

Figure S1. The electrophoretic result of PCR-RFLP band patterns of *RAPGEF6* gene at 15836649 bp in chicken. DNA was digested with the restriction enzyme *Mnll* and three types of genotypes (TT, TC and CC), which are shown above (M: DL600 plus).

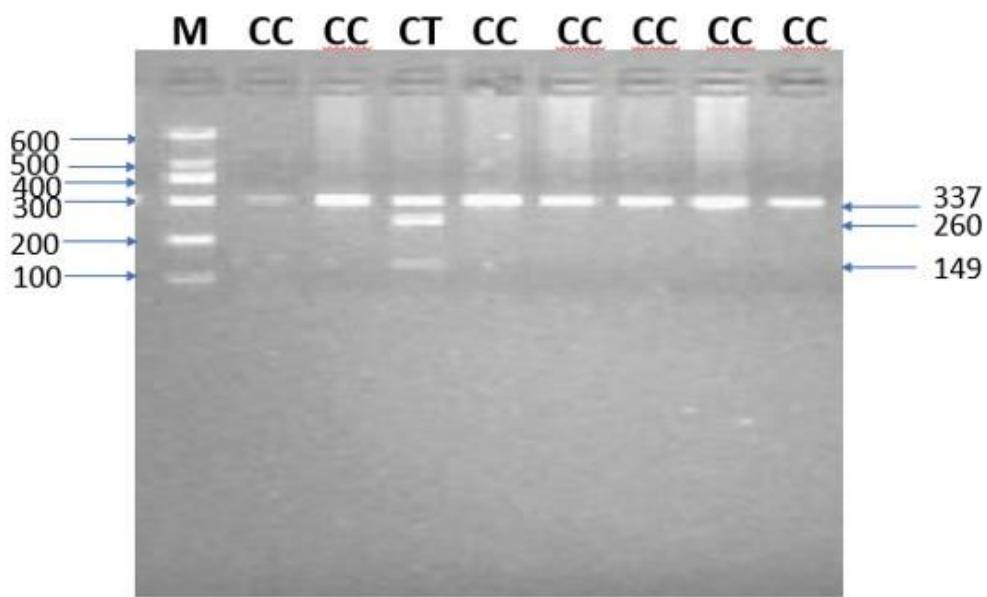


Figure S2. The electrophoretic result of PCR-RFLP band patterns of *RAPGEF6* gene at 15843452 bp in chicken. DNA was digested with the restriction enzyme *MboI* and two types of genotypes (CC and CT), which are shown above (M: DL600 plus).

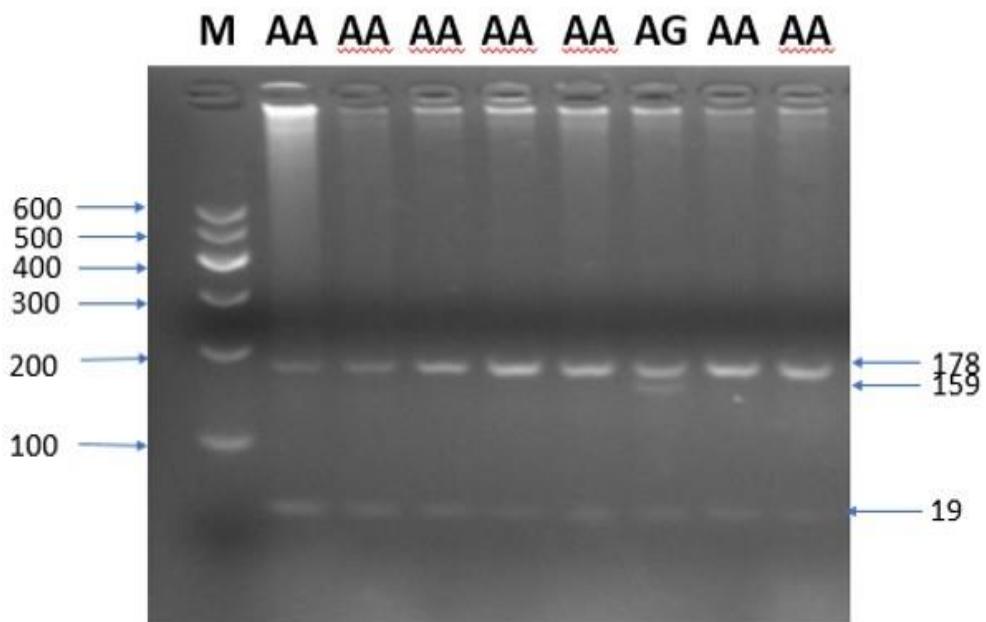


Figure S3. The electrophoretic result of PCR-RFLP band patterns of *RAPGEF6* gene at 15829057 bp in chicken. DNA was digested with the restriction enzyme *XmnI* and two types of genotypes (AA and AG), which are shown above (M: DL600 plus).

