SABRE

CUTTING EDGE GENOMICS FOR SUSTAINABLE ANIMAL BREEDING

Genes behind good bovine embryo quality

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Food Quality and Safety

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The aim of this study

- Find genes characteristic of good bovine embryo quality
- Use these genes to assess the quality of *in vitro* embryos derived from different IVM media
- Are there marker genes for good embryo quality applicable for the both *in vivo* and *in vitro* embryos?

Background

- Genes guide embryo development
- Known differences in gene expression between different quality embryos
- *In vitro* production affects embryonic gene expression

Materials and methods

- *In vivo*:
  - Biopsies from embryos resulting in calf delivery (CD) and no pregnancy (NP)

    -> Microarray analysis with Blue chip

    -> Validation of candidate genes with qPCR
- *In vitro*:
  - Embryo production by using three different IVM media => aim to find replacement for serum
  - QPCR study by using validated genes from *in vivo* study, and other quality-related genes

Microarray: *in vivo* candidate genes

Selected up-regulated genes in CD embryos

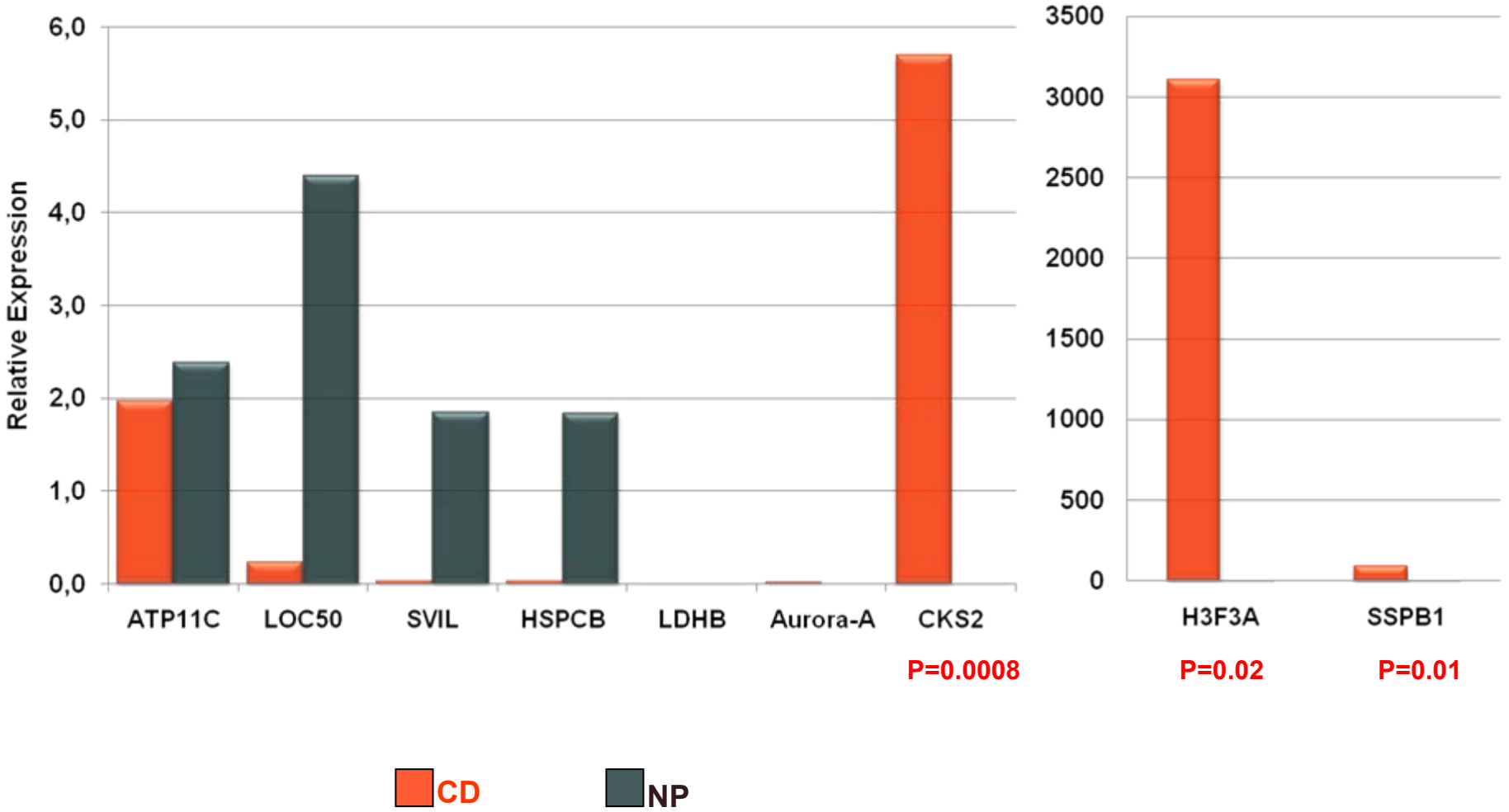
Gene name	Fold change
Lactate dehydrogenase B (LDHB)	8,0
Similar to single-stranded DNA binding protein 1 (SSPB1)	3,2
Aurora-A	2,8
H3 histone, family 3A (H3F3A)	2,4
CDC28 protein kinase regulatory subunit 2 (CKS2)	1,5

