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Functions and related mechanisms of lncRNA SULT1C2A in congenital scoliosis

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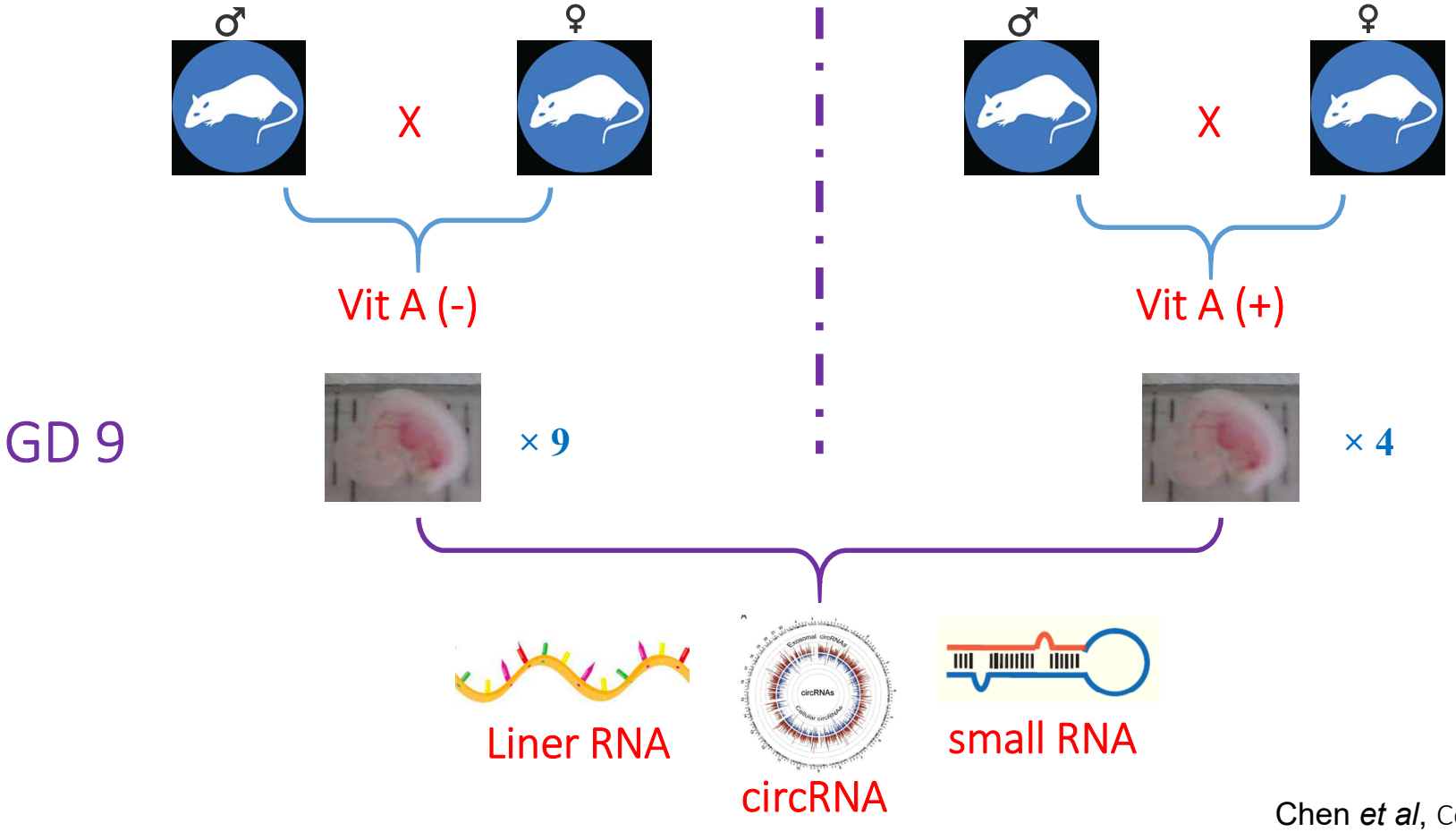
Transcriptome Assembly



