

Transcriptome of Porcine PBMCs over Two Generations Reveals Key Genes and Pathways Associated with Variable Antibody Responses post PRRSV Vaccination

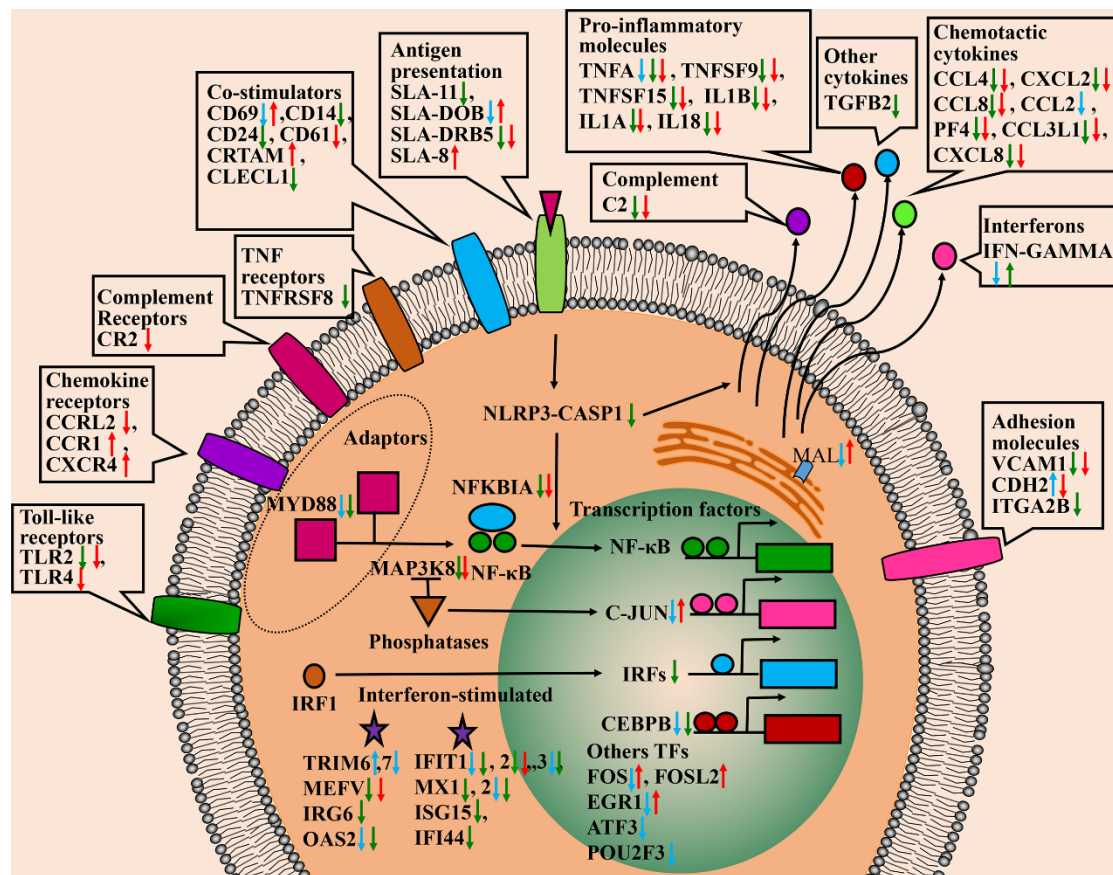
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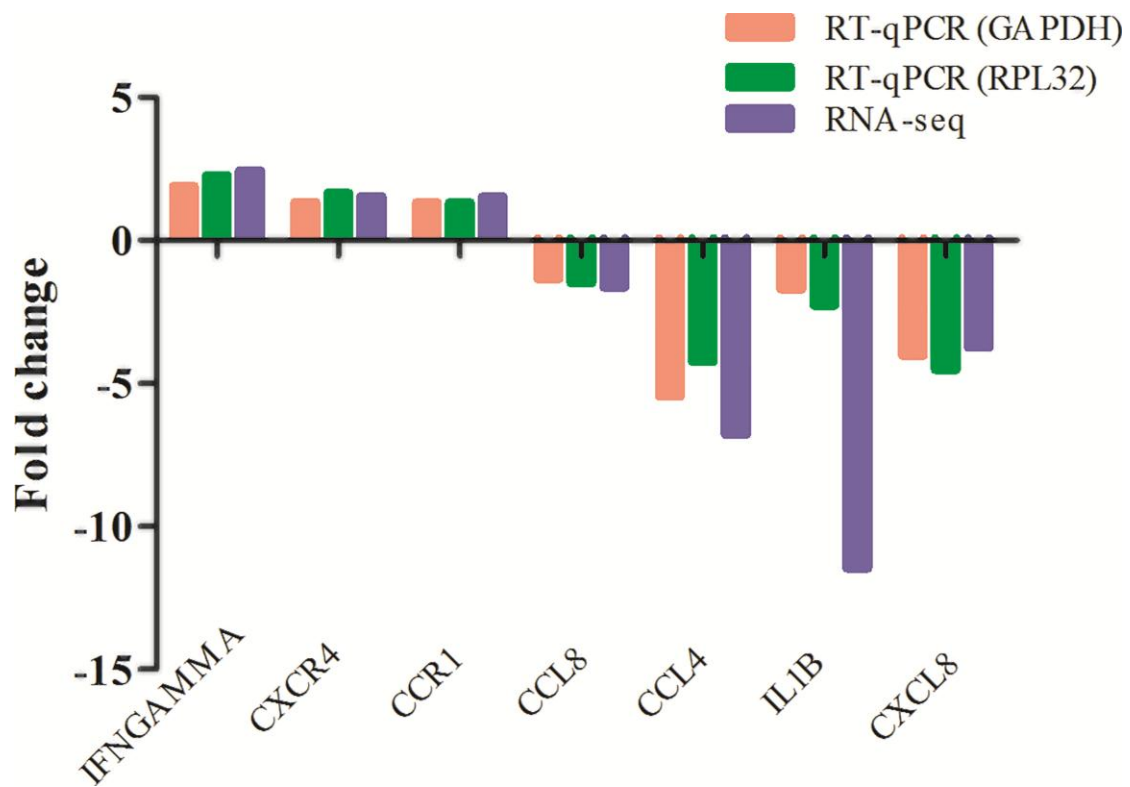
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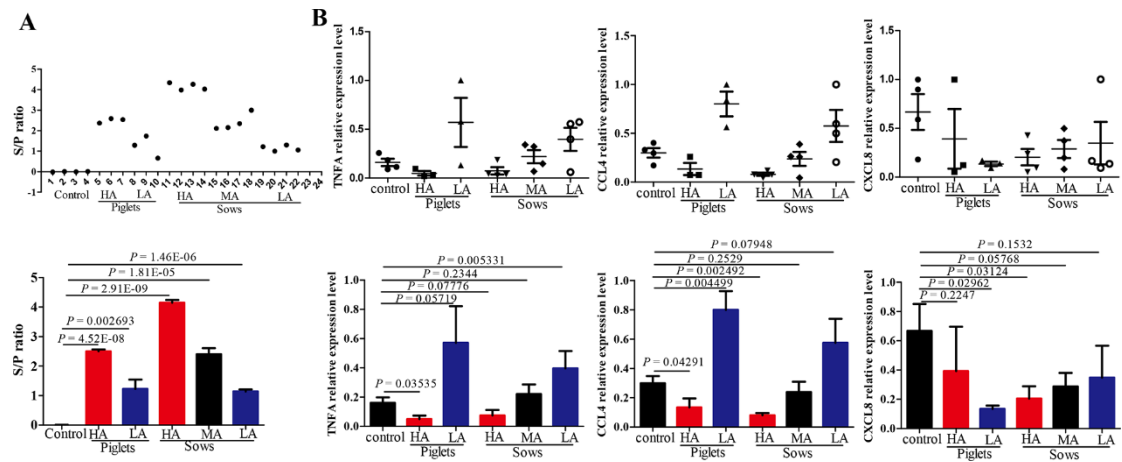
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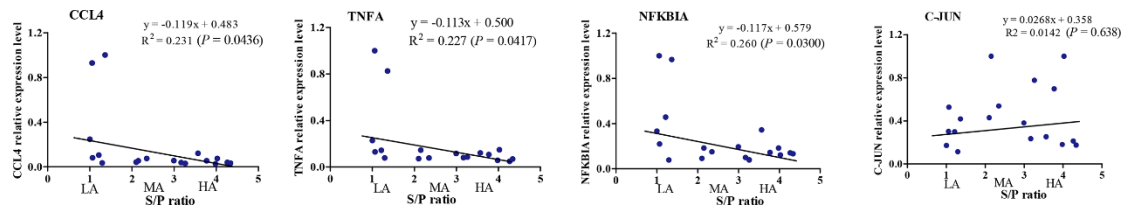
Supplementary Figure S1. Significant immune responses of porcine PBMCs at 35 days after PRRSV-MLV vaccination. DEGs of host response and their functions inside and outside PBMCs. Genes marked with blue, green, and red color arrows represent HA versus MA, HA versus LA, and MA versus LA, respectively. Genes marked with a downward arrow were downregulated, and the others were upregulated.



