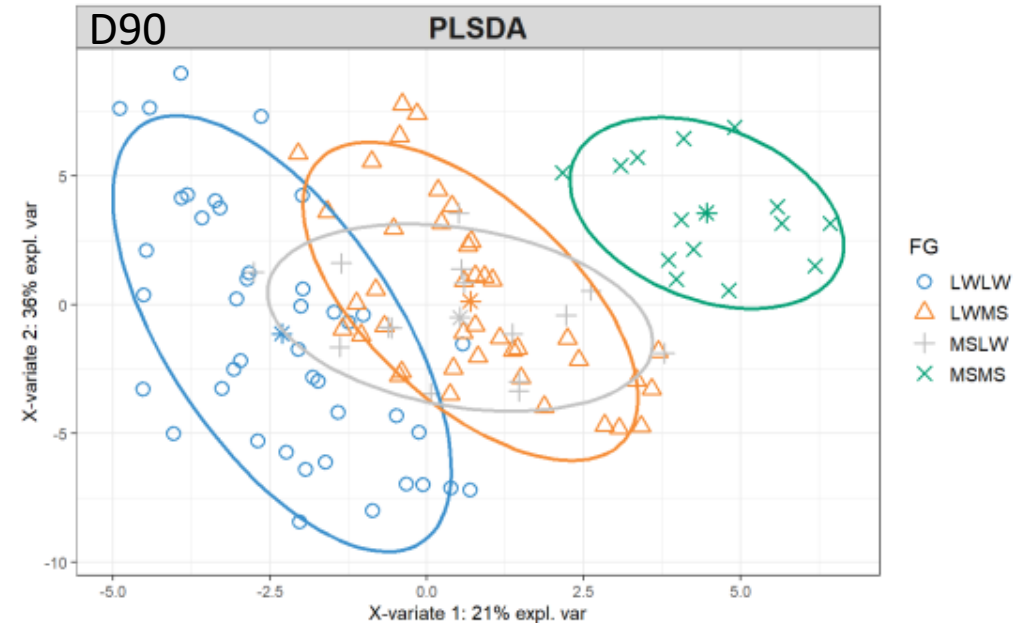
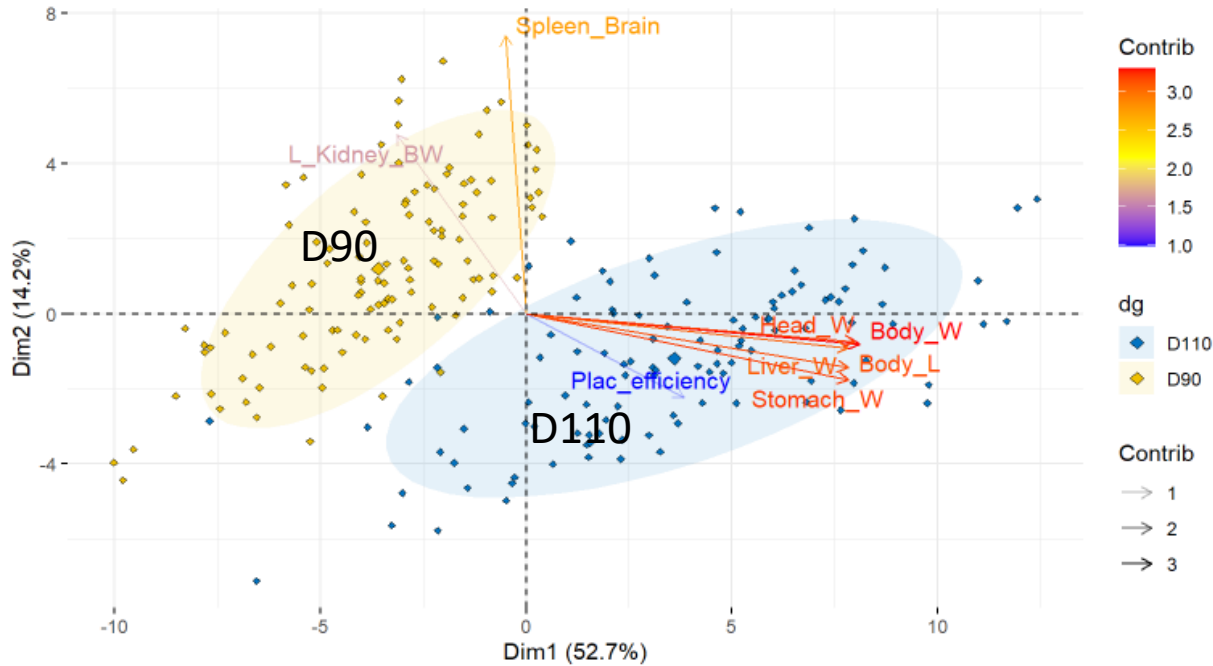