Chen *et al*, Cell Physiol Biochem. 2018

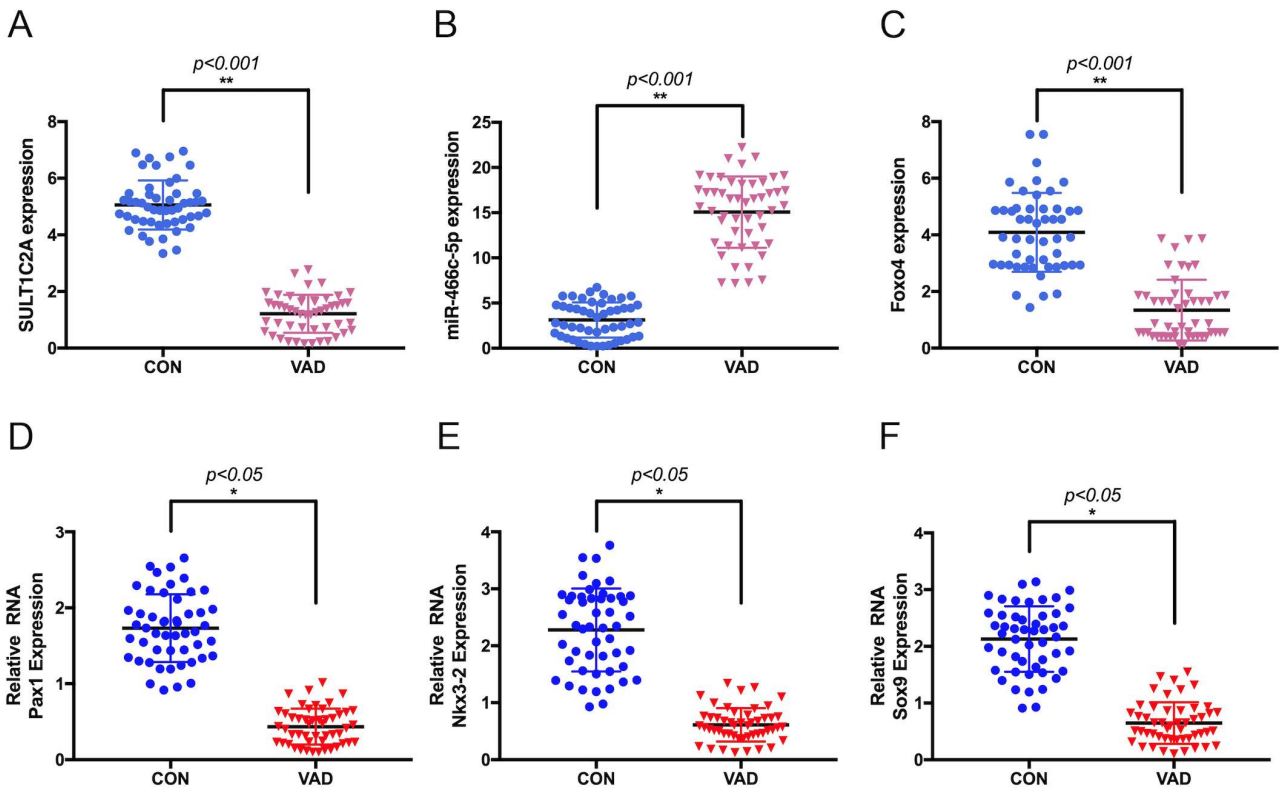
Co-expression network construction

NONRATG027649.1
chromosome 9 (4621424 -4624425 [+])

- **intron 4**
- **Length：1,828 bp**
- **Gene：Sult1c2a**

Weighted gene co-expression network analysis (WGCNA)

