Supplemental Information

IncRNA *GCAT1* is involved in premature ovarian insufficiency by regulating p27 translation in GCs via competitive binding to PTBP1

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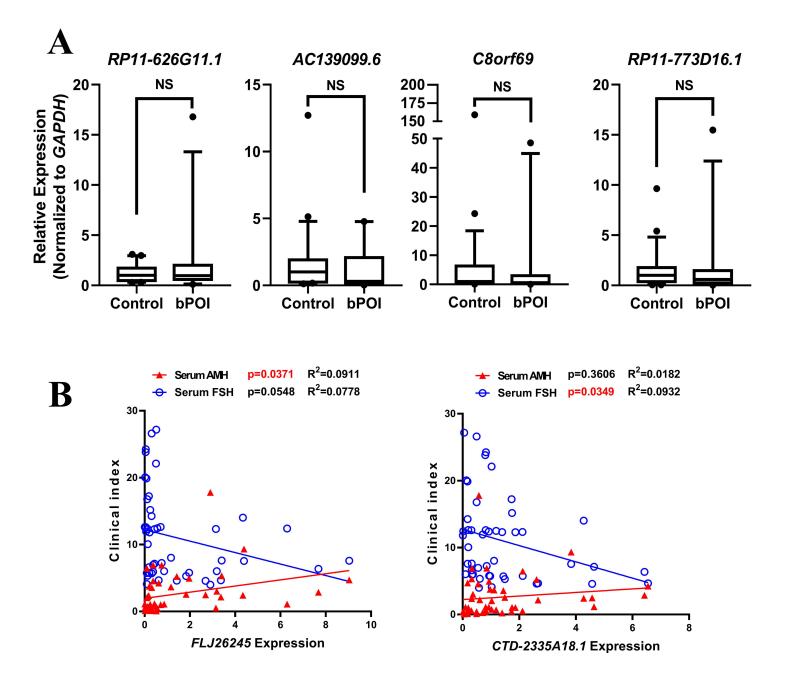


Figure S1. (A) The expression level of differentially expressed lncRNAs was validated by qRT-PCR in GCs from an independent cohort of patients with bPOI (n = 24) and controls (n = 24). Ct values were normalized to GAPDH. Data are presented as the median \pm interquartile range. NS = No significance. Two-tailed Mann–Whitney U-test. **(B)** The correlation between the expression levels of lncRNAs in GCs and the serum concentration of AMH and FSH was analyzed by Pearson Correlation Analysis.



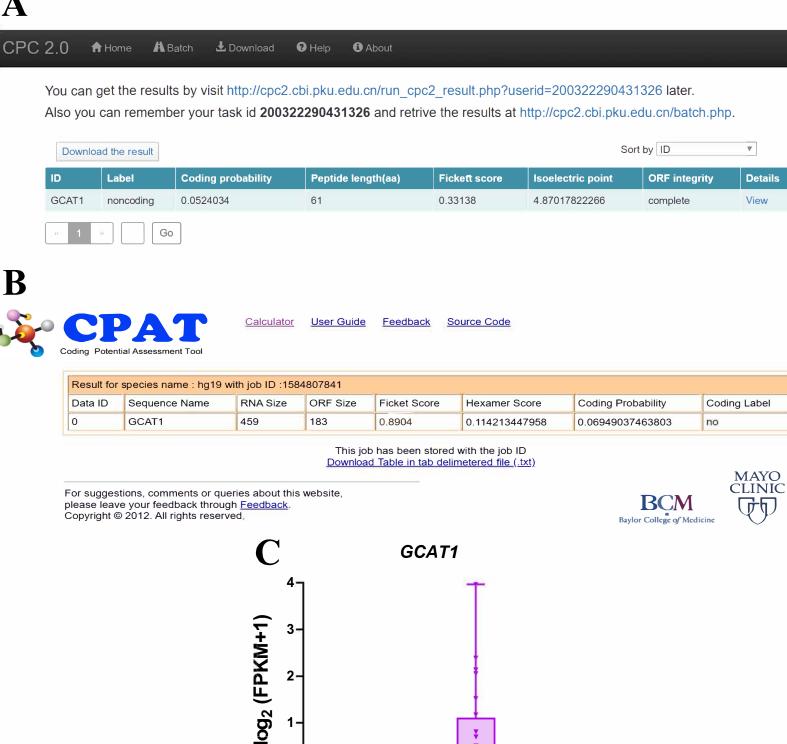
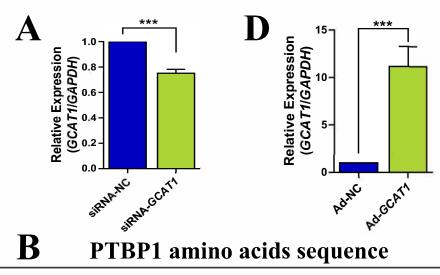


Figure S2. (A/B) Coding probability of *GCAT1* was assessed by Coding Potential Calculator (CPC2)**(A)** and Coding-Potential Assessment Tool (CPAT)**(B). (C)** The expression of *GCAT1* in human granulosa cells during folliculogenesis.



