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MiR-34c-5p Inhibit Uremia Vascular Calcification through Down-Regulating Fut8

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Objectives : To construct a more bionic vascular calcification model of uremia. The changes of Fut8 in the vascular calcification model were determined. To explore the mechanism of miR-34c-5p on vascular calcification.
Methods : Cells co-cultrue, Bioinformatics, Western and Lectin blotting, Quantitative real-time PCR analysis, Immunocytochemical staining, Luciferase reporter assay, Lectin pull-down assay, Alizarin red staining, Measurement of Fut8 activity, Calcium content assays.
Results : In endothelial/smooth muscle cell co-culture systems, overexpressed miR-34c-5p promoted uremia serum-induced calcification in HASMCs(Fig1). Fut8 was increased in calcification model(Fig1). miR-34c-5p targeted Fut8 and negatively regulate the calcification(Fig1). In our previous study, inhibition of Fut8 mitigated vascular calcification. Overexpression miR-34c-5p affects expression of Fut8 and suppresses uremic serum-induced HASMCs calcification. Oe-Fut8 effectively reverses the expression of miR-34-5p in vitro. (Fig2).
Conclusions : In uremic vascular calcification disease, miR-34c-5p can down-regulate the degree of calcification by inhibiting Fut8.

终图 1_1.jpg


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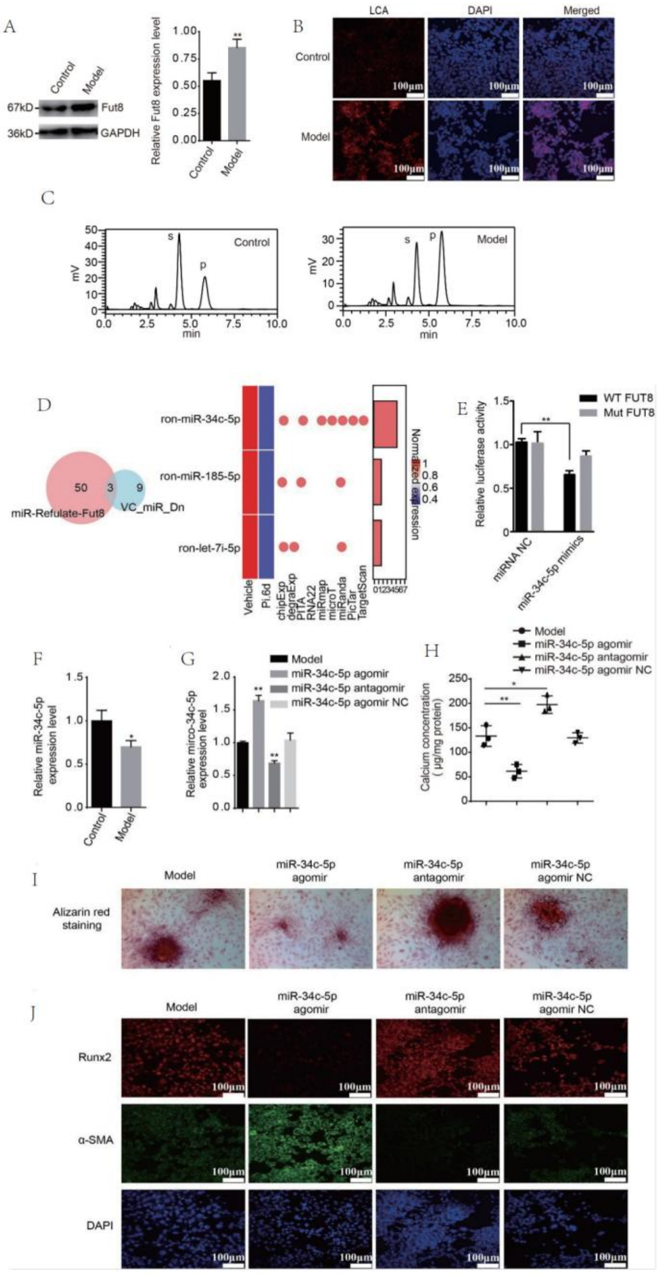
A
Control
Model
67kD
Fut8
36kD
GAPDH
Relative Fut8 expression level
B
LCA
DAPI
Merged
C
Control
Model
mV
min
D
miR-Refulate-Fut8
VC_miR_Dn
ron-miR-34c-5p
ron-miR-185-5p
ron-let-7i-5p
Vehicle
Pi_6d
chipExp
degraExp
PITA
RNA22
miRmap
microT
miRanda
PicTar
TargetScan
Normalized expression
E
WT FUT8
Mut FUT8
Relative luciferase activity
miRNA NC
miR-34c-5p mimics
F
Relative miR-34c-5p expression level
G
Model
miR-34c-5p agomir
miR-34c-5p antagomir
miR-34c-5p agomir NC
Relative mirco-34c-5p expression level
H
Calcium concentration (μg/mg protein)
I
Alizarin red staining
J
Runx2
α-SMA
DAPI

终图 1_1.jpg

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