Co RNA w - RNA- t a a o o TOR o a -

Ma Aa ,[1,2] A a G o ,[1,2] M a B a a,[3] Pa a B o c ,[1,2] A a Ba t o,[1,2] T o Ca o ,[1,2] Ma a K c,[4] Da F. Ka a,[3] Go a a D a a a ,[1,2] Na Ma t ,[1,2,8] o R ,[5] Jo a ,[6] K Ka ,[3] Ta C a a,[5,7] a G [1,2]

[1]M R . C (MRC) L I . M S . (LMS), L W12 0NN, U K ; [2]I . C S . (ICS), F . M , I C L , L W12 0NN, U K ; [3]MRC C . R M , U . E , E EH16 4UU, U K ; [4]W . G B . , L-1526 L , L ; [5]E . P , T B , I , C CB22 3AT, U K ; [6]R . I . M P . (IMP), V B (VBC), V 1030, A . ; [7]MRC H G . U , U . E , E EH4 2XU, U K

E o o t a c t o a c o OCT4, SO 2, KLF4, a M C (OSKM) o a o a t c t o - k o t t (PSC). R o a a ow a c t o , t a t a c a t o . O c a c a . To t o a o o a - , w o a o -w RNA a a o a OSKM. I t , w t o a o OSKM- a - a t o c as *1A*. o a o a a oa c t a t a - RNA . c (cRNA) w t t RNA c t o a t a c a o a c o o t t a a t. O a a a a o o a a , w c c a a t a o a c (TOR) o a . O o a , TOR t o t t c o o c c - t a (CDK) t o (CDKI), c 16[INK4a], 21[CIP1], a 15[INK4] , t OSKM- a . O t o t a , t o o TOR t t a -a o a t c t o o t (SASP), w c t a o o a . T o t a a c o o t t t o a t o o c a TOR a o o a - . O a , o t t a a t a o o k c o a c w t RNA t o a c a t t o o o t o wa o t o o o t o .

[*K* : SASP; . ; PSC ; . - RNA- ; . RNA .]

S . .

R F 20, 2017; . . , O 18, 2017.

D , (OCT4, SOX2, KLF4, MYC, - . S . OSKM) . , , ES , . T . (G 1962; W . 1997) . (ES) (T . 2001). I , . Y . (T . Y . 2006) . -

(Q . 2014; S . 2014; Y . 2014; C . 2015). A (K . 2010). T (, TERT 53) (Q . 2014), PSC (K L 2009; B G 2010). K 53, 16^{INK4}, 21^{CIP1} OSKM, (B . 2009; H . 2009; K . 2009; L . 2009; M . 2009; U . 2009). H S . A , (SASP) (K . 2010; S . 2014). S SASP (C . 2010). I (M S 2014), (B . 2016), (K 2008) (C S 2010). S (M . 2016). T IL-6 SASP, IL-6 MYC PIM1 (B . 2013). G . G (J . 2000; R 2005; A . 2008; T . 2016; W . 2016) (Q . 2014; S . 2014; Y . 2014).

I , RNA *a OSKM a A* , , T , *RN35-226 d 16* RNA *a a 352218 -236 -2283 -33 RN TJT -36 -8 -383* (RNA-) RNA . U , (TOR) *-3528 35 3512a a a 53,352* . M RNA-

S a

R

E a OSKM

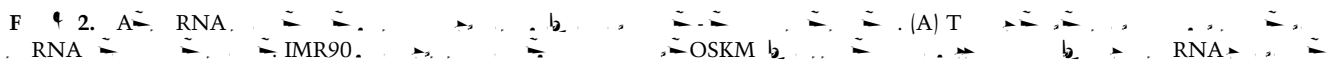
H (OSKM) IMR90 (F . 1A,B) (B . 2009). S RAS^{G12V}, OSKM (CDK) (CDKI) 15^{INK4}, 16^{INK4}, 21^{CIP1}, (F . 1C).