MDGIVPDIAVGTKRGSDELFSTCVTNGPFIMSSNSASAANGNDSKKFKGDSRSAGVPSRVIHIRKLPIDVTE GEVISLGLPFGKVTNLLMLKGKNQAFIEMNTEEAANTMVNYYTSV1TPVLRGQPIYIQFSNHKELKTDSSPNQARAQAALQAVNSVQSGNLALAASAAAVDAGMAMAGQSPVLRIIVENLFYPVTLDVLHQIFSKFGTVLKIITFTKNNQFQALLQYADPVSAQHAKLSLDGQNIYNACCTLRIDFSKLTSLNVKYNNDKSRDYTRPDLPSGDSQPSLDQTMAAAFGAPGIISASPYAGAGFPPTFAIPQAAGLSVPNVHGALAPLAIPSAAAAAAAAGRIAIPGLAGAGNSVLLVSNLNPERVTPQSLFILFGVYGDVQRVKILFNKKENALVQMADGNQAQLAMSHLNGHKLHGKPIRITLSKHQNVQLPREGQEDQGLTKDYGNSPLHRFKKPGSKNFQNIFPPSATLHLSNIPPSVSEEDLKVLFSSNGGVVKGFKFFQKDRKMALIQMGSVEEAVQALIDLHNHDLGENHHLRVSFSKSTI

C hnRNPK amino acids sequence

METEQPEETFPNTETNGEFGKRPAEDMEEEQAFKRSRNTDEMVELRILLQSKNAGAVIGKGGKNIKALRT

DYNASVSVPDSSGPERILSISADIETIGEILKKIIPTLEEGLQLPSPTATSQLPLESDAVECLNYQHYKGSDFD

CELRLLIHQSLAGGIIGVKGAKIKELRENTQTTIKLFQECCPHSTDRVVLIGGKPDRVVECIKIILDLISESPIK

GRAQPYDPNFYDETYDYGGFTMMFDDRRGRPVGFPMRGRGGFDRMPPGRGGRPMPPSRRDYDDMSPR

RGPPPPPPGRGGRGGSRARNLPLPPPPPPRGGDLMAYDRRGRPGDRYDGMVGFSADETWDSAIDTWSPSE

WQMAYEPQGGSGYDYSYAGGRGSYGDLGGPIITTQVTIPKDLAGSIIGKGGQRIKQIRHESGASIKIDEPLE

GSEDRIITITGTQDQIQNAQYLLQNSVKQYADVEGF

Figure S3. (A) The efficiency of GCATI knockdown via siRNA in KGN cells was detected by qRT-PCR. Values of qRT-PCR were obtained from triplicates and expressed as the mean \pm SD (n = 3). Two-tailed Student's t-test. ***P < 0.001. **(B)** The amino acid sequence of PTBP1 protein. Five unique peptides identified by MS from GCATI-binding protein were highlighted in red font and the RNA-binding domains were highlighted by gray background. **(C)** The amino acid sequence of hnRNPK protein. Seven unique peptides identified by MS from GCATI-binding protein were highlighted in in red font and the RNA-binding domains were highlighted by gray background. **(D)** The efficiency of GCATI over-expression via adenovirus in KGN cells was detected by qRT-PCR. Values of qRT-PCR were obtained from triplicates and expressed as the mean \pm SD (n = 3). Two-tailed Student's t-test. ***P < 0.001.

Table S1 List of primers used in this study

Gene (homo sapiens)	Forward (5'-3')	Reverse (5'-3')	
GCAT1	AGGTCTCCTGCCTCCAA	TCCTCTTCCTCCACCTCTGC	
GAPDH	GGGAAACTGTGGCGTGAT	GAGTGGGTGTCGCTGTTGA	
LMNB1	GAAAAAGACAACTCTCGTCGCA	GTAAGCACTGATTTCCATGTCCA	
CDKN1B	GTCAAACGTGCGAGTGTCTA	CATGTCTCTGCAGTGCTTCT	
PTBP1	ATTGTCCCAGATATAGCCGTTG	GCTGTCATTTCCGTTTGCTG	

Table S2 List of siRNA used in this study

siRNAs	Sense(5'-3')
siRNA- <i>GCAT1</i>	GAUGGCAGAGCAGAUGCAATT
siRNA-CDKN1B	GAGCAATGCGCAGGAATAAGG
siRNA-PTBP1	CCAGCCCATCTACATCCAGTT

Table S3 List of antibodies used in this study

Antibody	Supplier	Catalog#	Application
Anti-PTBP1	Abcam	ab133734	WB
Anti-PTBP1	Cell Signaling Technology	57246S	RIP
Anti-hnRNPK	Abcam	ab39975	RIP
Anti-p27 Kip1	Cell Signaling Technology	3686S	WB
Tubulin	Proteintech	66031-1-lg	WB
Actin	Proteintech	66009-1-lg	WB