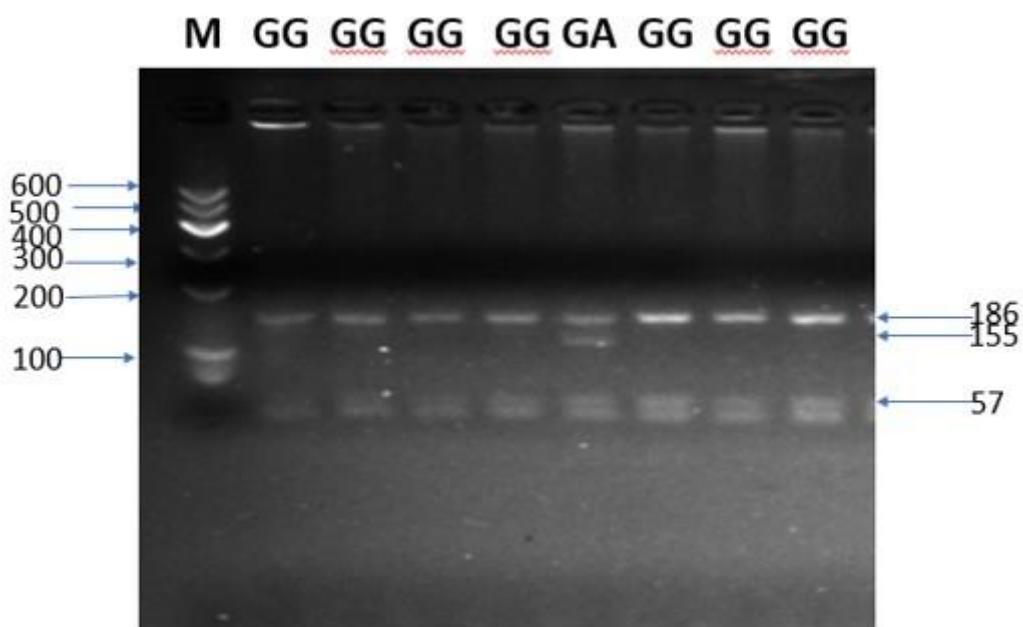


Figure S4. The electrophoretic result of PCR-RFLP band patterns of *RAPGEF6* gene at 15845449 bp in chicken. DNA was digested with the restriction enzyme *MwoI* and two types of genotypes (GG and GA), which are shown above (M: DL600 plus).

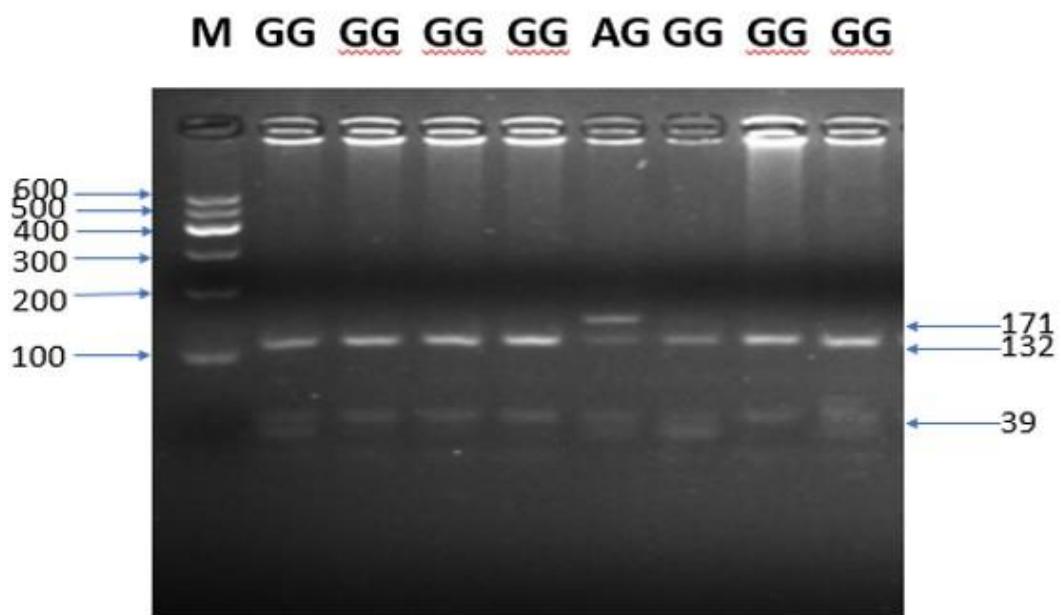


Figure S5. The electrophoretic result of PCR-RFLP band patterns of *RAPGEF6* gene at 15829303 bp in chicken. DNA was digested with the restriction enzyme *HphI* and two types of genotypes (AG and GG), which are shown above (M: DL600 plus).

