COMPARISON OF THE PORCINE GENE EXPRESSION PROFILES BETWEEN THE MAJOR SITES FOR LIPID METABOLISM, LIVER AND FAT

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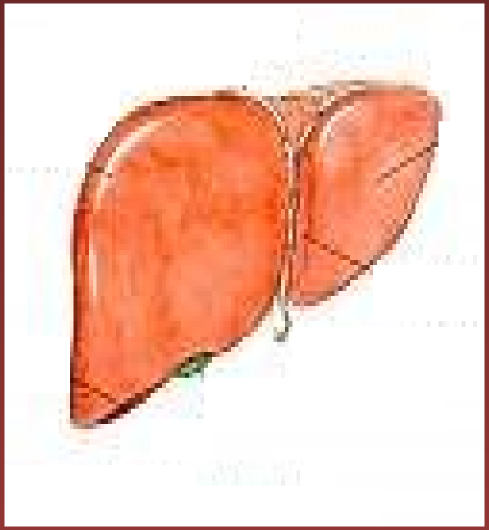
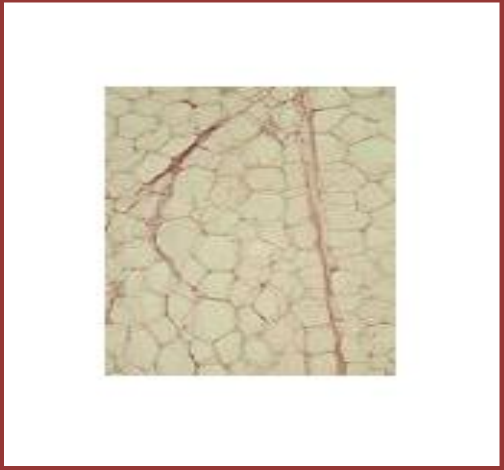


Liver plays vital roles in vertebrates

Glycogen storage
Decomposition of red blood cells
Protein synthesis
Hormone production
Detoxification
…
Lipid metabolism



Adipose tissues are not only stores of triacylglycerols

Substances secretion
Hormones secretion

Lipid metabolism

The main sites for fat metabolism in animals:

Liver and Adipose tissue

However,

there are important differences among species:

Humans /Rodents / Birds	Dogs/ Cats/ Cattle/Sheep/ Pigs
De novo fatty acid synthesis occurs mainly in liver	*De novo* fatty acid synthesis occurs mainly in adipose tissues

Important aspect
mainly when applying results from animal models to human

Aim

To investigate the lipid metabolism profile of liver and adipose tissue in pig

- Comparison of gene expression patterns using microarrays

Material and Methods

Experimental design

20 Iberian pigs ; 7 months old

10 males	10 females
5 High feeding level	5 High feeding level
5 Low feeding level	5 Low feeding level

Gene expression analyses:

Affymetrix GeneChip Porcine ExpressionArray
(24.123 probes ≈ 20.200 genes)

8 individuals selected for hybridizations
(hepatic and adipose samples)

qRT-PCR validation
(hepatic and adipose samples from 17 pigs)

Material and Methods

Statistical analyses:

GCRMA normalization (BRB - Simon & Peng Lam)

ANOVA . Bioconductor (BRB - Simon & Peng Lam)

Model:

$$Y_{jkln} = mean + gender_j + feeding\ level_k + tissue_l + error_n$$

False Discovery Rate < 0. 01 (Benjamini y Hochberg, 1995)

Annotation and biological interpretation

Affymetrix annotation file + NetAffx tool

David database (http://david.abcc.ncifcrf.gov/)

Results: Global expression differences $p < 10^{-3}$ and FDR<0.01

Number of DE probes by	
Gender	14
Feeding level	0
Tissue	7847

Number of DE probes by	
Interaction gender/tissue:	55
Interaction feeding level/tissue	0
Interaction gender /feeding level	0