Expand Sample Verification

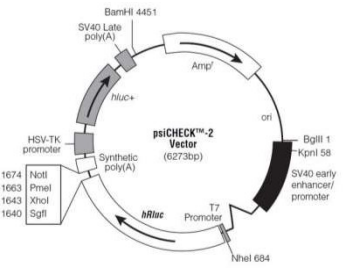


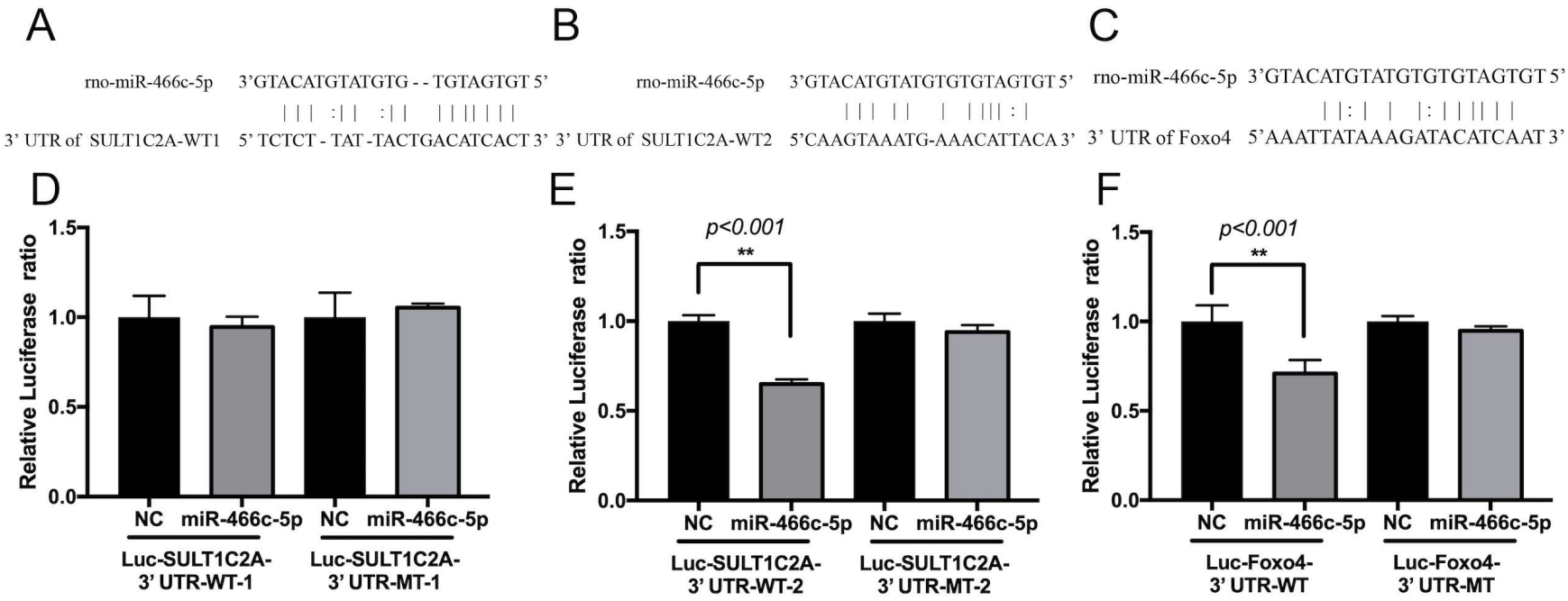
GD 9 , n=50

**P＜0.05 **P＜0.01*. VAD *vs* Control

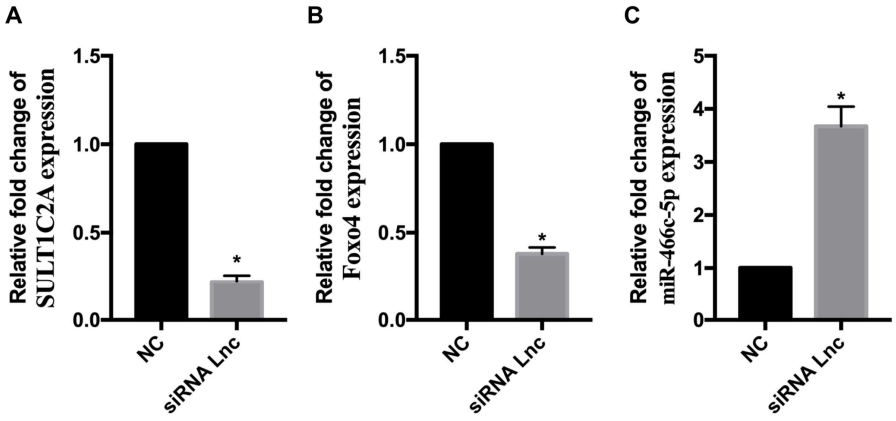
Gene	Primer Sequence (5'- 3')
SULT1C2A-F	TGGCCCAGAATGAGAGGTTTGATGA
SULT1C2A-R	CGGTGTCACAGTCCTTGGCATTTAC
FOXO4-F	GGCGGCAAGGGTGGCAAGG
FOXO4-R	CCGGCCTCATTGGGGACAGC
Pax1-F	CAATGCCATCCGCCTACGAAT
Pax1-R	AGAGACCCGCAGTTGCCTA
Nkx3-2-F	AGCGCCGCTTTAACCATCA
Nkx3-2-R	GCGTTTGGTCTTGTAGCGAC
Sox9-F	AGTACCCGCATCTGCACAAC
Sox9-R	ACGAAGGGTCTCTTCTCGCT
GAPDH-F	TGGGGTGATGCTGGTGCTGAGTAT
GAPDH-R	AGCGGAAGGGGCGGAGATGAT
rno-miR-466c-5p-F	TGTGATGTGTGTATGTAC
U6-F	CTCGCTTCGGCAGCACA

