www.ThePharmaJournal.com

The Pharma Innovation



ISSN (E): 2277-7695 ISSN (P): 2349-8242 NAAS Rating: 5.23 TPI 2022; SP-11(6): 2859-2863 © 2022 TPI

www.thepharmajournal.com Received: 07-03-2022 Accepted: 10-04-2022

Mandeep Singh Azad

Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu, R.S. Pura, Jammu, Jammu and Kashmir, India

Dibyendu Chakraborty

Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu, R.S. Pura, Jammu, Jammu and Kashmir, India

Kawardeep Kour

Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu, R.S. Pura, Jammu, Jammu and Kashmir, India

Vikas Khajuria

Animal Husbandry Department Rajouri, Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu, R.S. Pura, Jammu, Jammu and Kashmir, India

Corresponding Author

Dibyendu Chakraborty Sher-e-Kashmir University of Agricultural Sciences and Technology of Jammu R.S. Pura, Jammu, Jammu and Kashmir, India

Molecular characterization of inhibitor of apoptosis gene (*IAP*-1) in native chicken of Poonch region from international borders of India and Pakistan

Mandeep Singh Azad, Dibyendu Chakraborty, Kawardeep Kour, Simran Singh, D Kumar, Nishant Kumar, Aditi Lal Koul and Vikas Khajuria

Abstract

Apoptosis of programmed cell death is an important phenomenon of body immune response. Inhibitor of Apoptosis gene (IAP-1) is involved in different biological activities like binding and inhibiting caspases, regulating cell cycle progression, and modulating receptor-mediated signal transduction. The present study was undertaken with the main objectives of molecular characterization of IAP-1 gene in local native chicken of Poonch region. RNA Purification Kit (HiPura) was use for RNA extraction from Poonchi blood. After cDNA synthesis, IAP-1 gene of 544 bp size was amplified from cDNA using specific primers designed by Primer 3 software. Direct sequencing was carried out by Sanger sequencing with both forward and reverse primers to obtain 544 bp sequence product. The sequences were studied by using BioEdit and MEGA X softwares. The sequence results of IAP-I gene of Poonchi chicken and Kadaknath chicken were compared with, Leghorn, Fayoumi breed of chicken and also with other sequence of Gallus gallus complete Cds, Centrocercus urophasianus, Mus musculus, Gallus Gallus partial Cds, Rattus norvegicus, Tympanuchus phasianellus. AT content were higher then GC content for IAP-1 gene. Sequence results were analyzed for synonymous (S) and non-synonymous (NS) changes. No SNP variation within Poonchi population as well as with Kadaknath population were observed. Fayoumi breed showed 2 (NS) changes. Highest number of NS changes were observed with Mus musculus. The Genetic distance result showed no dissimilarity between Poonchi chicken and Kadaknath breed. Highest genetic distance was observed with Tympanuchus phasianellus.

Keywords: IAP-1, sequencing, ClustalW, genetic distance, Poonchi chicken, India

1. Introduction

The complex immune system of poultry provides an opportunity for investigating polygenic regulation of immune response in chickens (Lamont, 1998) ^[10]. Genetic resistance to diseases is a great resource for control and prevention of diseases and for improvement of productivity in poultry (Bacon et al., 2000; Bates et al., 1998)^[2, 3]. Differences in disease resistance between strains of chickens have long been described (Bearse et al., 1939; Lambert, 1932)^{[4,} ^{11]}. Local native chicken breeds are considered more adaptive and disease resistant than the exotic breeds. In recent years exploration of native germplasm has been a focused area worldwide. However, it is difficult to ascertain whether a single or a set of gene (s) define this property to indigenous chickens. There has been special emphasis on development of improved genetic stock for disease tolerance. Native Indian chicken with better immune competence has been indicated earlier by higher complement activity, higher serum lysozyme level, and antibody response (Haunshi and Sharma, 2002; Baelmans et al., 2005)^[9, 1]. The inhibitor of apoptosis protein-1 (IAP-1) gene is a member of the IAP family, which is involved in host antIAPoptotic mechanisms (Deveraux and Reed, 1999). The IAP prevents apoptosis by binding and inhibiting caspase activity at various steps of the apoptotic signaling pathway (Deveraux et al., 1997; Roy et al., 1997)^[6, 13]. Chicken IAP1 has been mapped to chromosome 1, and its sequence has 85% homology to the human IAP gene (Goodenbour et al., 2004)^[7].

Poonch is one of the remotest districts of the Jammu and Kashmir (UT) and situated on line of Control. This native poultry population is quite hardy and thrives well in adverse climatic conditions. It is very imperative to identify and characterize these native chicken population in order to use them for development of disease resistant and early maturing strains to ameliorate the socio-economic status of economically weaker and the poor resource less nomadic and other farmers of the remote district of U.T of Jammu and Kashmir.