T OSKM- . RNA (RNA-). G (GSEA) SASP OSKM (F . 1D). O OSKM- RAS- . F , TGF-β- (S F . S1A). I . RAS OSKM , F , RAS OSKM (S F . S1B), RAS (F . 1E). O , RAS OSKM (S = 0.33) (F . 1F). A (F . 1G; S F . S1C), (GO) ( ) (F . 1H; S F . S1D). B , OSKM RAS . F , GO OSKM RAS (F . 1I; S F . S1E). O , OSKM .

a 3 35223 -118a -2 a 12 12 a

F ¶ 1. Ħ . OSKM IMR90 . (*A*) S - IMR90. ( ), OSKM RAS. A 12 ( ), β- (SA-β-G) ( ), B U 18- ( ). B , 100 M. (*B*) Q . B U SA-β-G . (***) $P < 0.001$. (*C*) Q RT–PCR (RT–PCR) RNA . *CDKN2B* (15^{INK4}), *CDKN2A* (16^{INK4A}), *CDKN1A* (21^{CIP1}) . (*) $P < 0.05$; (***) $P < 0.001$. (*D*) G (GSEA) SASP OSKM IMR90 . (NES) N . (*E*) H . (C . 2004). IMR90 , RAS, OSKM. L 2 ( ) *Z*- , RAS OSKM . B . (*F*) S 2 (FC) RAS OSKM . G ([FDR] < 0.05; 2 < –1 2 > 1) . (*G*) V - RAS OSKM . D 2 < –1 FDR < 0.05. (*H,I*) G (GO) OSKM- RAS- (*H*) OSKM- (*I*). F , ( ) 2 < –1 ($P < 0.05$) . N , M (://). GO . S - . O ($P < 0.05$) .

S F S2A. A RNA (RNA ; 3153 RNA) (F . 2A). S RNA OSKM- (37 0) (F . 2B,C). A RNA RNA (*CDKN1A*, *MTOR*, *MYOT*, *UBE2E1*) OSKM- (F . 2D; S F . S2D).



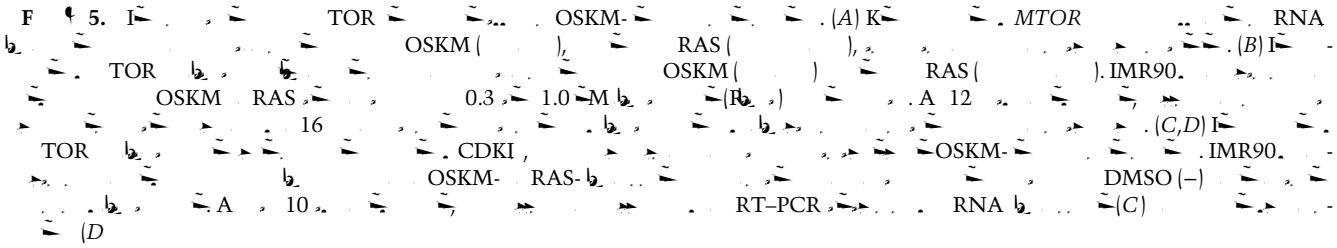
T , IMR90 OSKM RNA . W RNA (S F . S3A–C). *MYOT* , RNA ( ). T RNA *CDKN1A*, *MYOT*, *MTOR*, *UBE2E1* OSKM- (F . 2E), B U (F . 2F; S F . S3D), β- (SA-β-G IMR90 OSKM (F . 2G,H; S F . S3E). S 21^{CIP1} (B . 2009),

RNA- a a a a a a a a RNA

A . RNA- RNA- (T R 2017). W . RNA- RNA , .

T . RNA (F . 3A). T . RNA R-E, R-30- (F . 2013), . RNA II (P II)- (D . 2005). W . RNA 3′ (LTR) . , -A. (ATTAAA) 3′ . R-E, . T . RNA , RNA R-E (S F . S4A). T . RNA (S F . S4B) . RNA (F . 3B). M . RNA (116 . 300) (F . 3C), . RNA (310 . 359 . RNA) (F . 3D).

N , . RNA- . IMR90 OSKM . RNA (F . 3E). C . RNA (RNA *MTOR* *CDKN1A* *MYOT* *UBE2E1*,). T . RNA- . RNA (F . 3E), . *TP53*, *CDKN1A*, *MTOR*, *UBE2E1* RNA (F . 3F). M , *CDKN1A*, 53 , OSKM– 53 . W *MYOT* . F , RNA- . (F . 3G; S F . S4C). T . RNA . W (. 06 19.6720T 19.7353.6( )