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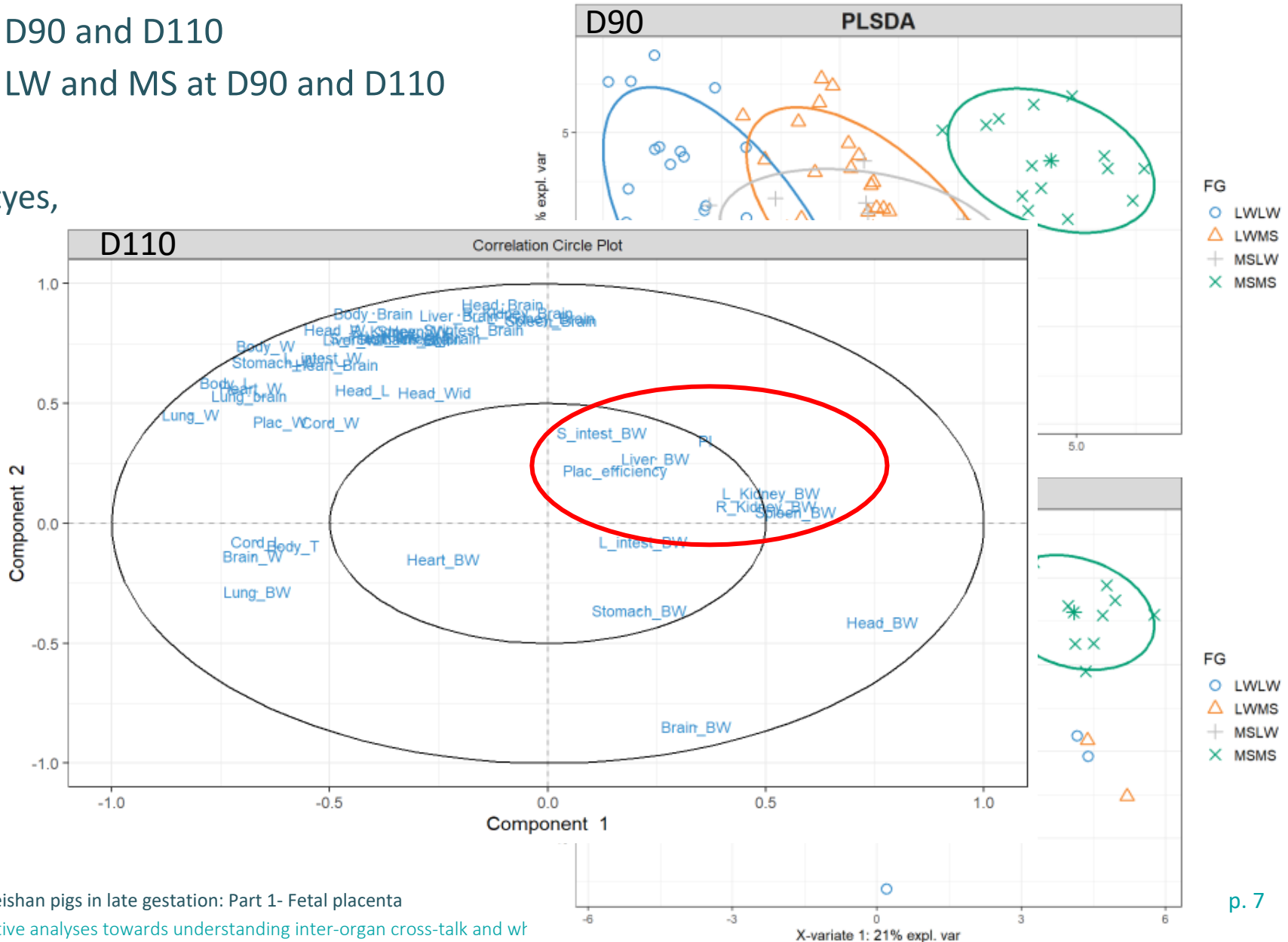
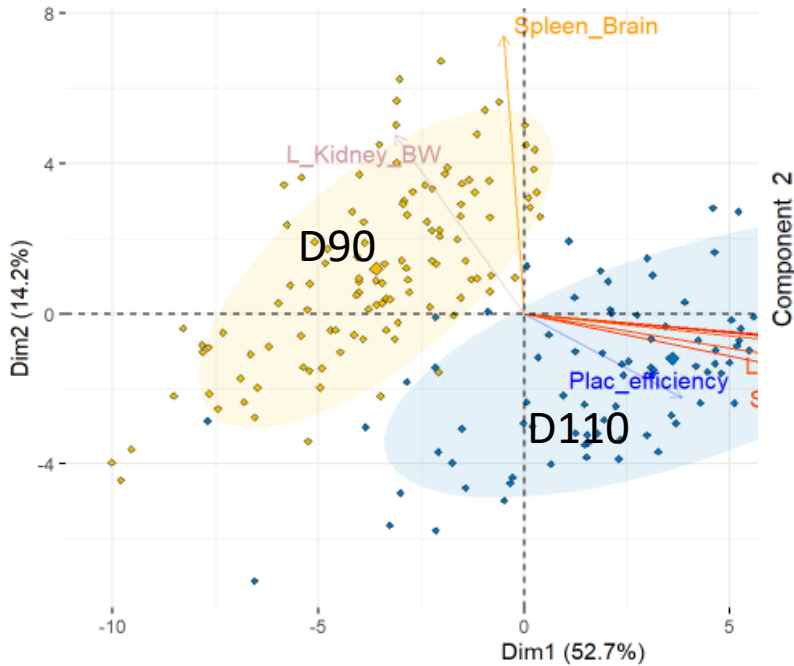


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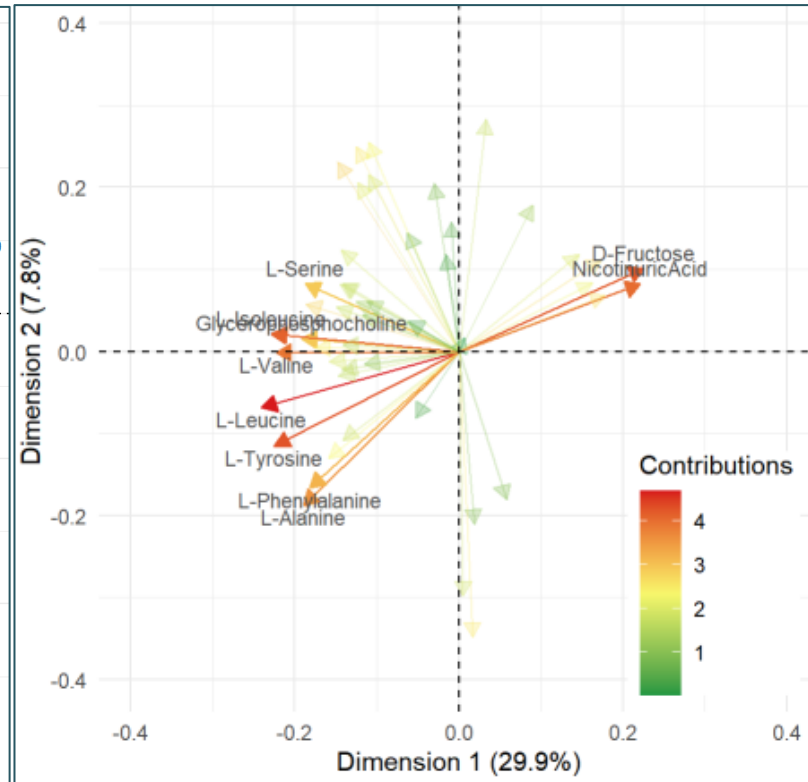
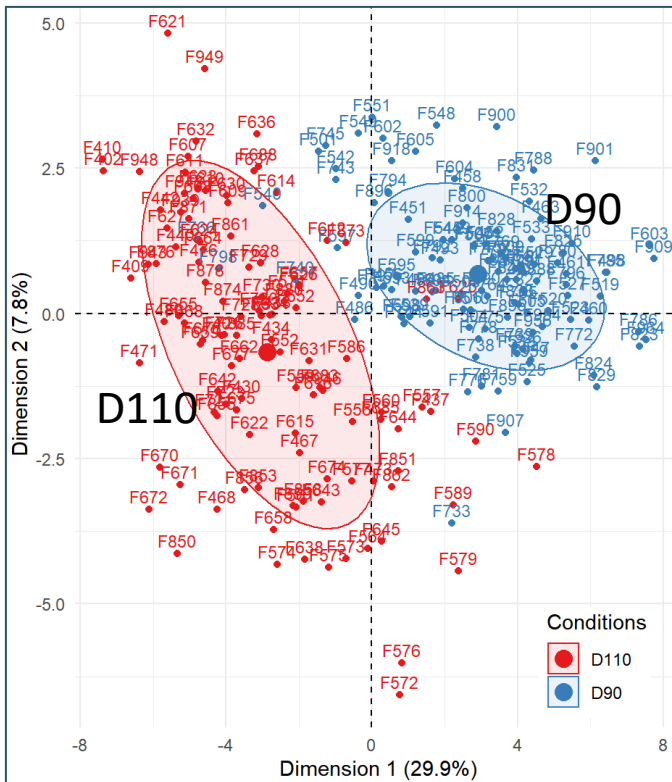
# ➤ Evolution of the placental metabolome at the end of gestation