Therefore, the present study was undertaken with the objective to characterize the immune related inhibitor of apoptotic protein (*IAP*-1) in Poonchi chicken.

2. Materials and Methods

Fresh blood sample from Poonchi Chicken were collected from the Brachial Vein in EDTA coated vacutainers. RNA isolation was done using RNA Purification Kit (HiPura). Concentration and purity of RNA was checked using Nanodrop-spectrophotometer. Samples having OD ratio (260nm/280nm) of approx. 2 was used for further analysis. Quality of RNA was checked on 1% agarose gel and visualized under transilluminator. Presence of three intact bands of 28s,18s and 5s with smearing indicated good quality and intactness of RNA. cDNA was synthesized using Revert aid first strand cDNA synthesis kit.

Forward primer 5'- GTAACTACTAGGGCTGCCGA-3' and reverse primer 5'- AACTCTCCTCCTTTCACACG-3' were designed using Primer 3 software. PCR was performed in a total volume of 25μ l.

Thermocycler conditions for *IAP-1* gene were 95 °C for 3 min, then 34 cycles of 95° C for 0.30 min, 55 °C for 0.45 min, and 72 °C for 1.30 min, with a final extension step of 5.00 min at 72 °C. Positively amplified PCR products were then checked on 2% agarose gel along with 100bp DNA ladder. The products with size 554 kb were sent to Biologia Research India Pvt Ltd, New Delhi, for sequencing.

2.1 Sequence Analysis

IAP-1 gene sequences for Poonchi chicken were then subjected to BLAST analysis. Sequence was analyzed with MEGAX and BioEdit softwares and compared with other reported sequences of different breeds and species obtained from NCBI (National Center for Biotechnology Information). Multiple sequence alignment was done by ClustalW method using MEGA X software to see the identities, similarities and differences. The phylogenetic tree was constructed using Neighbor-Joining method (Saitou and Nei, 1987)^[14], based on the aligned sequences. The evolutionary distances analyses were conducted using the Maximum Composite Likelihood model (Tamura *et al.*, 2004)^[15]. The relative frequencies of the nucleotide composition were computed for all the sequences used.

3. Results and Discussion

Immune response is mainly controlled by immune system consisting of different cells. It is very important to study and understand the polygenic role of various genes in prevention of disease. Inhibitor of apoptotic protein (*IAP*-1) prevent apoptosis by binding and inhibiting the caspase (Deveraux *et al.*, 1997 & 1998; Roy *et al.* 1997; Takahashi *et al.* 1998; Tamm *et al.*, 1998) ^[5-6, 13, 16] which should act in concert to produce an effective response (Pinard-van der Laan *et al.*, 1998) ^[12].

The sequence results of *IAP*-I gene of Poonchi chicken and Kadaknath chicken were compared with, Leghorn, Fayoumi breed of chicken and also with other sequence of *Gallus gallus* complete Cds, *Centrocercus urophasianus*, *Mus musculus*, *Gallus Gallus partial* Cds, *Rattus norvegicus* and *Tympanuchus phasianellus*. Sequencing results showed no SNP variation within Poonchi population as well as with Kadaknath population (Fig. 1a-e). Also, no variation was observed with Leghorn sequence but variation was seen with Fayoumi breed of chicken.

	4		86				620				30	•	•••	864		•	•••	11	11		1.		866		· ·			70				SBC		1.5		169		11		1			•••	1	11		1.		87:					1 * 731		1	
	<u> </u>		_			8	621			9.6	30			0.65	10			06	50														·			103	ν.			9.1	00			0	111				0.0	20			0	10			
Poonchi 1	9	GOTO	CIG	8.MG3	VCC.	PCT 1	CAC	ACT	7N	161	TCI	AA	act	TC.	NC A	AC	ce:	12.0	CA	76	лœ	7.A.(ag		GA	AG	77	GG	TV	CG.	CA	11	TC	ΞX.	IGI	M	GG	CDO	101	GN	r G i	.11	CT	CT	CG	100	G	CG.	NG	-10	aca e	382	LA A	SC.	rgg	ave.	27
Poonchi 2																																																									
Gadaknath	1.																			•••																												• •	• • •								
Cadaknath	2.																																																• • •								
F221082.1	σ,																																																								
F221083.1	a.		.c.																																												Α.										
F008592.1	ā.		.c.																																																						
(F579274.1	21		.e.				.е.									11	11		11																												λ.		T.							φ.	12
M 007464	- 1		a.c		40	soe	10	Ċ9.	6		2.2	. e	1.0	ĊΤ.			άŪ.	а.	10		a.	e		. 6	а.	e	e.,	. a	1.1	. 9	-	÷.	40	a	12.				æ.		-9			e .	- 2			22				e.,			A	C1	éČ.
2494054 1	°, ľ																																																								
10011000 1	10		181			••••							• • •																																		1										17
P311203.1	10		121																																															•••				•••			
M 023987	~" i				· ·		-																																						121	11	<u> </u>		•••			12.7				۰.	17
DC 023987.	2		· · · ·																																			'						· · ·			<u>ع</u> :	<u>.</u>							***	- 1	· · ·
dF579281.1	т.																				•••					•••			• • •		•••			•••	••••	54.	•••		• • •					•••			•••	•••	- 1	•••		• • •				х.	• •