F 5. I TOR OSKM- . (*A*) K . *MTOR* . RNA OSKM ( ), RAS ( ), . (*B*) I - . TOR OSKM ( ) RAS ( ). IMR90. OSKM RAS 0.3 1.0 M (R) . A 12 , 16 . . (*C*,*D*) I . TOR . CDKI , OSKM- . IMR90. - OSKM- RAS- DMSO (–) . A 10 , . RT–PCR . RNA (*C*) - (*D*

- . C . RNA- , *CDKN2B* OSKM, - OSKM- RAS- (S F . S6A). B . RNA- TOR (F . 5C). A - , , . *CDKN2A*, *CDKN2B*, *CDKN2A* *CDKN1A*, *CDKN1A* TOR . RAS OSKM- (RAS-) . - CDKI . A-

CDKI OSKM RAS -
( )
OSKM-
(F . 5C). A 16^{INK4a} W
16^{INK4a}
OSKM-
RAS- (F . 5D; S F .
S6B).
W
CDKN1A, CDKN2A, CDKN2B OSKM-
. D *CDKN2B*
(F . 4E, 5C), *CDKN2B*
. RNA
OSKM (F . 5E; S F . S6C;
). O , *CDKN1A*

F 6. D .. . TOR PSC . (*A*) R . C, 9- TNG MKOS MEF 1 *Ba* MKOS RNA RNA . N N -GFP⁺ 14. S F S7, B C, . E - SD . (*) $P < 0.05$; (**) $P < 0.01$; (***) $P < 0.001$; ( ) . (*B*) D - - . R (0.3 1.0 M) 3, 6, 14 MKOS R PSC . (AP) 14 . D . E SD (0–3 0–6). D 0–14. (*) $P < 0.05$; (**) $P < 0.01$; (***) $P < 0.001$. (*C*) D - . R (0.3 1.0 M) 3 MKOS - R PSC 14 . (*D*) R - C, 9- N - GFP MEF *Ba* MKOS- - O U6- RNA . 500 M TGF-β RI (A 5 ; T , A83-01) 5 M (R). N N -GFP⁺ 14. E SD . (*) $P < 0.05$; (**) $P < 0.01$; ( ) . (G) S N⁺ ; ( ). . (*E*) R - MKOS MEF (CM). MEF , RAS, RAS RNA *M* . CM 3 4. MEF . A (AP⁺) CM . RAS/ . E SD . (**) $P < 0.01$; (***) $P < 0.001$. R . (*F*) S . MTOR . PSC . (G .) ; ( ) .

(S F S7F). O , TOR . F . 6F).

D

A M (Q . 2014; S . 2014; Y . 2014). S . (B . 2009). D , - , - .

53, 16^{INK4}, , 21^{CIP1} (K L 2009; B G 2010). T , RNA OSKM- . O OSKM- . W . RNA . : *CDKN1A*, *MTOR*, *MYOT*, *UBE2E1*. W *CDKN1A* (21^{CIP1}) *MTOR* , *MYOT* *UBE2E1* - . *MYOT* Z- . (O . 2005). D . RNA *MYOT* . RNA , . *MYOT* IMR90 . UBE2E1

E2 (N . 1996).
P - -
(D -S
. 2013). W UBE2E . G 21CIP1

P a

F RE- RNA , 97- - RNA PCR- . RE-X RE-E O - RLL (F 2013). RNA T S1.

RNA a a

D RNA M .

L a a a RNA

G DNA 10^6 G P (Q) . S (3 [ ]. GIPZ 4 RLL), PCR 2 µ DNA . I PCR - QIA (Q), Q 2.0 F - (40). P , - S PCR B A DNA (A T -) . O. , 12 M I G A II H S 2500 - (50-),

S a a a a RNA

FASTA CASAVA 1.8. T RNA B (0.12.8). C F ! *P-* R. T F ! *P-* , (F. 1925).

I a - a a

C 4%. 30 , PBS, 0.2% T X-100. 5 .

A . , JC, O'L ~A, B, ~ A, G , MV, A A, R,
S, F , M, D, C . , M, B ~C, P N, . 2008.
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. ~ . *C* **133:** 1006–1018.
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. . , .. ~ . ~ ~ ~ ~ . *C*
167: 1867–1882 1821.
B, DJ, C . BG, D M, W . ME, S ~CJ, Z ~ J,

P S, F OR, B AK, W G, S S, S R. 2014. F RNA- S 2. *Na P* **9:** 171–181.

Coupling shRNA screens with single-cell RNA-seq identifies a dual role for mTOR in reprogramming-induced senescence

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