Results: Expression differences conditional on gender

14 DE probes by gender effect

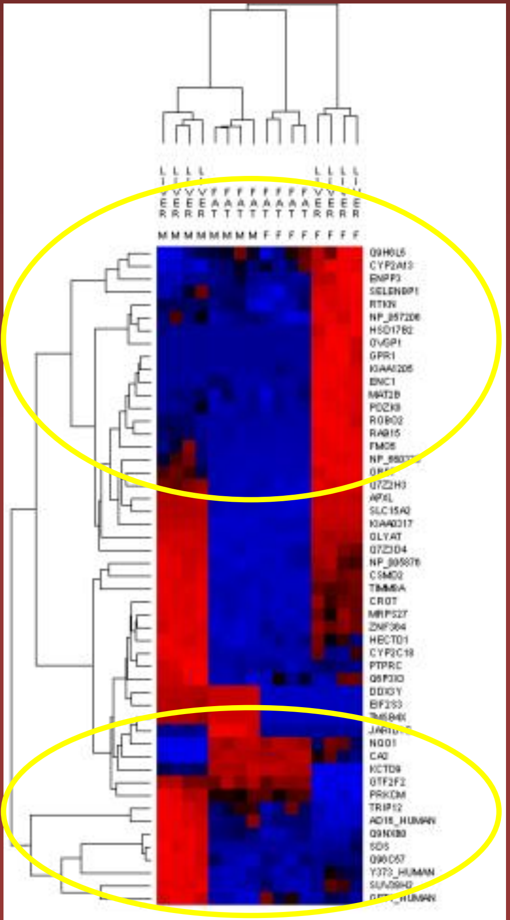
EIF1AY
PTPRM
NP_950252
DDX3Y
XIST
HIPK2
CLOCK
LPHN2
EIF2S3
TMSB4X
UTY
EIF2S3
LRP1B
DDX3X

12 genes
consistently differentially expressed between genders across tissues

(Ferraz et al., 2008 and Yang et al., 2006)

Results: Expression differences conditional on gender

Several other genes (55 interactions gender/tissue) differentially expressed between genders in liver or adipose tissue



Results: Expression differences conditional on tissue

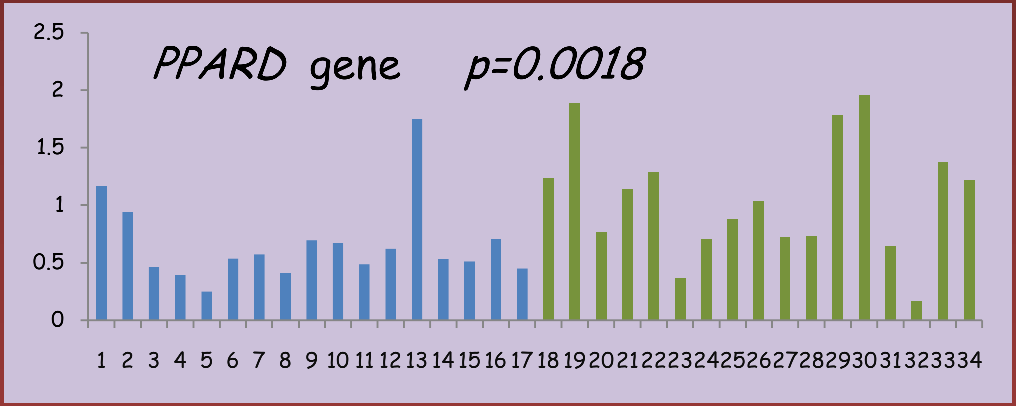
Number of DE probes by	
Tissue	7847

Biological interpretation using *DAVID database*

Probes related with lipid metabolism	334
Genes related with lipid metabolism	243

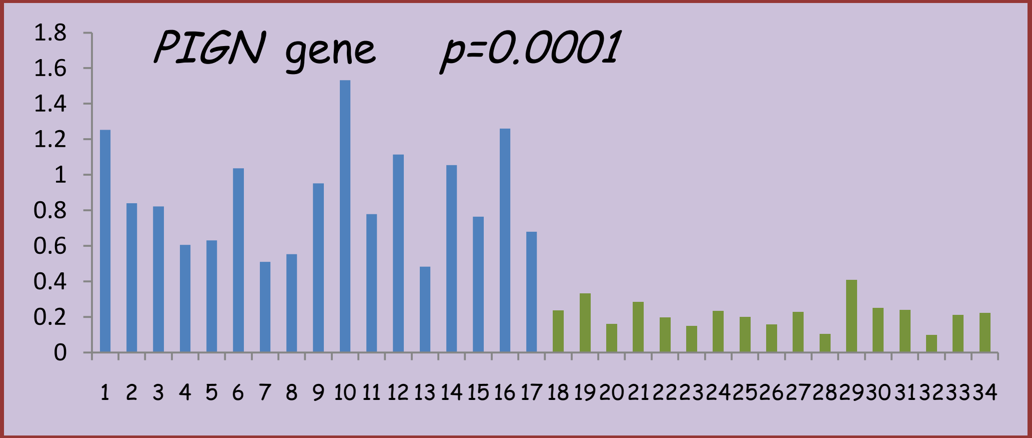
		Fold change				
Tissue	Upregulated probes	1.3-2.5	2.5-5	5-10	>10	Upregulated genes
Adipose	148	59	36	17	37	113
Liver	186	50	52	32	54	130

Results : Validation by qRT-PCR of two genes with the lowest expression differences detected by microarray technology