Selected down-regulated genes in CD embryos

Gene name	Fold change
Supervillin (SVIL)	2,8
Homo s. mRNA for ATPase, Class VI, type 11C (ATP11C)	2,6
mRNA for aromatase cytochrome P450 pseudogene (LOC50)	2,4
Heat shock 90kDa protein 1, beta (HSPCB)	2,0

Differentially expressed genes were identified by SAM at a false discovery rate (FDR) of ≤ 5% and $P \leq 0.05$

qPCR validation of selected *in vivo* genes

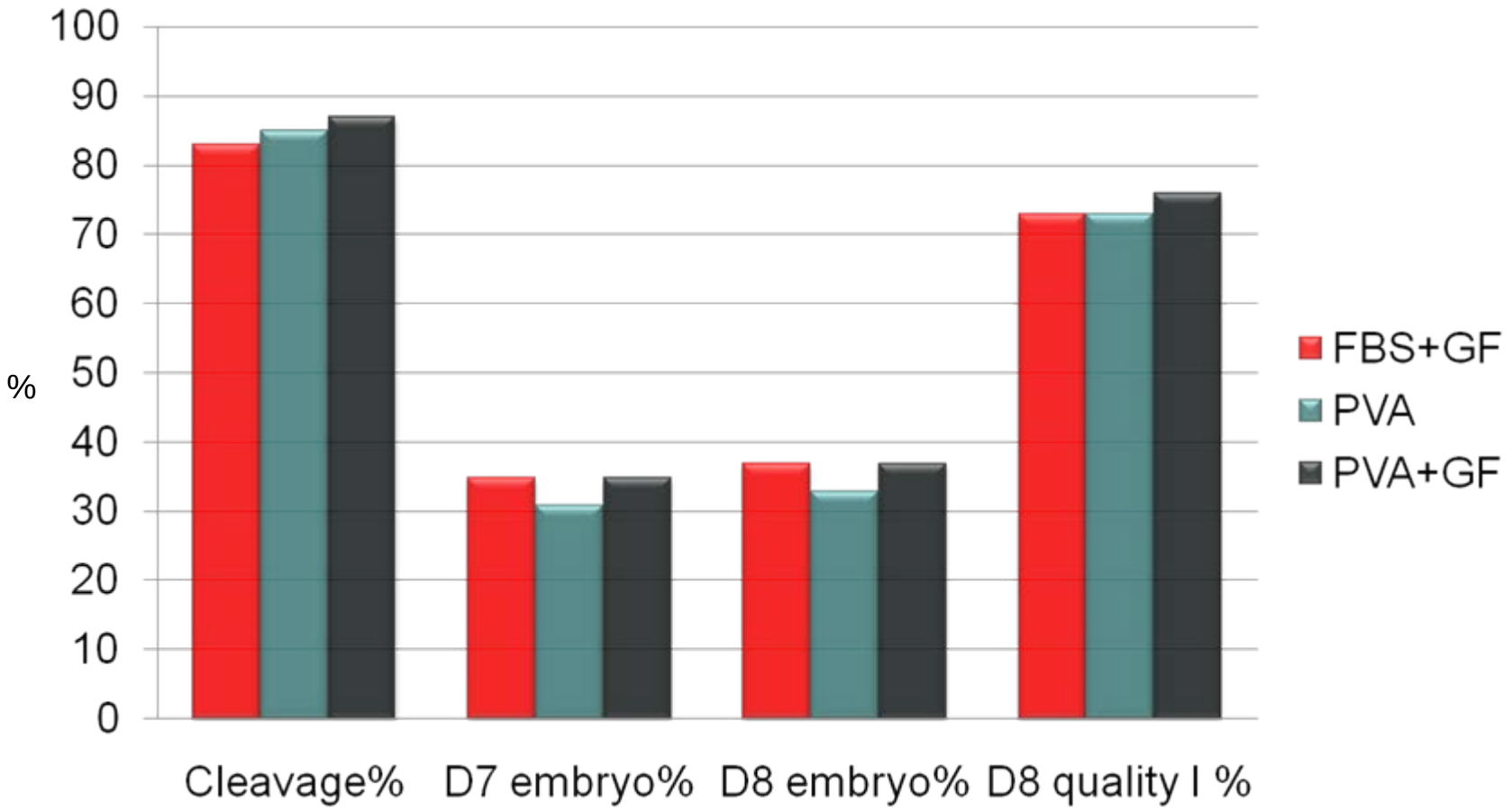


In vitro embryo production

- Oocyte maturation in three different media
  - FBS+GF*
  - PVA+GF*
  - PVA

    *GDF9, BMP15, IGF-I, EGF,BDNF, FGF8, LEP
- Embryo culture in G1/G2 media (Vitrolife)
- Measurements:
  - Embryo cleavage% on day 3
  - Embryo yield and morphological quality on day 7 and 8