Supplementary Figure S2. RT-qPCR validation of RNA-Seq data. *GAPDH* and *RPL32* genes were as internal controls separately.



Supplementary Figure S3. Immune responses and gene expression quantification of naive piglets, vaccinated piglets, and pregnant sows to PRRSV-MLV vaccination. (A) S/P ratio among controls ($n = 4$), vaccinated piglets ($n = 3$), and sows ($n = 4$). (B) Expression levels of *TNF- α* , *CCL4*, and *CXCL8* genes in controls ($n = 4$), vaccinated piglets ($n = 3$), and sows ($n = 4$) with RT-qPCR. P means significant level of t -test.



Supplementary Figure S4. Regression analysis of divergent PRRSV-MLV-specific antibody levels (x-axis) with gene expression levels (y-axis) in sows and piglets. HA: high antibody group ($n = 8$); MA: median antibody group ($n = 4$); LA: low antibody group ($n = 6$).

Supplementary Table 1. The basic statistics of sequencing reads aligned to genome for different pigs.

Sample ID	Total tags (Million)	Mapping rating %
H37	37.8	82.1%
H48	41.58	81.5%
H78	42.34	83.9%
H90	37.5	83.2%
M35	39.47	85.7%
M44	40.14	81.9%
M49	38.92	82.6%
M91	42.16	81.2%
L39	41.12	81.8%
L45	40.44	85.1%
L51	37.28	80.7%
L63	38.22	83.4%

Supplementary Table 1. Primer sequences for qRT-PCR validation

Gene name	Primer sequence (5'-3')	Amplified size (bp)	Annealing temperature (°C)
<i>CCL4</i>	F:CTGCTGCTTCACATACACCGT R:CAGACCTGCCTGCCCTTTT	123	60
<i>CCL8</i>	F: CAAGAATCACCAACAGCCAGT R: CAGTCCAGGTAGGAAGGTTCAA	164	58
<i>CCR1</i>	F:CAGAAACAAAGACTTCGTGGACA R: CACAGGTCAAAGGAAATGGTT	96	58
<i>CXCL8</i>	F: TAGGACCAGAGCCAGGAAGA R: AATTTGGGGTGGAAAGGTGT	191	58
<i>CXCR4</i>	F: CGGGTTC CGTATATTC ACTTCA R: ACCCACTATGCCAGTTAAGAAGAT	163	60
<i>IFN-γ</i>	F:CCTAAATGGTAGCTCTGGGAAAC R:ATGAGTTCACTGATGGCTTTGC	99	60
<i>IL1B</i>	F:CTGTTATTTGAGGCTGATGGC R:TCATCGGCTCCTCCTTTGC	166	60
<i>IFNα</i>	F:GGCACAAATGAGGAGAATC R:CACAGCCAGGATGGAGTC	307	60
<i>TLR3</i>	F:AAAATCTCCAAGAGCTTCTATTAGCAA R:TTGTATTTGATTTGATGACA ACTCTAATCTTT	119	60
<i>RPL32</i>	F:CGGAAGTTTCTGGTACACAATGTAA R:TGGAAGAGACGTTGTGAGCAA	94	58-60
<i>GAPDH</i>	F:ACTCACTCTTCTACCTTTGATGCT R:TGTTGCTGTAGCCAAATTCA	100	58-60