

Supplementary Material

1 Supplementary Figures and Tables

Supplementary Table 1. Sequenced Primer product for the bovine TDG gene

Gene	Primer	Sequenced Product	Percent Identity
TDG	F: GAACGCGGGCAGCTATTCTC R: GTCTCTCGTGTGGGTTCTG	NNNNNNNNNNNNNNNNNNNNNTTCCNTTCCNA CAAATGATGACTGAAGCTCCTGACATGGCA GTTATAAACGCACAAGAAATGCCAGCCGAAGTTC CTGCCCCAGCACCTGCTCAGGAACCCAC ACGAGAGACA	98%

Bos taurus thymine DNA glycosylase (TDG), mRNA

Sequence ID: [NM_001083696.2](#) Length: 3198 Number of Matches: 1

Range 1: 195 to 307 [GenBank](#) [Graphics](#)

[▼ Next Match](#) [▲ Previous Match](#)

Score	Expect	Identities	Gaps	Strand
202 bits(109)	4e-50	111/113(98%)	0/113(0%)	Plus/Plus
Query 1	TTTCCNTTCCNACAAATGATGACTGAAGCTCCTGACATGGCAGTTATAAACGCACAAGAA	60		
Sbjct 195	TTTCCATTCCAACAAATGATGACTGAAGCTCCTGACATGGCAGTTATAAACGCACAAGAA	254		
Query 61	ATGCCAGCCGAAGTTCCTGCCCCAGCACCTGCTCAGGAACCCACACGAGAGAC	113		
Sbjct 255	ATGCCAGCCGAAGTTCCTGCCCCAGCACCTGCTCAGGAACCCACACGAGAGAC	307		

Bos taurus thymine-DNA glycosylase, mRNA (cDNA clone MGC:140185 IMAGE:8168686), complete cds

Sequence ID: [BC134451.1](#) Length: 3199 Number of Matches: 1

Range 1: 196 to 308 [GenBank](#) [Graphics](#)

[▼ Next Match](#) [▲ Previous Match](#)

Score	Expect	Identities	Gaps	Strand
202 bits(109)	4e-50	111/113(98%)	0/113(0%)	Plus/Plus
Query 1	TTTCCNTTCCNACAAATGATGACTGAAGCTCCTGACATGGCAGTTATAAACGCACAAGAA	60		
Sbjct 196	TTTCCATTCCAACAAATGATGACTGAAGCTCCTGACATGGCAGTTATAAACGCACAAGAA	255		
Query 61	ATGCCAGCCGAAGTTCCTGCCCCAGCACCTGCTCAGGAACCCACACGAGAGAC	113		
Sbjct 256	ATGCCAGCCGAAGTTCCTGCCCCAGCACCTGCTCAGGAACCCACACGAGAGAC	308		