Supplementary Table S1. Information of five variants (SNPs) in chicken *RAPGEF6* gene.

Sl. No.	Probe Set ID	Affy SNP ID	Position in the genome	Flank	Allele A	Allele B
1	AX-75745366	Affx-50618245	15836649	CCTGTTAGCC[T/C]CCTTAAGAAG	T	C
2	AX-75745380	Affx-50618257	15843452	TCATAACCGA[C/T]CCAATTGC	C	T
3	AX-75745340	Affx-50618223	15829057	AGAGACACAG[A/G]AATGAGTGCC	A	G
4	AX-75745388	Affx-50618264	15845449	TATGAGCCAA[G/A]CCATTACTGC	G	A
5	AX-75745341	Affx-50618224	15829303	TAGCAGGATC[A/G]CCTCAAAGT	A	G

Supplementary Table S2. Primer information for SNP fragments in amplified chicken *RAPGEF6* gene.

Sl. No.	Oligo name	Primers	Sequences (5' to 3')	Base	GC%	Product Length	Annealing Temperature(°C)	nmol/OD	MW
1	EGG-A1-F012	Forward	GCACGAATATGGTTTCAG	20	40.0	261	54.6	5.1	6147.1
	EGG-A1-R253	Reverse	AGGGCTCCTTGGCTTCATT	20	55.0			5.7	6075.0
2	EGG-A2-F064	Forward	GGAGGGAGAGAGGCCACTAA	20	55.0	486	53.5	4.7	6249.1
	EGG-A2-R533	Reverse	CTGGACGTCTGAAAATATG	20	45.0			5.0	6181.1
3	EGG-A3-F148	Forward	CTGTAACACAAGAGACATGAA	21	38.1	178	52.0	4.5	6456.3
	EGG-A3-R305	Reverse	AAAATAGTACACATGGAACTC	21	33.3			4.5	6431.3
4	EGG-A4-F197	Forward	CTCCATTCTATTGTACTT	20	35.0	259	53.5	5.7	5984.0
	EGG-A4-R436	Reverse	TAAAATGTAGAAAGCAGGGT	20	35.0			4.6	6238.2
5	EGG-A5-F171	Forward	TTAACCTTGAAACACACT	20	40.0	197	50.2	5.4	6005.0
	EGG-A5-R348	Reverse	GCTTCTTTCAGGTTACCA	20	40.0			5.5	6049.0

Supplementary Table S3. The ingredients of *RAPGEF6* PCR-RFLP optimized Restriction Enzyme (RE) digestion mixture.

Sl. No.	SNP ID	Restriction Enzyme	Composition of Digestion Mixture (μL)				Digestion Temperature (°C)	% of Agarose Gel
			10 × buffer	PCR products	Enzyme	ddH ₂ O		
1	AX-75745366	<i>Mnl</i> I (10 U/μL)	1.0	5.0	0.03	4.0	37	5
2	AX-75745380	<i>Mbo</i> I (10 U/μL)	1.0	5.0	0.03	4.0	37	5
3	AX-75745340	<i>Xmn</i> I (10 U/μL)	1.0	5.0	0.03	4.0	37	5
4	AX-75745388	<i>Mwo</i> I (10 U/μL)	1.0	5.0	0.03	4.0	60	5
5	AX-75745341	<i>Hph</i> I (10 U/μL)	1.0	5.0	0.03	4.0	37	5

Supplementary Table S4. The restriction enzymes with their recognition site/cut position used to digest the PCR products.

Sl. No.	Position in VLDR	Name of Restriction Enzyme	Recognition Sequence	Recognition Site/Cut Position
1	T15835549C	<i>MnII</i>	5' CCTCN ₇ 3' GGAGN ₆	5' CCTC(N)7↓3' 3' GGAG(N)6↑5'
2	C15843452T	<i>MboI</i>	5' GATC 3' CTAG	5' ↓GATC 3' 3' CTAG↑5'
3	A15829057G	<i>XmnI</i>	5' GAANNNNTTC 3' CTTNNNNAAG	5' GAANN↓NNTTC 3' 3' CTTNN↑NNAAG 5'
4	G15845449A	<i>MwoI</i>	5' GCNNNNNNNGC 3' CGNNNNNNNCG	5' GCNNNNN↓NNGC 3' 3' CGNN↑NNNNNCG 5'
5	A15829303G	<i>HphI</i>	5' GGTGAN ₈ 3' CCACTN ₇	5' GGTGA (N)8↓3' 3' CCACT (N)7↑5'