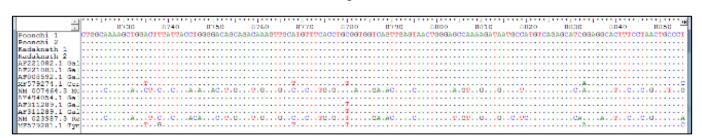


Fig 1a

Fig 1b

																						_																										_
	× 11						1.2.2.2	6 Y 1 Y		111			112		111	11	111				12.5	111		111		111		111		111		111			111		1.22	***	111		1.1.1		1.5.5	4 * * p	1	1 N F		ыł
		1040		1050		117	0450			070			101	11			102	0		11	200			12	10		1	192			11	200			0.94			11	250			11	9440	1		0.2	/70 °	-
Poonchi 1		271700	TAAC	21600	CPT	172	PG27	AAA7	ACC1	CAT	906	1464	CC2	LAC(CAA	GI!	PZC	λM	GT	170	CAA	161	GA	CM	IGC	A2.3	CC	CAT	SAA	800	00	191	ZAA	AAC	ATT	CAT	PTA3	ACP	020	20A7	ACT	031	82.7	/TC:	LAG'	ITC	AGC	C
Poonchi 2	- I-																																															
Kadaknath 1	- I-																																															•
Kadaknath 2	- I.																																															
AF221082.1 (3a1																																							a a								۰.
AF221003.1 (Se 1														• • •	• •												• • •			• • •	• • •			• • •			• • •	• • •							• • •		•
AF008592.1 (Sal														• • •	• •																						• • •										•
xr579274.1 (Cuz				ç.																			_																								-
NM 007464.3	- MG	P C	c.e.		.0.	.CI	.88.	e.	T .	999	1.1	SIC	160	TT	.e.	ъ. В	. А.	.c.		с.,	r.,	cc,				· G.	- 6	C	.с.			. Я.				.10				.1	2	÷.,	190	:A . 7	£		122	•
57494054.1	5a1 -														• • •	• •	• • •																															
AF311289.1	Ga_										• • •					• •																• • •			• • •			• • •	• • •							• • •		•
Ar311289.1	201				121		12.27								•••											121				1.12	• • •	• • •	•••	• • •		• • •			• • •	• • •						• • •		•
NM 023987.3	Ba	e e	c.c.		- 6-	.01	.88.	e.	1501	966	1.4	017	. AC	211	e	AA.	. А.	.c.		c	r.,	66,			• • •	· G.	• •	C	.c.			•••		• • •	a	.10		24.4			2.1	÷.,	000	11.13	IC.		· ¥.	•
MF579281.1	7 ym				c.	${\bf x} = {\bf x}^{T}$																																										- 1



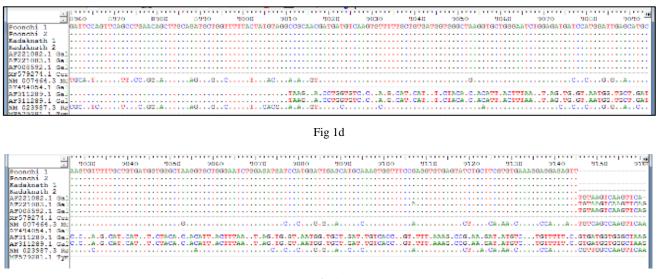


Fig 1e

Fig 1a, b, c, d, e: shows the nucleotide base pair changes between different breeds and species in the present study