PREDICTED: Bos taurus thymine DNA glycosylase (TDG), transcript variant X1, mRNA

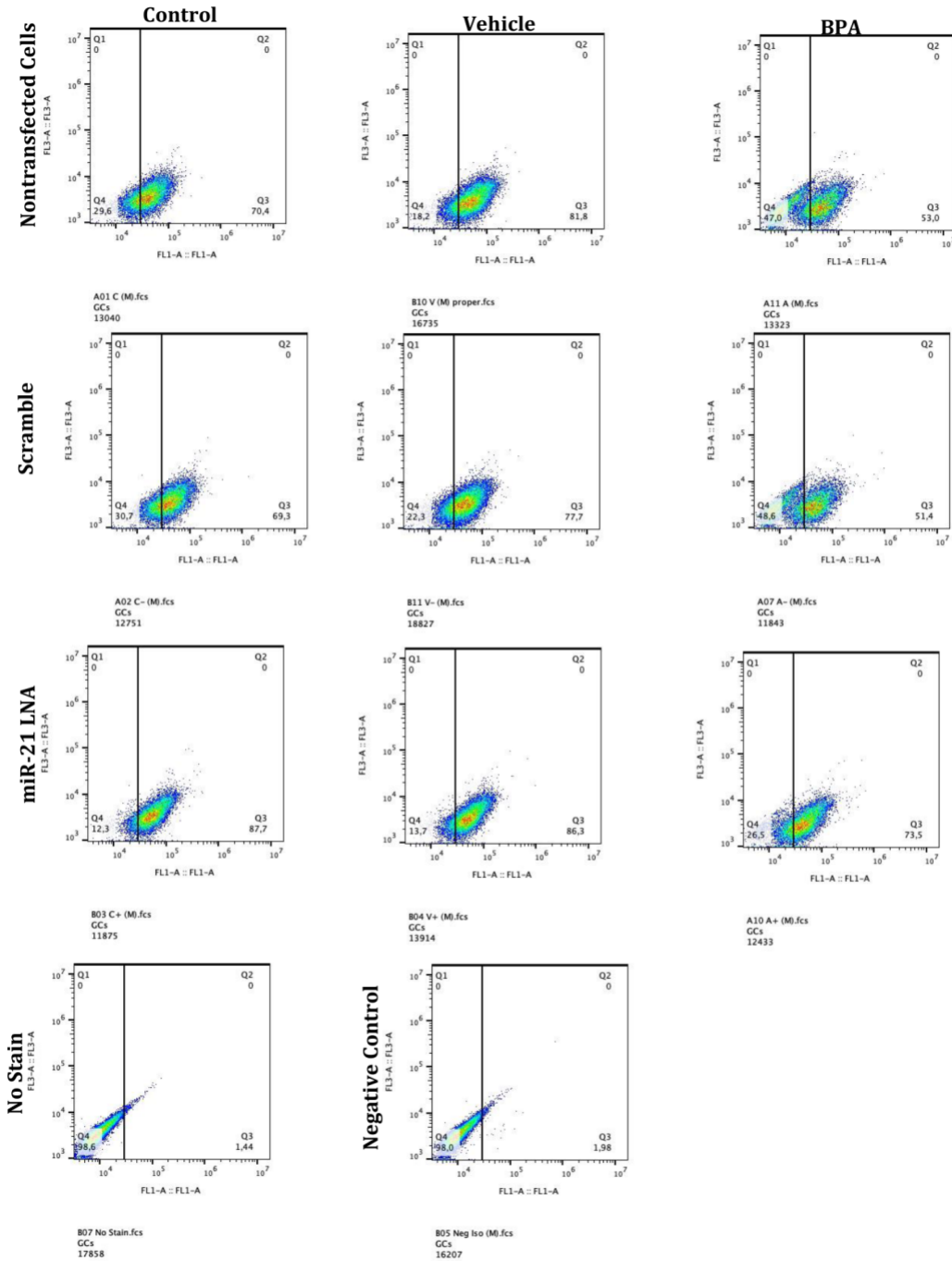
Sequence ID: [XM_024991735.1](#) Length: 3354 Number of Matches: 1

Range 1: 363 to 475 [GenBank](#) [Graphics](#)

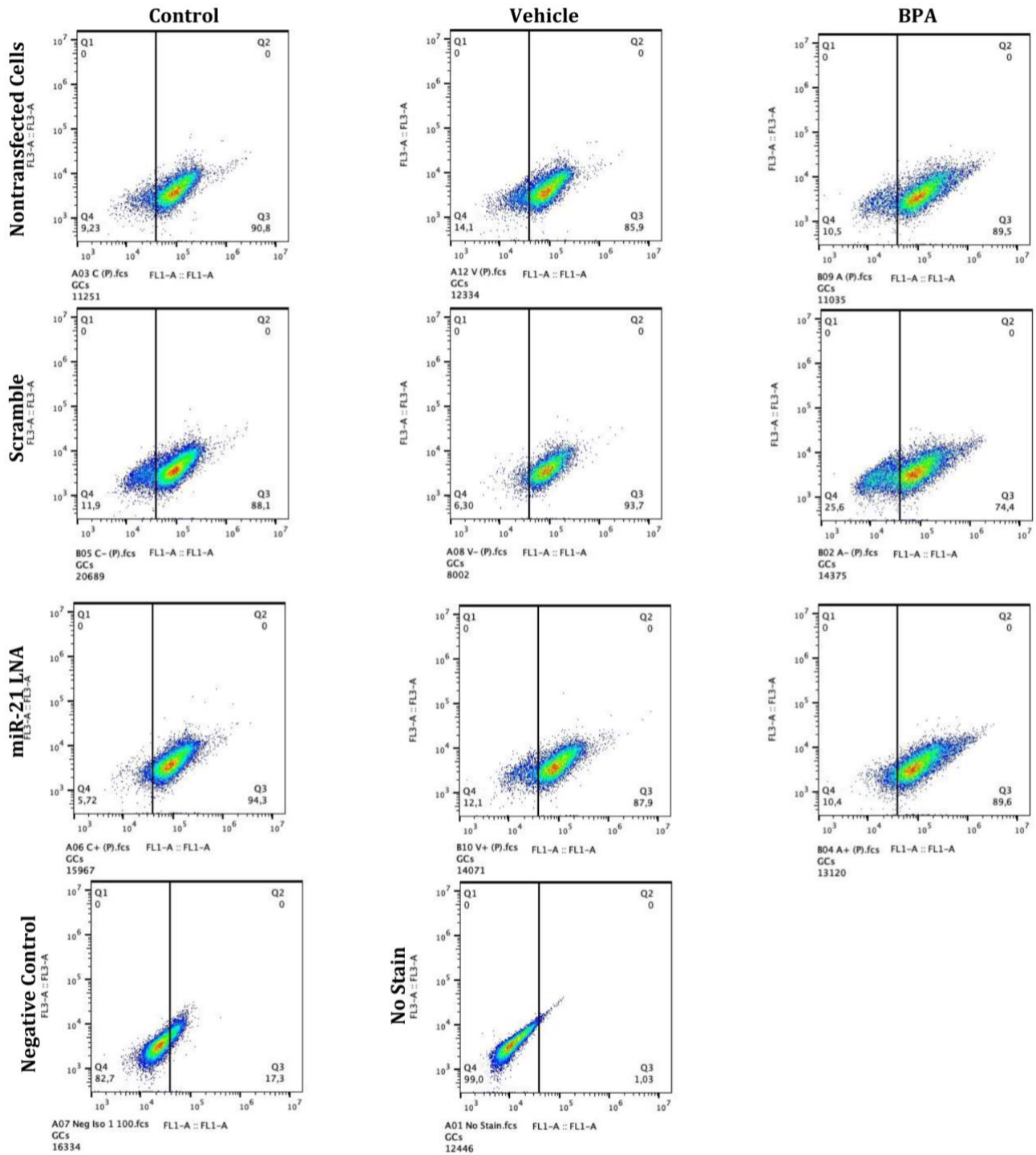
[▼ Next Match](#) [▲ Previous Match](#)