Comparison 90D vs 110D

Analysis: PCA

Individual plot

Variable plot



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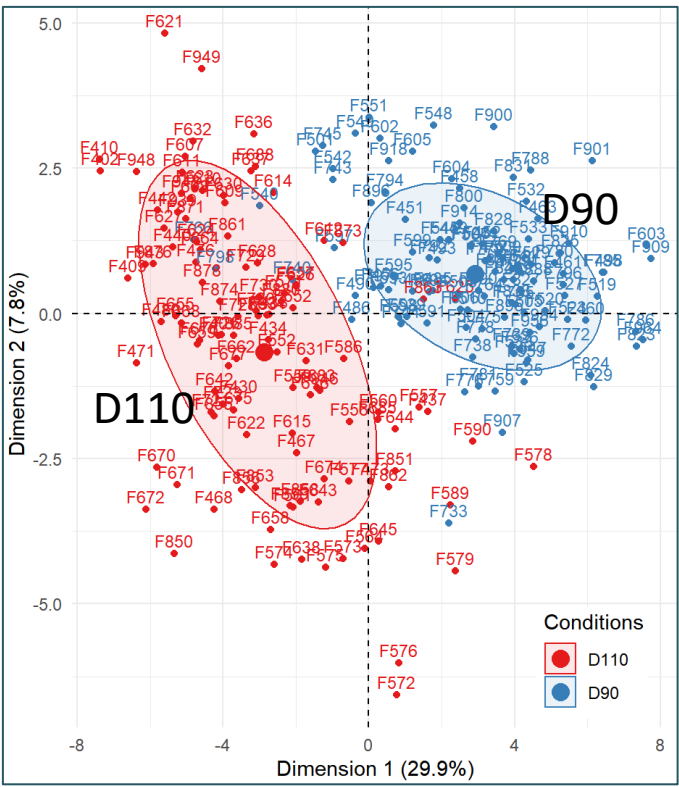


Comparison 90D vs 110D

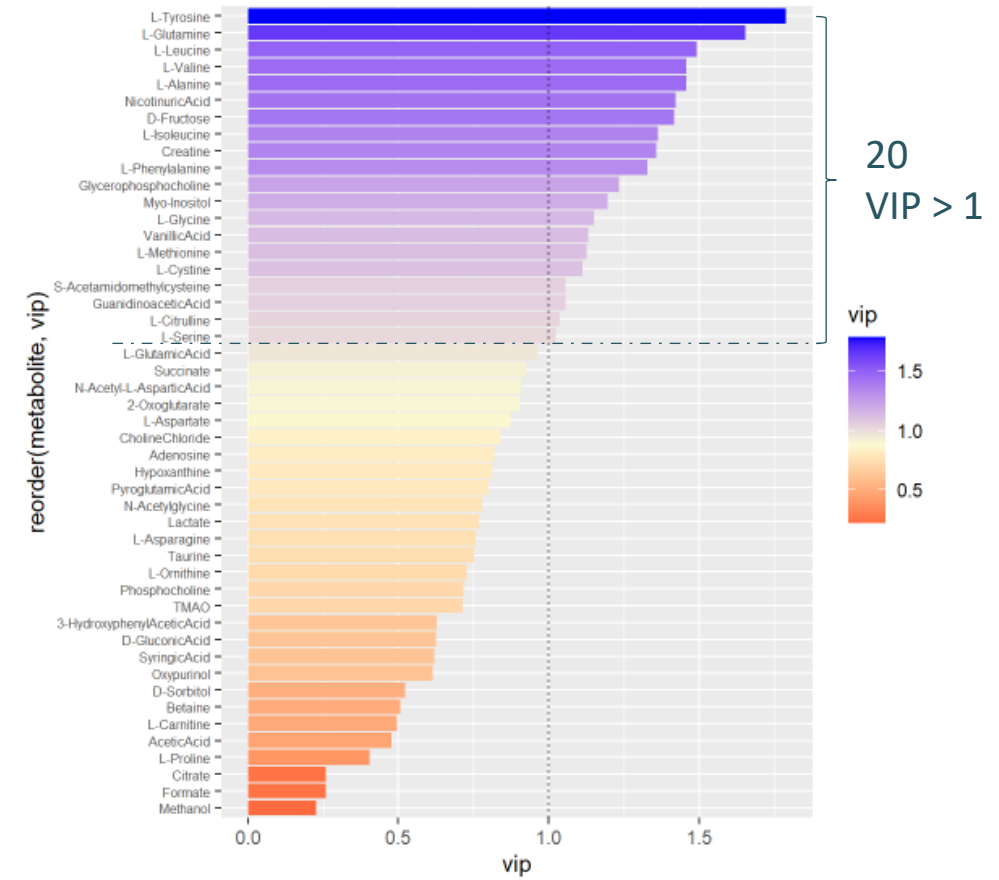
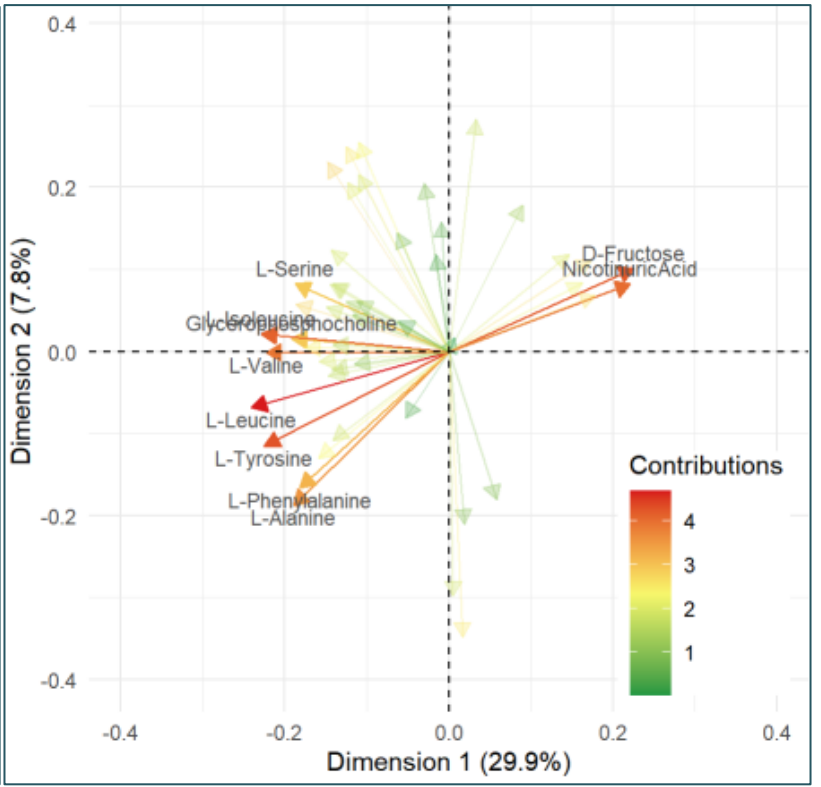
Analysis: PCA