Luciferase Reporter Assays

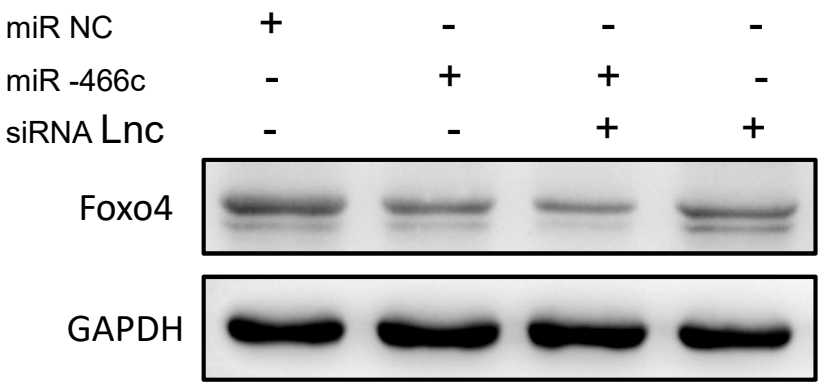


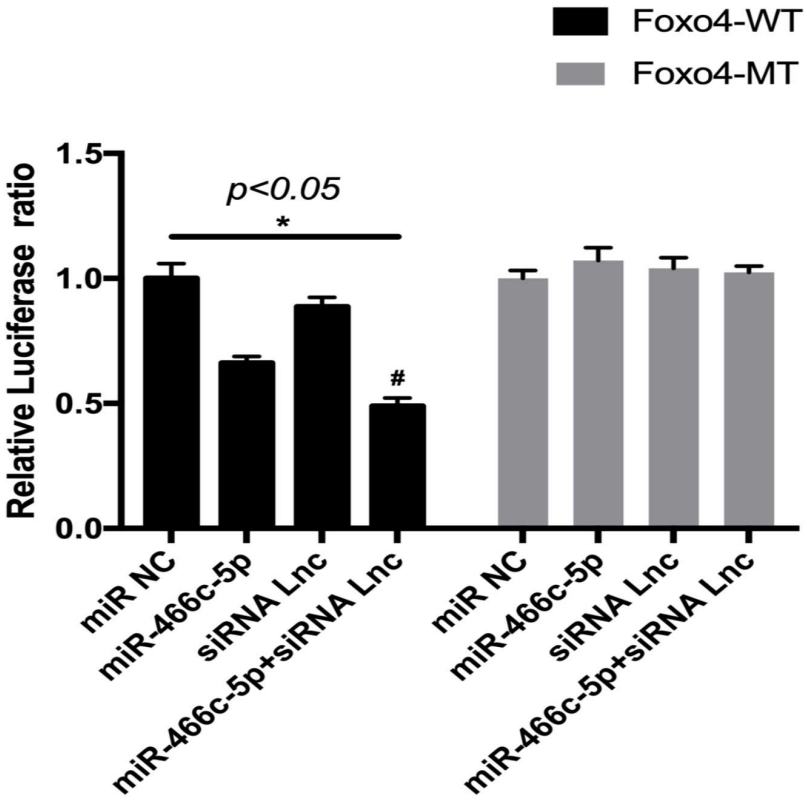


siRNA Experiments



siRNA Lnc： SULT1C2A and Foxo4↓,
miR-466c-5p ↑



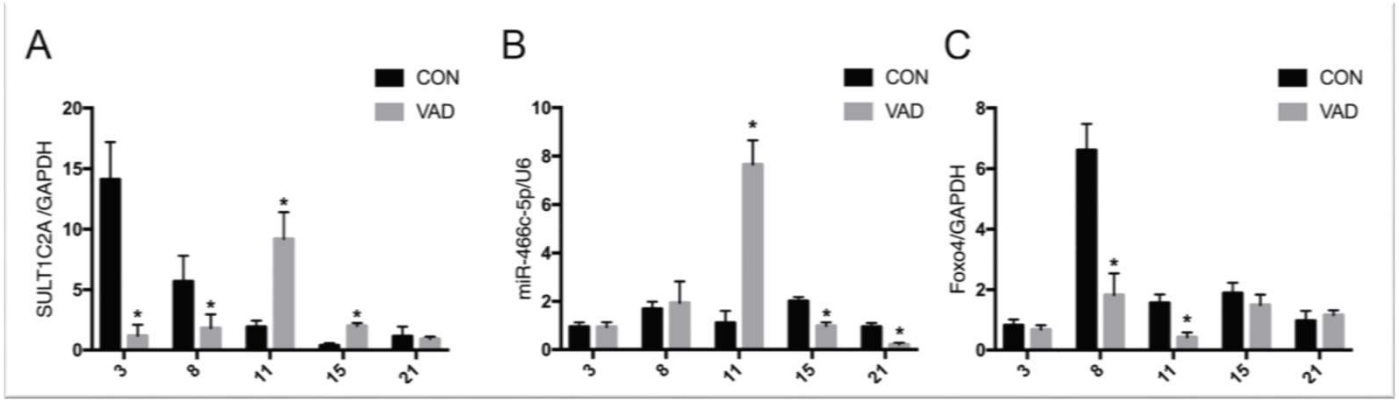


miR-466c mimics 、 siRNA-lncRNA

SULT1C2A ： Foxo4-WT ↓

SULT1C2A-miR-466c-*Foxo4* axis