Score	Expect	Identities	Gaps	Strand
202 bits(109)	4e-50	111/113(98%)	0/113(0%)	Plus/Plus
Query 1	TTTCCNTTCCNACAAATGATGACTGAAGCTCCTGACATGGCAGTTATAAACGCACAAGAA	60		
Sbjct 363	TTTCCATTCCAACAAATGATGACTGAAGCTCCTGACATGGCAGTTATAAACGCACAAGAA	422		
Query 61	ATGCCAGCCGAAGTTCCTGCCCCAGCACCTGCTCAGGAACCCACACGAGAGAC	113		
Sbjct 423	ATGCCAGCCGAAGTTCCTGCCCCAGCACCTGCTCAGGAACCCACACGAGAGAC	475		

Supplementary Figure 1 – Nucleotide Blast Results from sequenced TDG product. The product sequence was run through NCBI nucleotide blast resulting in 3 matches with 98% identities to the bovine TDG transcript.



Supplementary Figure 2 – Flow cytometry of 5’methylcytosine (5mC) in Bovine Granulosa Cells. Transfected and treated GCs were fixed and permeabilized to allow for immunostaining of 5mC. Immunodetection of 5mC in GCs was achieved by utilizing the same Alexa Fluor 488 conjugated secondary antibody coupled with flow cytometry. Representative flow histograms are represented in for all 9 groups where the x-axis represents the FITC fluorescence from 5mC and the y-axis represents Hoechst stain from nuclear staining.



Supplementary Figure 3 – Figure 9. Flow cytometry of 5’hydroxymethylcytosine (5hmC) in Bovine Granulosa Cells. Transfected and treated GCs were fixed and permeabilized to allow for immunostaining of 5hmC. Immunodetection of 5hmC in GCs was achieved by utilizing the same Alexa Fluor 488 conjugated secondary antibody coupled with flow cytometry. Representative flow histograms are represented in for all 9 groups where the x-axis represents the FITC fluorescence from 5hmC and the y-axis represents Hoechst stain from nuclear staining.

Supplementary Table 2. Statistic reports from parametric and non-parametric analysis on mRNA data in COCs & GCs.

Gene	Normality	Statistic Report
Cumulus Oocyte Complexes (COCs)		
DNMT1	Skewed	[H(8)=20.0847, p=0.01002]
DNMT3A	Skewed	[H(8)=20.7973, p=0.007706]
TET1	Skewed	[H(8)=19.5026, p=0.01239]
TET2	Skewed	[H(8)=15.7989, p=0.04535]
TET3	Skewed	[H(8)=20.3919, p=0.008951]
TDG	Skewed	[H(8)=22.8694, p=0.003536]
Granulosa Cells (GCs)		
DNMT1	Skewed	[H(8)=20.4656, p=0.008711]
DNMT3A	Skewed	[H(8)=18.0175, p=0.0211]
TET1	Normal	[F(8,45)=3.840836, p=0.00162406]
TET2	Normal	[F(8,45)=2.51995, p=0.0234676]
TET3	Skewed	[H(8)=31.7993, p=0.0001012]
TDG	Skewed	[H(8)=30.0522, p=0.0002069]