Analysis: PLS-DA

Individual plot



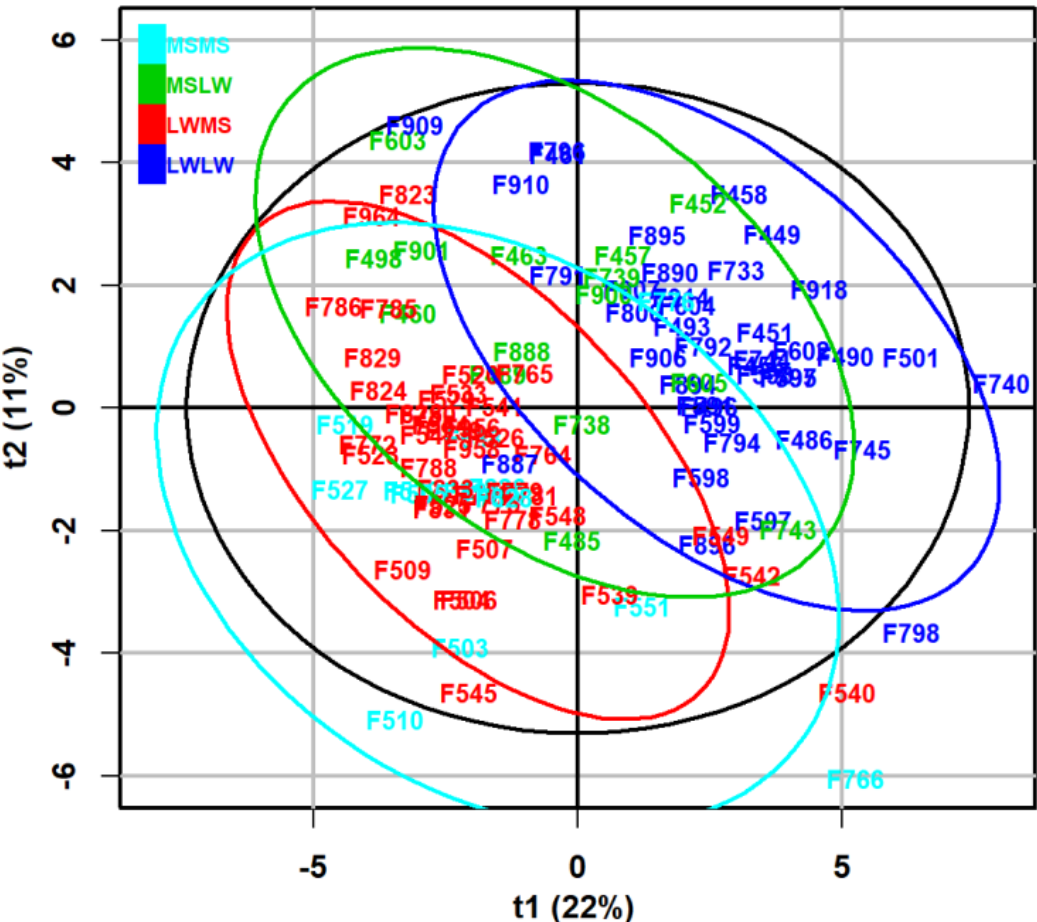
Variable plot



# ➤ Comparison of the placental metabolome between genotypes

D90

Analysis: PLS-DA



19 metabolites  
VIP > 1

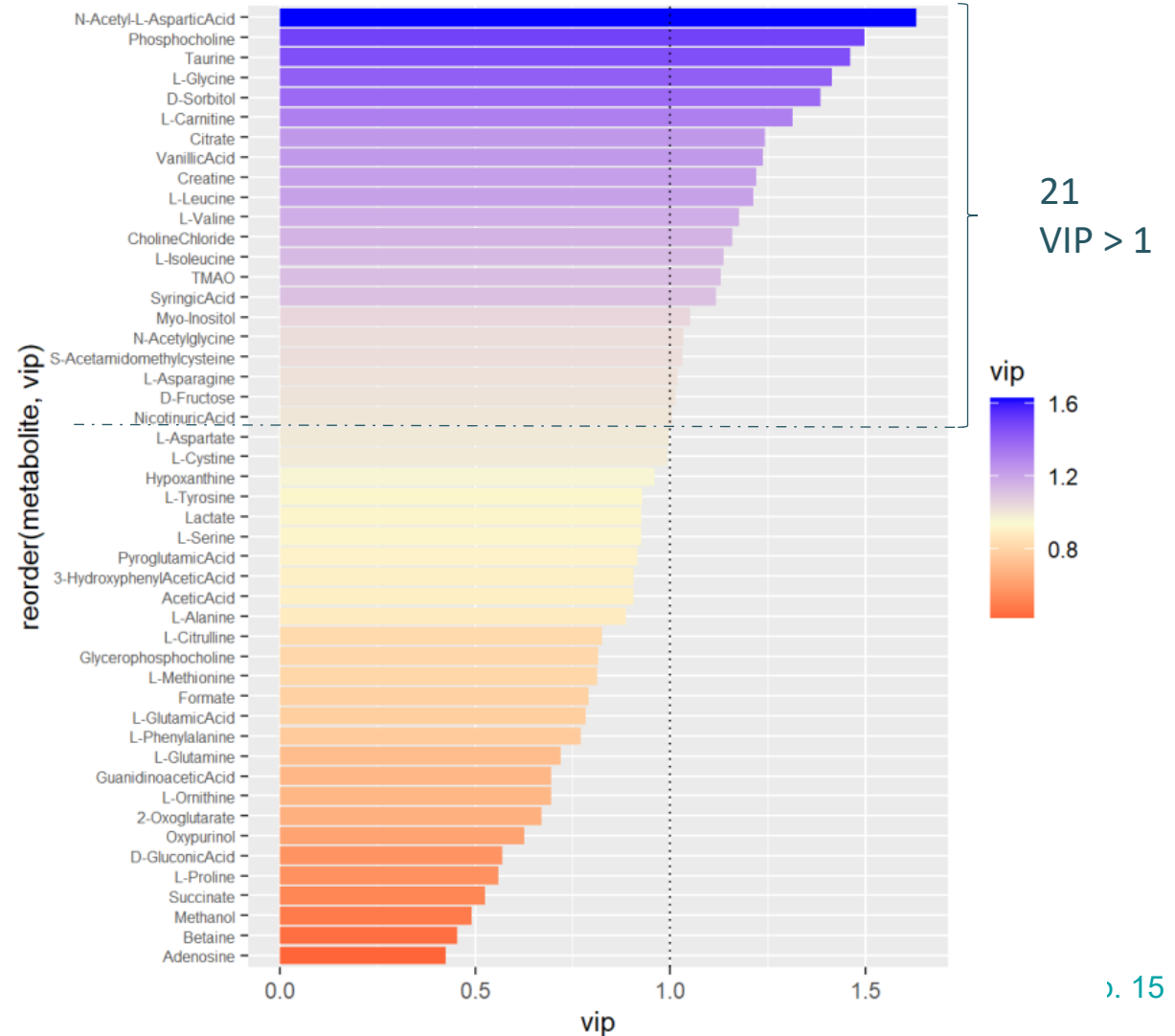
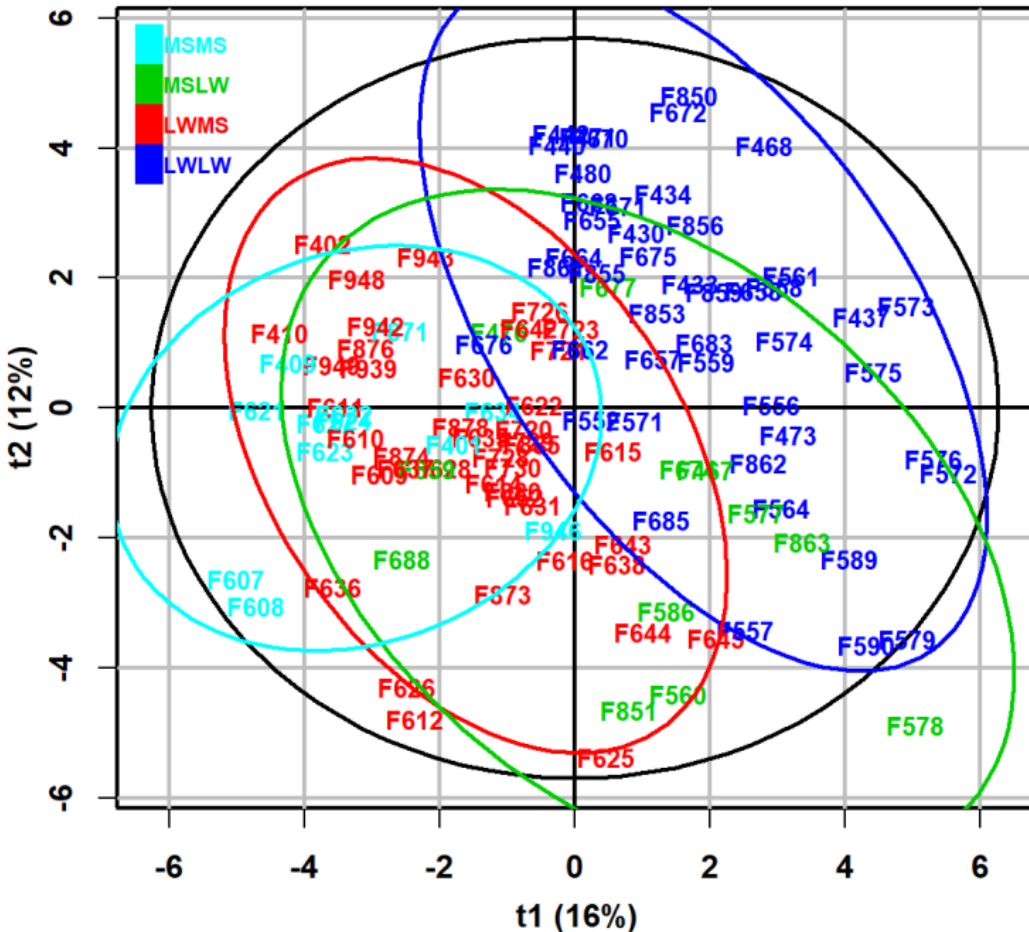
- 2-oxoglutarate
- D-Fructose
- Glycerophosphocholine
- GuanidinoaceticAcid
- L-Alanine
- L-Citrulline
- L-Cystine
- L-Glutamine
- L-Glycine
- L-Isoleucine
- L-Leucine
- L-Methionine
- L-Phenylalanine
- L-Tyrosine
- L-Valine
- Myo-Inositol
- NicotinuricAcid
- S-Acetamidomethylcysteine
- VanillicAcid