qRT-PCR



Foxo4

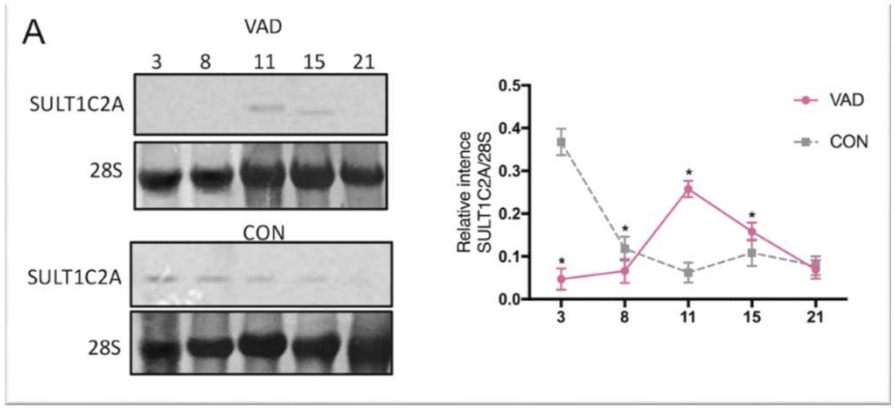
GD 11.5
EMAGE:1315

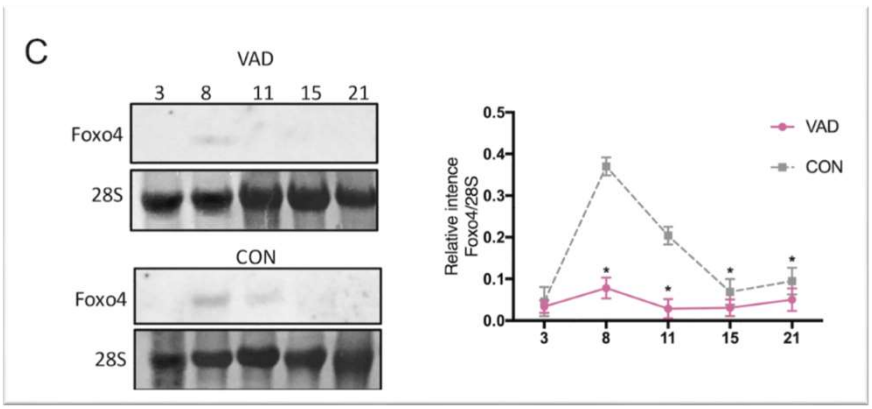
GD 15.5
EMAGE:13835

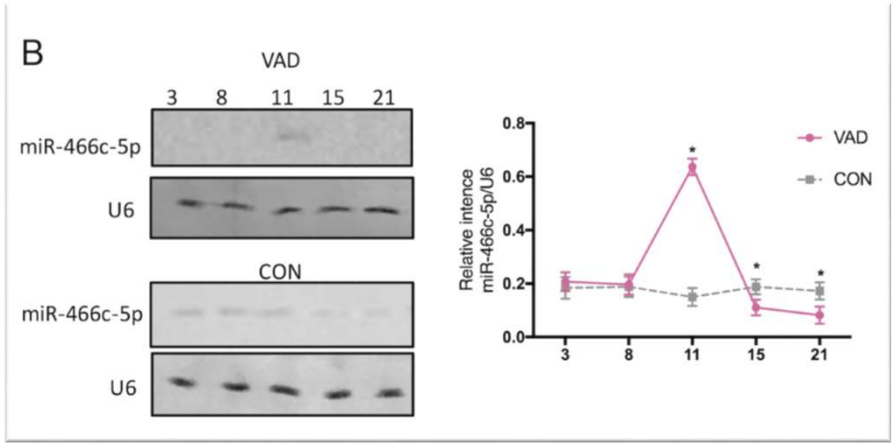
- **SULT1C2A : GD 3 ， 8↓，GD 11 ， 15↑;**
- **miR-466c-5p : GD 11↑，GD 15 ， 21↓;**
- **Foxo4 : GD 8 ， 11↓;**