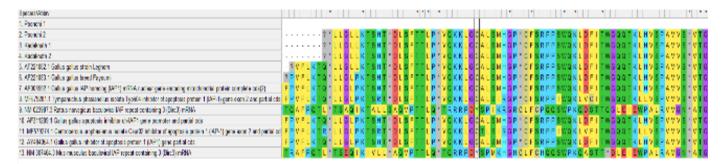
Supakon (2016) studied *IAP*-I gene in Thai native chicken and reported a 394 bp product. PCR-RFLP by using *Bg*II restriction enzymes revealed 3 genotypes and 2 alleles with 0.70 & 0.30; 0.75 & 0.25; 0.72 & 0.28 and 0.77 & 0.23frequencies in Luenghangkhao (LK), Pradhuhangdum (PD), Chee (CH) and Red Jungle Fowls (RJF), respectively. In contrary to the present finding by sequencing, Supakorn (2016) observed within and between population variations. You *et al.* (1997) sequenced the full length chicken *IAP*-1 gene and reported SNPs and synonym and non-synonym changes.

Between different species high variations were observed. In the study of nucleic acid composition of Poonchi chicken and other breeds and species it was observed that AT content were higher then GC content for *IAP*-1 gene in all the breeds and species in the present study (Table 1).

	i deleonde et					
	A%	С%	G%	Т%	G+C%	A+T%
Poonchi_1	25.45	22.74	25.63	26.17	48.38	51.62
Poonchi_2	25.45	22.74	25.63	26.17	48.38	51.62
Kadaknath_1	25.45	22.74	25.63	26.17	48.38	51.62
Kadaknath_2	25.45	22.74	25.63	26.17	48.38	51.62
Leghorn	25.68	22.96	25.00	26.36	47.96	52.04
Fayoumi	25.89	23.35	24.70	26.06	48.05	51.95
Gallus Gallus complete Cds	28.68	20.16	22.27	28.89	42.43	57.57
Centrocercus urophasianus	24.46	26.93	21.67	26.93	48.61	51.39
Mus musculus	27.84	23.05	24.61	24.50	47.26	52.34
Gallus Gallus partial Cds	25.38	27.41	21.32	25.38	48.73	50.76
Gallus Gallus partial Cds	25.75	22.60	24.13	27.52	46.73	53.27
Rattus norvegicus	28.26	23.03	24.73	23.98	47.26	52.24
Tympanuchus phasianellus	24.46	26.93	21.98	26.63	48.92	51.08

Table 1: Nucleotide content for IAP-1 gene

The amino acid sequence results were analyzed for synonymous (S) and non- synonymous (NS) changes. No variations within Poonchi population as well as with Kadaknath population were observed. Synonymous and nonsynonymous changes are shown in Fig. 2. Fayoumi breed showed 2 (NS) changes. The highest number of NS changes were seen with *Mus musculus*a (121) followed by with *Tympanuchus phasianellus* (116) changes with Poonchi chicken. Supakorn (2016) reported G/A synonymous SNP in native Thai chicken. Zhou *et al.* (2001) observed T/A substitution using PCR-RFLP.