# ➤ Comparison of the placental metabolome between genotypes



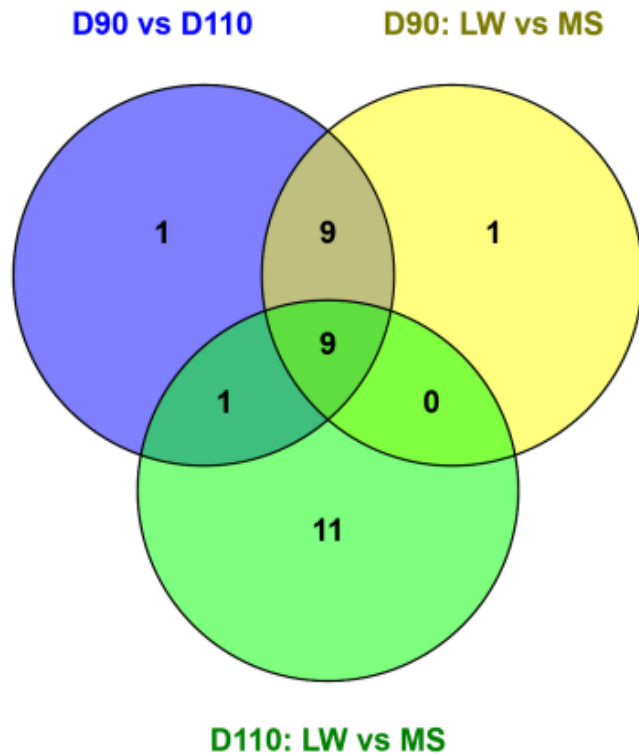
Analysis: PLS-DA

D110



## ➤ Characteristics of a placenta associated with higher maturity

Venn diagram of discriminant metabolites previously identified

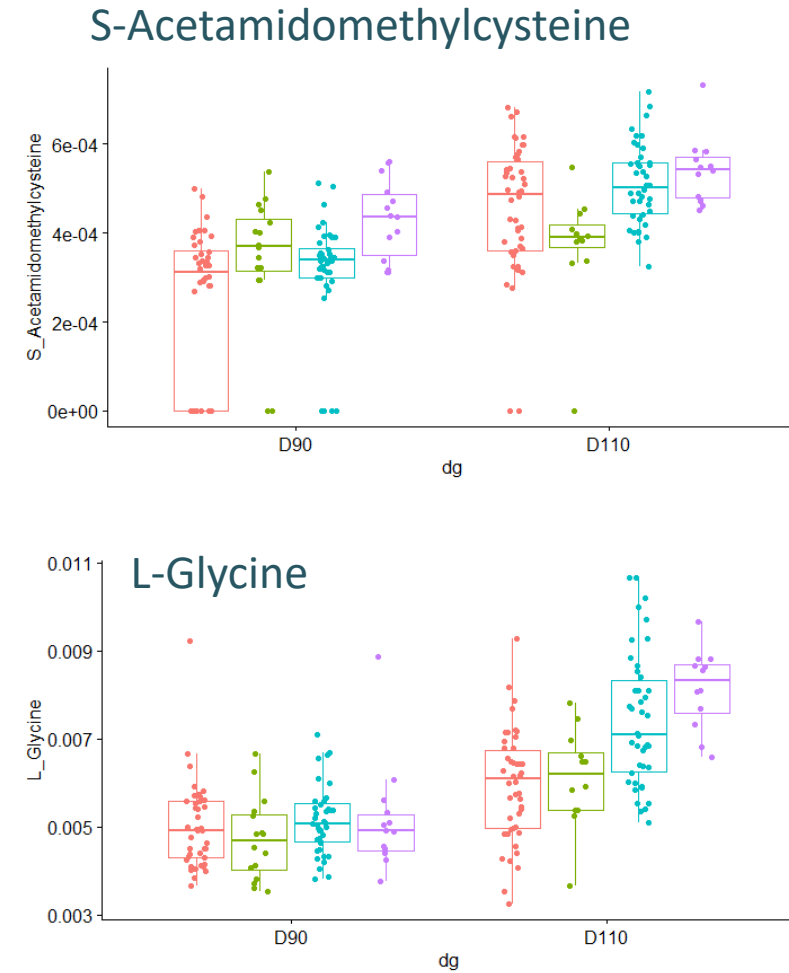
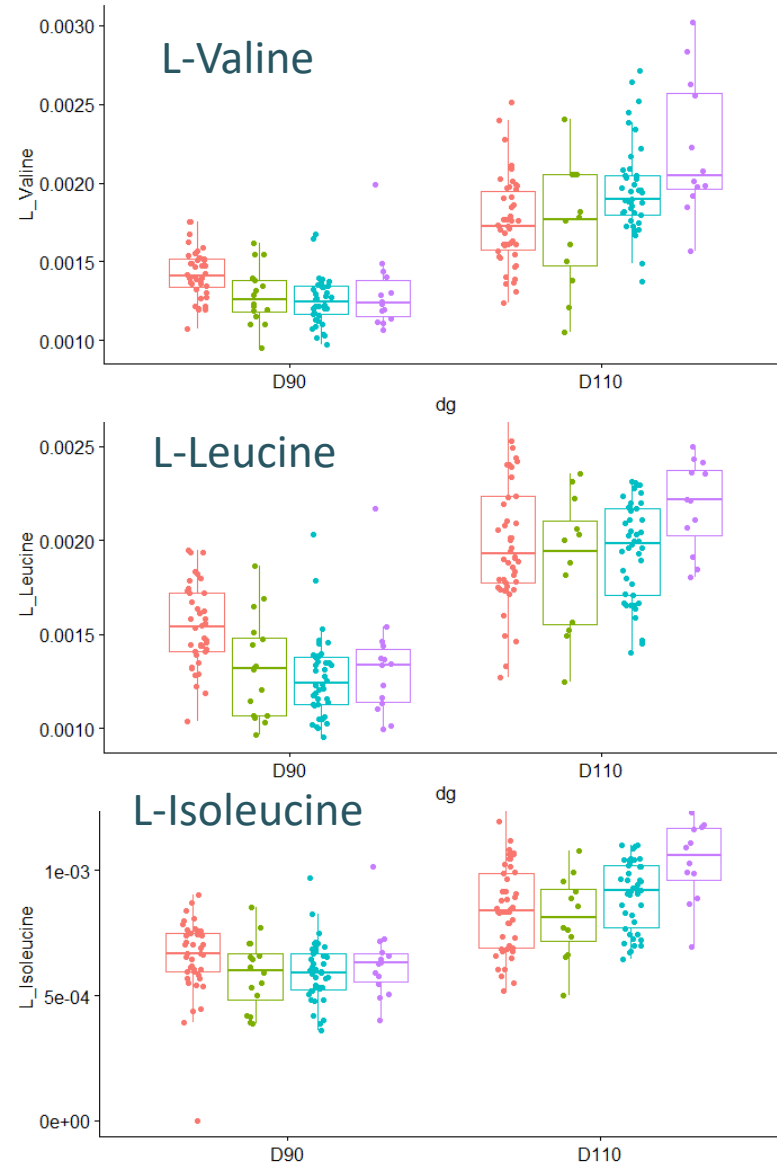
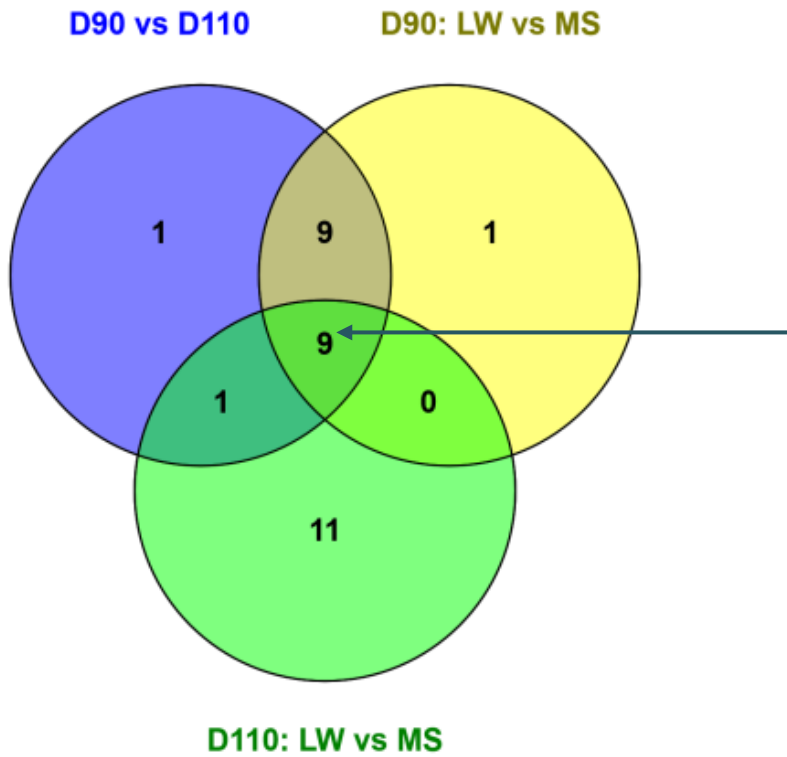