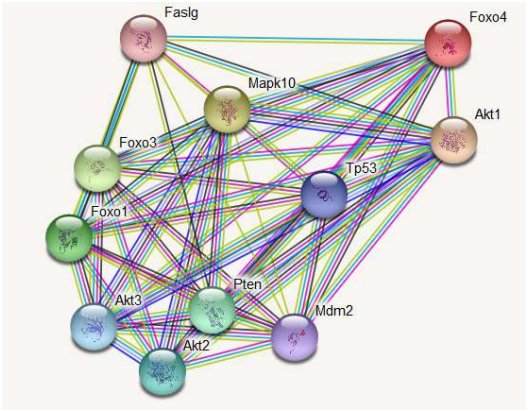
SULT1C2A-miR-466c-*Foxo4* axis

Northern Blot



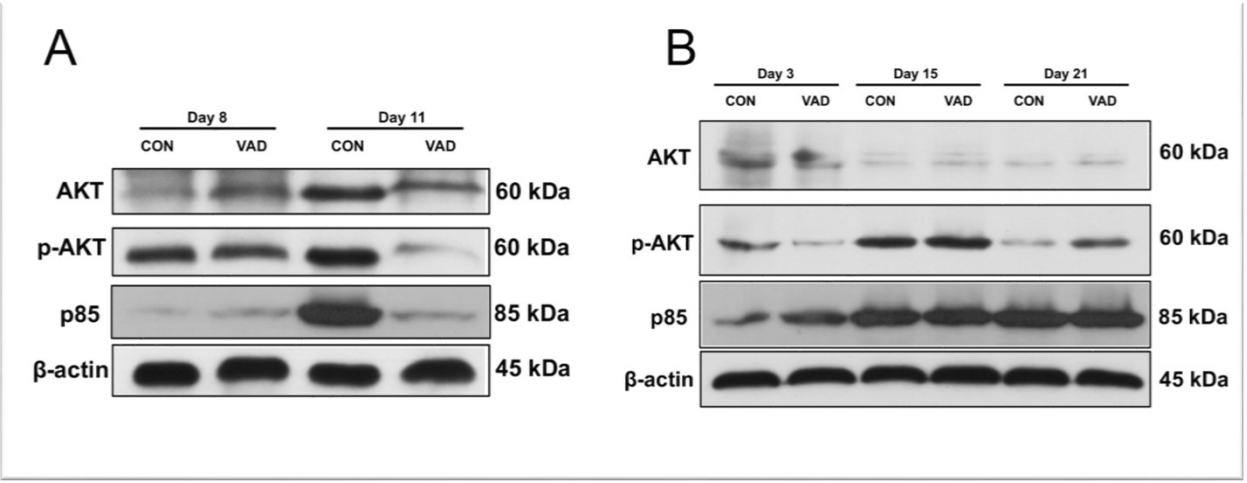


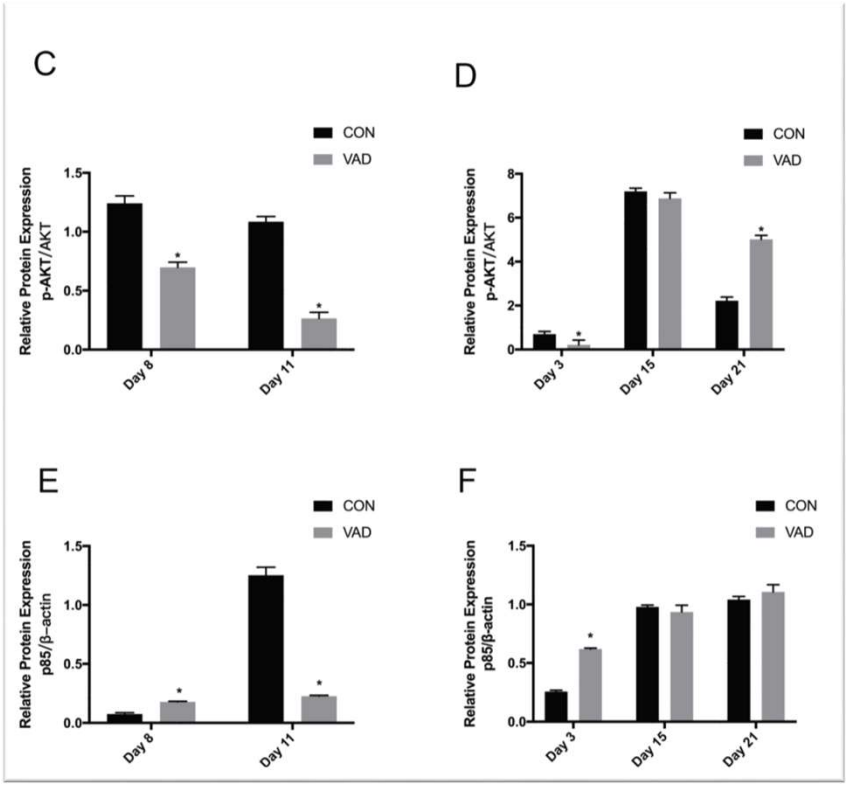




PI3K-AKT-Foxo4 pathway

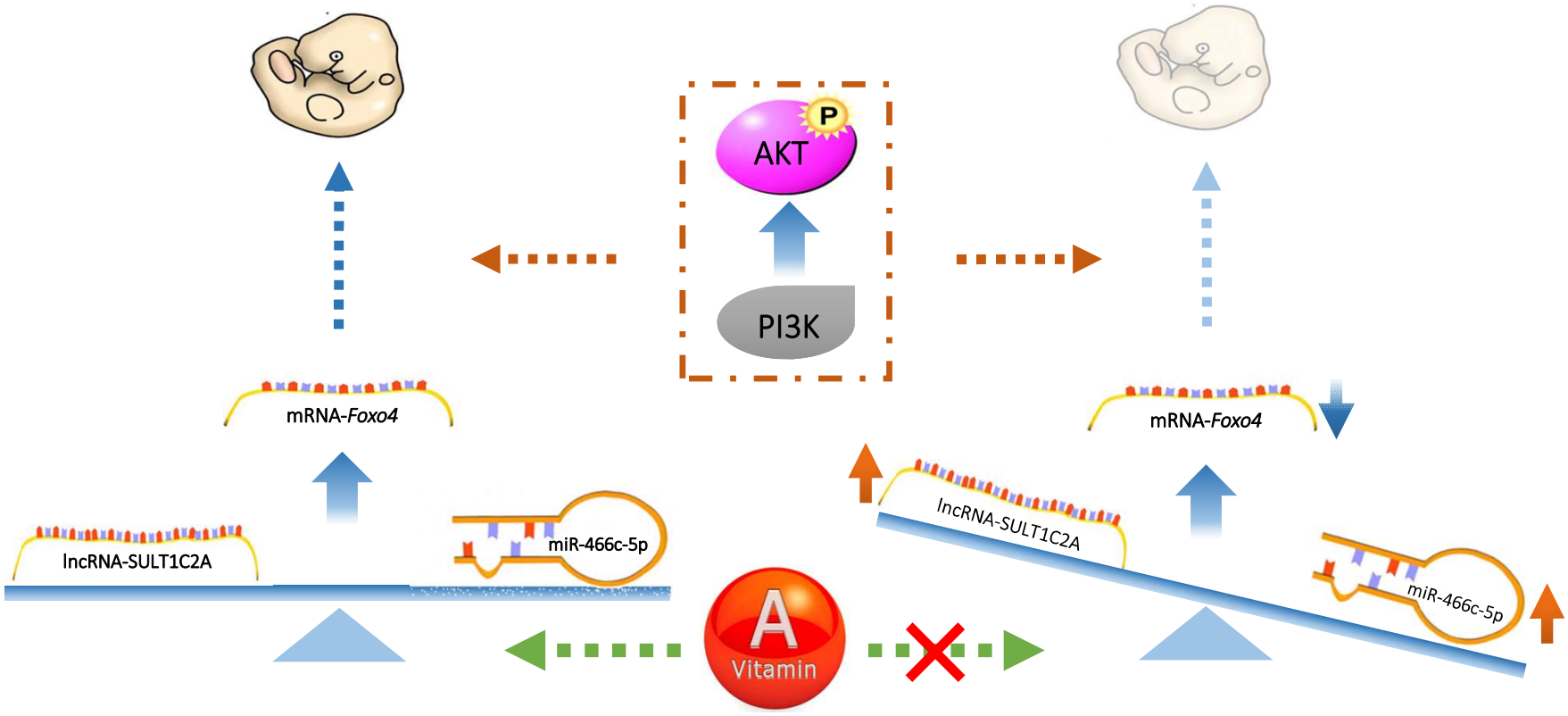
Western Blot





- p-AKT : GD 3 ， 8， 11 ↓， GD 21 ↑;
- p85 : GD 11 ↓ ， GD 3 ， 8 ↑ ;

Schematic Diagram of SULT1C2A-rno-miR-466c-5p-Foxo4 axis