Adipose tissue 1.75 folds more expression than hepatic tissue

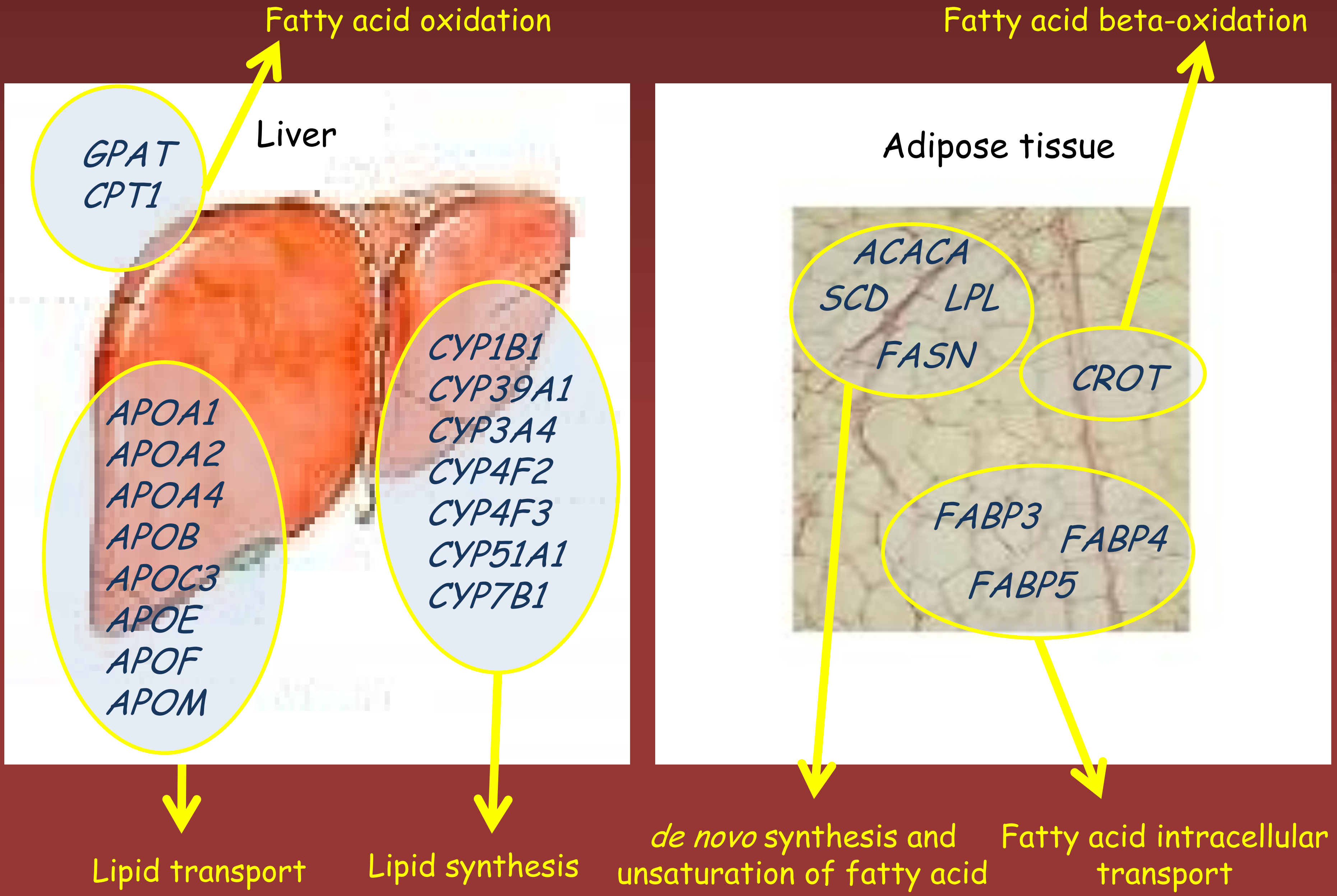
PPARD:
anti-lipid oxidation and anti-adipogenic



Hepatic tissue 3.41 folds more expression than adipose tissue

PIGN:
glycosylphosphatidylinositol (GPI)-anchor biosynthesis

Interpretation of tissue expression differences



Liver

GPAT
CPT1

Fatty acid oxidation

APOA1
APOA2
APOA4
APOB
APOC3
APOE
APOF
APOM

Lipid transport

CYP1B1
CYP39A1
CYP3A4
CYP4F2
CYP4F3
CYP51A1
CYP7B1

Lipid synthesis

Adipose tissue

ACACA
SCD
LPL
FASN

de novo synthesis and unsaturation of fatty acid

CROT

Fatty acid beta-oxidation

FABP3
FABP4
FABP5

Fatty acid intracellular transport

CONCLUSIONS

- High proportion of DE probes (33%) between both tissues

- Besides liver, adipose tissue has potential interest for studying the genetic basis of porcine traits related to fat metabolism

- Differences on transcription level of some key genes support known tissue-specific processes

(*ACACA* and *FASN* - *de novo* fatty acid synthesis in adipose tissue)

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