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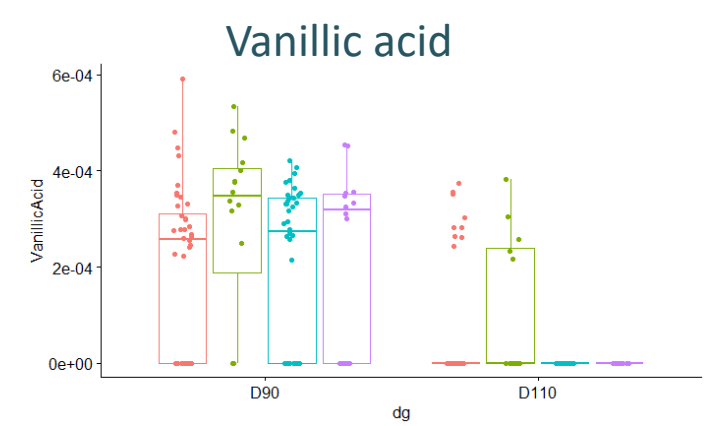
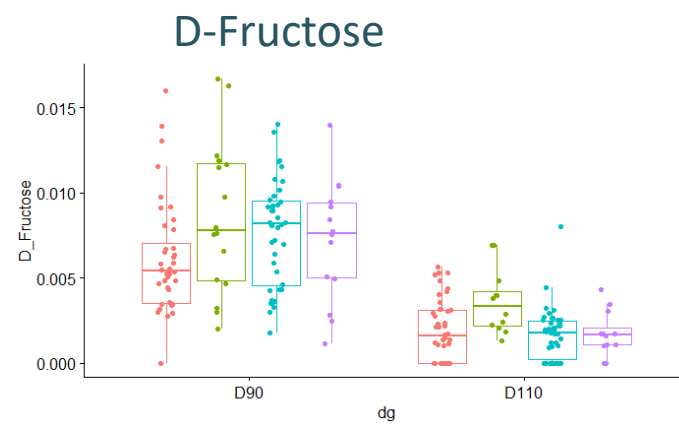
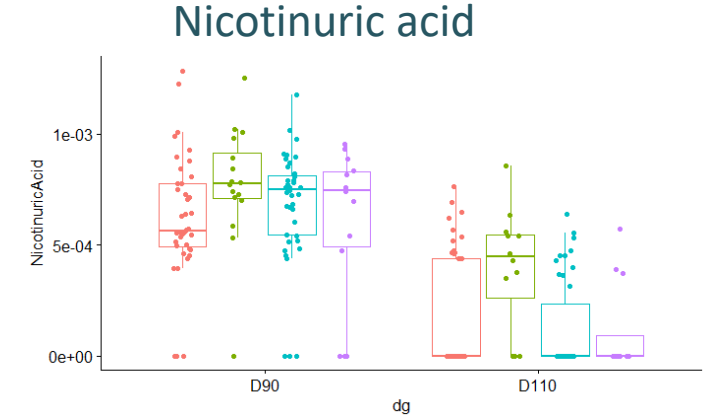
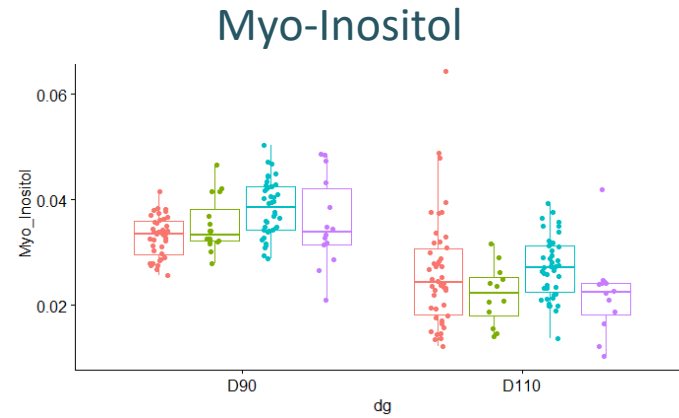
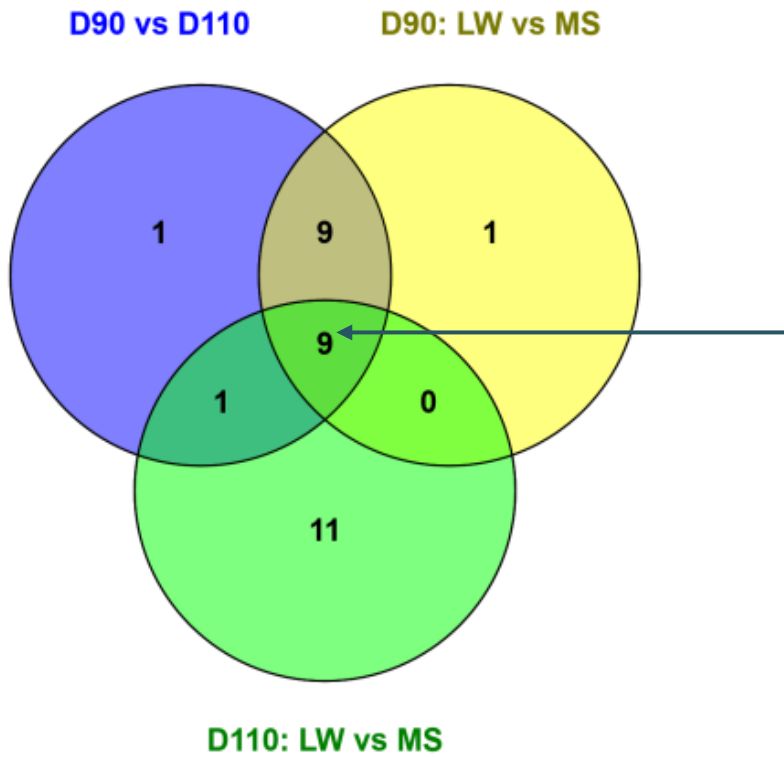
Venn diagram of discriminant metabolites previously identified





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Venn diagram of discriminant metabolites previously identified