Results of embryo production

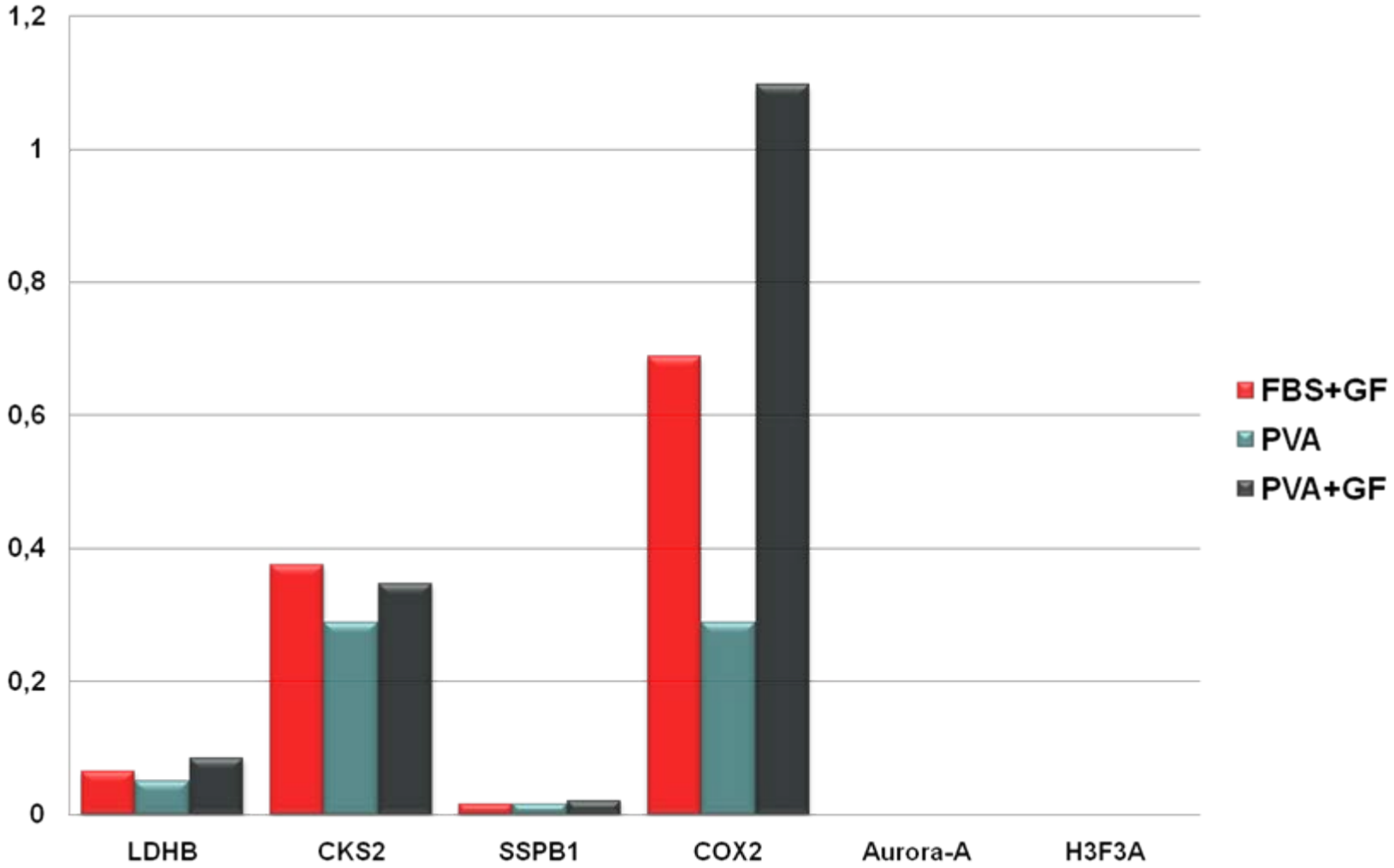


qPCR study of *in vitro* embryos

- Genes validated from *in vivo* material
  - Up-regulated: CKS2, SSPB1, H3F3A, LDHB, Aurora-A

- Genes known to be expressed in good quality *in vitro* embryos (El-Sayed et al,. 2006)
  - Up-regulated: COX2

qPCR results of *in vitro* embryos



P=0.01

Discussion I

- Genes expressed in good quality embryos are different between *in vivo* and *in vitro*
- CKS2 a possible good quality embryo marker for *in vivo* embryos

Discussion II

- FBS can be replaced with PVA without compromising embryo yield or morphological quality
- However, embryos derived from PVA IVM medium have lower COX2 expression levels

  => possibly indicating lower quality
- Similar gene expression of COX2 between FBS and PVA group is gained by adding GFs into PVA medium

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