

Fig. S1 Dose-response gradient assay of luciferase report vector in CNE cells.

To find the optimal concentration for transfection of pRL-TK in CNE cells, dose-response gradient assay was performed. pRL-TK was transfected to CNE cells with concentration from 20 ng/mL to 800 ng/mL. Then, the luminescence was detected. 10 ng pGL3-Basic Vector was co-transfected in each assay to normalize the *Renilla* luciferase activity.



Fig. S2 Differentiation assay and dose-response gradient assay in mESC.

mESCs grown under feeder free condition (undifferentiation, LIF+) were induced with RA (differentiated, LIF-, 72h), and the degree of differentiation was assessed by examining the expression level of Oct4 and Sox2 with real time PCR (A and B). Dose-response gradient assay of pRL-TK vector was performed in mESCs. pRL-TK was transfected to undifferentiated mESCs with dose from 20 ng/mL to 800 ng/mL. 10 ng pGL3-Basic Vector was co-transfected in each assay to normalize the *Renilla* luciferase activity (C).





Real time PCR (A) was performed to verify our microarray result (B). The two data have a strong correlative relationship (Spearman r= 0.79, P=4.58E-11, n=46). The gene with microarray fluorescence intensity above 1000 is considered as "expression". CCND1 in normoxia CNE has the smallest fluorescence intensity above 1000, so CCND1 relative expression in normoxia CNE was set as 1. The relative expression was normalized by B2M.



Fig. S4 Luciferase expression of 3'UTR constructs.

CNE cells were transfected with luciferase report vectors bearing different 3'UTRs with concentration from 40 to 480 ng/mL and the cells were collected for RT-PCR to detect the expression of luciferase. The 3'UTR luciferase report vectors include ACTB, ARPC3, ATP5B, ATP6AP1, CCND1, CCND3, LDHA, PGK1, POU5F1 and VEGFA. The relative expression was normalized by B2M (A-I). Along with increased concentration of these constructs, the expression of luciferase - 3'UTR transcripts also increased.

Fig. S5 The expression of endogenous GAPDH and the luciferase transcript.

CNE cells were transfected with pRL-TK report vector bearing GAPDH 3'UTR with no predicted MREs. The cells were collected and the expression of endogenous GAPDH mRNA (A) and luciferase mRNA (B) were determined by RT-PCR. The genes' expression was normalized by B2M and presented as the relative expression. Along with increased concentration of the construct, the expression of luciferase transcript increased but the expression of endogenous GAPDH mRNA did not have significant change.