FG LWLW MSLW LWMS MSMS

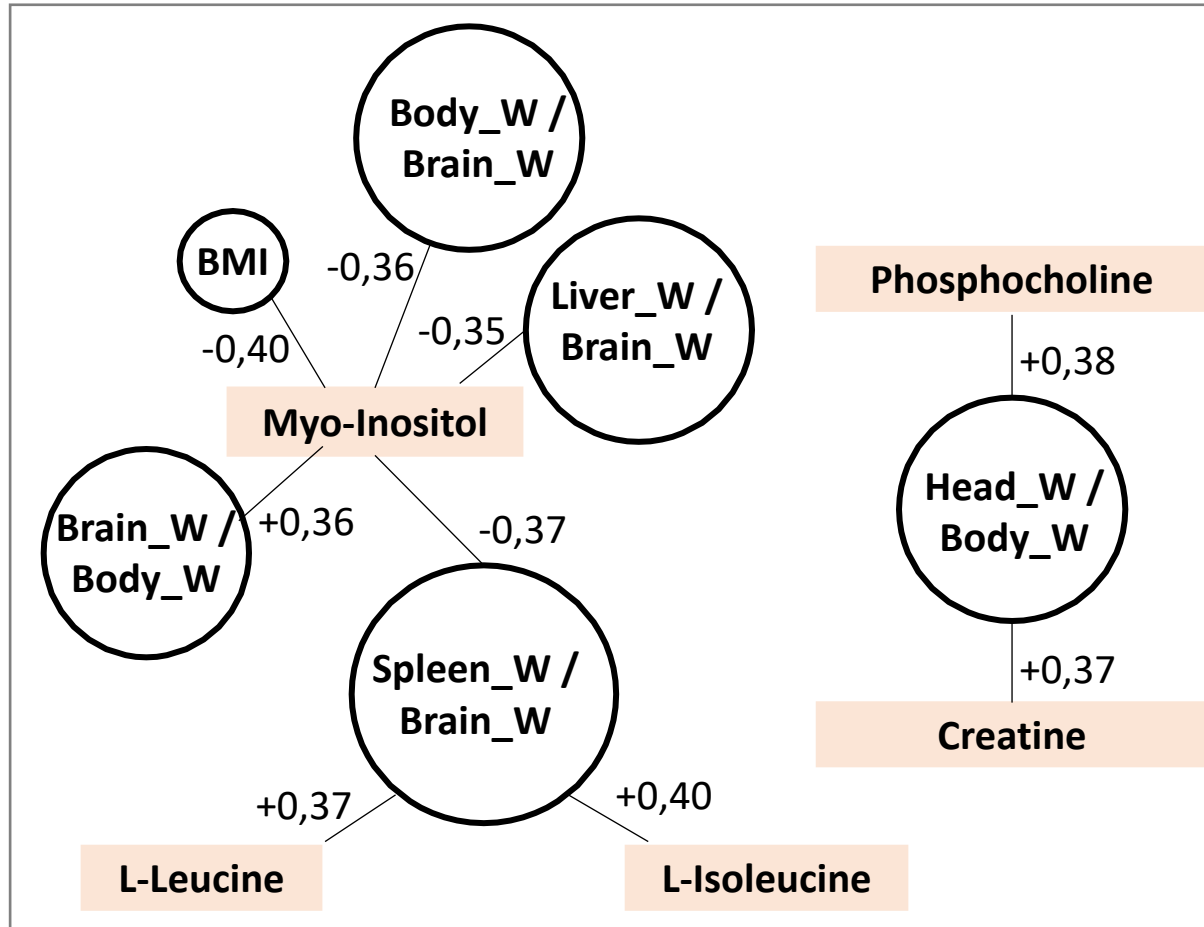


INRAE



## ➤ Correlations between fetus phenotypes and placental metabolites

Bi-partite plot





## > Conclusion and perspectives

A new step in the knowledge of the role of placenta in piglet maturity development

### **Metabolites associated with intra-uterine fetal growth restriction (IUGR):**

- Myoinositol : confirmation in both humans (Dessi & Fanos, 2013; Barberini et al., 2014) and pigs (Nissen et al. 2011; Lefort et al. 2020)
- Fructose: confirmation in pigs (Lefort et al. 2020)
- Nicotinuric acid and vanillic acid: new potential biomarkers of IUGR

**Further analyses are required to confirm their potential role in IUGR**

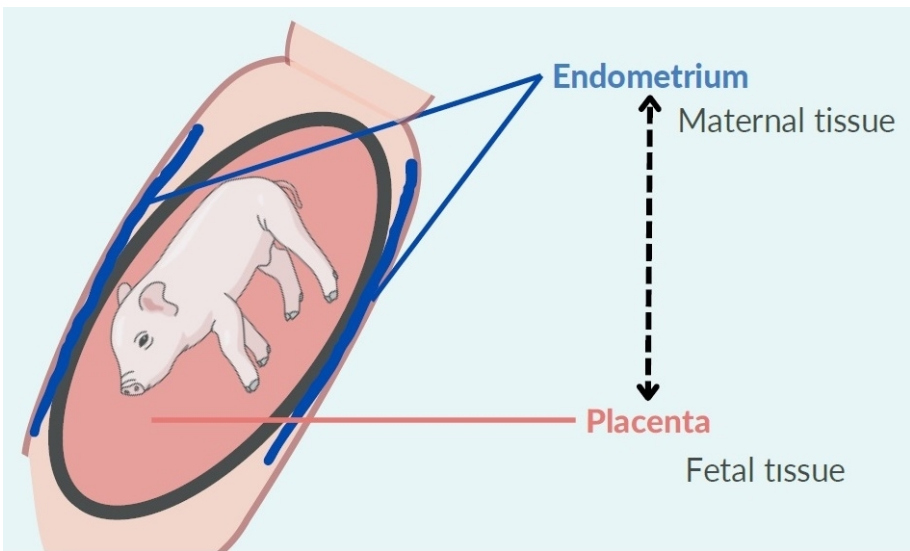
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# ➤ Conclusion and perspectives

Foeto-maternal interactions

EAAP / Session 37



Placenta  
Metabolomics

➤ 1H-NMR metabolomic study of Large White and Meishan pigs in late gestation: part 1- fetal placenta  
J. Guibert, A. Imbert, N. Marty-Gasset, L. Gress, C. Cécile, L. Canario, Y. Billon, A. Bonnet, L. Liaubert and CMD Bonnefont

Endometrium  
Metabolomics

**1H-NMR metabolomic study of Large White and Meishan pigs in late gestation: part 1- fetal placenta**

**Context:** Genetic selection applied in sows led to a reduction of piglet mortality. Mortality is associated to a later survival age after birth. Understand precise causes early in the life of gestation (from day 90 to the term at day 114) and may help improve maternal resources.

**Endometrium & Placenta:** No interface for the regulation of the maternal-fetal exchange between mother and fetus, making separate mechanisms.

**Objectives:** Addressing the trade-off between fetal growth and neonatal survival, targeting late gestation and genetic influences of the foeto-maternal interface in endometrial view.

**Material and Methods:** Experimental protocol (LW and Meishan) and 1H-NMR metabolomic analysis.

**Results:** 1. 1607 differential transcripts (DEGs) in the placenta for days of gestation (D05, D90, D114). 2. Changes in placental gene expression according to days of gestation (D05, D90, D114). 3. Change in placental gene expression, according to H0-1548 DEGs.

**Conclusion:** The study shows an important transcriptional change in the end of gestation in placenta and endometrium genes involved in nutrient transfer. This may be related to the trade-off between fetal growth and neonatal survival. Further studies should investigate the role of the endometrium in the foeto-maternal interface.

Placenta  
Transcriptomics

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Transcriptomics

**Transcriptome of the foeto-maternal interface in pigs in late gestation: part2- fetal endometrium**

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**Material and Methods:** Experimental protocol (LW and Meishan) and transcriptomic analysis.

**Results:** 1. 1607 differential transcripts (DEGs) in the endometrium for days of gestation (D05, maternal breed (MB) and their interactions. 2. Changes in endometrial gene expression according to days of gestation (D05, D90, D114). 3. Important change between maternal breed (MB) 3,724 DEGs (1,422 DEGs with a FDR < 0.05, LW vs MB).

**Conclusion:** The study shows an important transcriptional change in the end of gestation in placenta and endometrium genes involved in nutrient transfer. This may be related to the trade-off between fetal growth and neonatal survival. Further studies should investigate the role of the endometrium in the foeto-maternal interface.

Conclusion Perspectives



1H-NMR metabolomic study of Large White and Meishan pigs in late gestation: Part 1

29/08/2023 – EAAP - Session 37 "Omics and integrative analyses towards understanding inter-organ cross-talk and whole body physiology of livestock"

## > Thanks to

### UE Genesi INRAE

Yvon BILLON  
+ all the staff

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Agnès BONNET  
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Céline BRAVO  
Julien GUIBERT  
Julien HENRY  
Laure GRESS  
Laurence LIAUBET  
Laurianne CANARIO  
Lisa BLUY  
Nassim DUPRAT  
Nathalie MARTY-GASSET

### METATOUL platform

Cécile CANLET

**anr**<sup>®</sup> PORCINET

**anr**<sup>®</sup> CO-LOCATION

Thank you  
for your attention

**INRAE**

1H-NMR metabolomic study of Large White and Meishan pigs in late gestation: Part 1- Fetal placenta

29/08/2023 – EAAP - Session 37 "Omics and integrative analyses towards understanding inter-organ cross-talk and whole body physiology of livestock"



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