GD 9 → GD 11

Conclusions

- VAD initially reduces the expression of *Foxo4* and subsequently up-regulates miR-466c-5p；
- To maintain the relative balance of embryonic development, lncRNA SULT1C2A is up-regulated on GD 11 and functions as a sponge to antagonize miRNAs；
- The dynamic changes in the expression of the SULT1C2A-rno-miR-466c-5p-*Foxo4* axis may be initiated on GD 9 and were most obvious on GD 11 during the early-mid stage of somitogenesis；
- Dynamic profile of AKT phosphorylation, an important signaling pathway for somitogenesis, was found in the VAD group, and this novel finding warrants further research；

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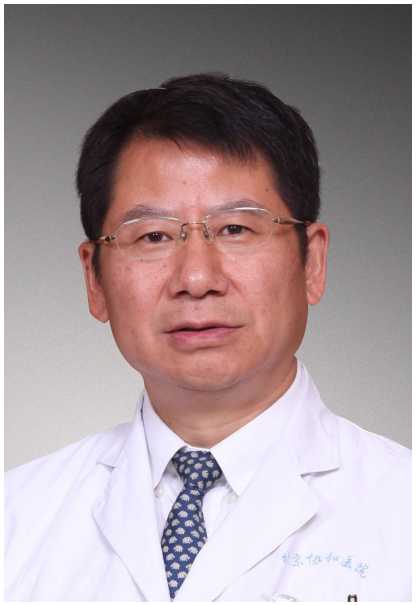

Thanks