The Pharma Innovation Journal

akreaun.																								
1. Proncini 1																								
2 Ponti 2	B C K	IVPCCS	136	FL	TAUL	n (1	SC	ETNQ	VSИ	FPN	' P C	K P <mark>N</mark> ((PVL	€ H S	L T G C	L G F (1 F 5 I	H S I	CN1	L V F	N-	44T	V N S S	VEA
1 Kadakash 1	s o k	INPCOS	136	FL	TALL	i ki	SC	E T N Q	V S M	FPN	' P C I	K P <mark>N</mark> (CP V L	KHS	LTGC	LGEC	FSI	L N S L	CNI	LVE	N.	A A T	<mark>ии</mark> s s	VEA
4. Kadakesth 2	s a k	I V P C C S	136	FL	TALL	n ki	SC	E N N Q	V S M	FPN	' P C I	K P <mark>N</mark> (CP V L	(H S	LTCC	L G F C	1 F 5	L H S L	CNI	LVF	N.	66 T	и и з з	VEA
5. AF221082 1 Galus galus shair Leghan	B G K	I V P C C S	136	FL	TALL	- K)	SC	E T N Q	V S M	FPN	' P C I	(P <mark>N</mark>)	(PVL	K H S	L T G C	L B F C) F 5 (L H S L	CNI	LVF	Иг	4 A T	и и в в	VEA
8 AF2/1001 1 Galus galus bread Fayouni	B C K	I V P C C S	136	T F L	TALL	i ki	S C	E T N Q	V S M	FPN	' P C I	K P <mark>N</mark> ((P <mark>VL</mark>	K H S	LTGC	L G F C	D F S I	L H S L	CN1	LVE	N-	4 A T	<mark>ии</mark> s s	VEA
7. AF00852 1 Galus galus IAP homolog (IAP1) mRNA nucleor gene encoding mitochondrial protein complete cds (2)	s di K	INPCOS	136	FL	TALL	n Ki	sc	E T N Q	V S M	FPN	' P C I	K P <mark>N</mark> (CP V L	< H S	LTGC	LGEC	1 F 5 I	L N S L	CNI	LVF	N	6.6 T	<mark>ии</mark> s s	VEA
IL MF579281.1 Tympanichus plasianellus isolate TyphO4 inhibitor of apoptosis protein 1 (IAP-1) gane even 2 and partial pot	se di K	I V P C C S	IRG	FL	t <mark>a pi</mark> l	n (I	S C	1																
3. HM (22907.2 Ratius nanegicus bacalorini) AP repeat containing 3 (Bird) mRHA	N R	N I L C C S	TSD	i P	A A H B	• K	r v	8 8 <mark>L H</mark>	H T L	ELT:	A C	R P T C	HVL	ERS	L P G L	LAL	FII	L R H L	0 V	RAF.	13	D T V	V T S S	AFA
10. AF311289.1 Galles galles apoptosis inhibitor ch KP1 gave promotes and partial cols	s a k	INPCOS	136	FL	TALL	ιK	sc	E T N Q	V S M	FPN	PC	K P N P	CP V L	KHS	LTGC	L G F C	1 F 5 I	L H S L	CN1	LVF	И	V R K	POVT	DVI
11. NFEREN 1 Certocercus unphasismus isolate Ceu C2 inhibitor of apoptosis protein 1 (AP-1) gave econ 2 and partial of	£CK	I V <mark>P</mark> C C S	IRG	FL	T <mark>a p l</mark>	n ki	SC	7																
12. AV454054,1 Gallus gallus inhibitor of apoptosis protein 1 (AP1) gave partial cels	E G K	I V <mark>P</mark> C C B	136	FL	I A L L	n ()	SC	E î NQ	V S M															
																			1.0.0					



Species/Abbrv																				
1. Poonchi 1																				
2. Poonchi 2	VΜV	/ G	* G	A	G N	LE	ΞM	I H	GΙ	S I	M Q	SG	FR	G \	/ S	I C	F۷	* K	(E I	E S 7
3. Kadaknath 1	VΜV	/ G	* <mark>G</mark>	A	G N	LE	ΞМ	I H	GΙ	. s I	NQ	SG	FR	G ۱	/ S	I C	F۷	* K	(E I	5 S 7
4. Kadaknath 2	VΜV	/ G	* <mark>G</mark>	A	G N	LE	ΞМ	I H	GΙ	. s I	NQ	S G	FR	G ۱	/ S	I C	F۷	* K	(E I	5 S 7
5. AF221082.1 Gallus gallus strain Leghorn	νмν	/G	* <mark>G</mark>	A	ΒN	LE	ΞМ	I H	GΙ	. s I	МQ	S G	FR	G۱	/ S	I C	F۷	* K	(E I	E S I
6. AF221083.1 Gallus gallus breed Fayoumi	νмν	/G	* <mark>G</mark>	A	ΒN	LE	ΞМ	I H	GΙ	. s I	МQ	S G	FQ	G١	/ S	I C	F۷	* K	(E I	ΞSΙ
7. AF008592.1 Gallus gallus IAP homolog (IAP1) mRNA nuclear gene encoding mitochondrial protein complete cds(2)	νмν	/G	* <mark>G</mark>	A	ΒN	LE	ΞМ	I H	GΙ	. s I	NQ	S G	FR	G۱	/ S	I C	F۷	* K	(E I	ΞSΙ
8. MF579281.1 Tympanuchus phasianellus isolate Typh04 inhibitor of apoptosis protein 1 (IAP-1) gene exon 2 and partial cds				-																
9. NM 023987.3 Rattus norvegicus baculoviral IAP repeat-containing 3 (Birc3) mRNA	VΜV	/ G	* D	A	G N	LE	Т	ΤP	G۷	V N I	ΤP	S G	FQ	G١	/ S 1	T *	S E	S K	(<mark>A</mark> I	K S S
10. AF311289.1 Gallus gallus apoptosis inhibitor ch-IAP1 gene promoter and partial cds	HIA	١T	sт	F	ΤL	М	* V	V *	M١	/ A	R Y	VΤ	LΟ	F۲	K G I	R N	D D	νĸ	C I	E C C
11. MF579274.1 Centrocercus urophasianus isolate Ceur02 inhibitor of apoptosis protein 1 (IAP-1) gene exon 2 and partial cd				-																
12. AY494054.1 Gallus gallus inhibitor of apoptosis protein 1 (IAP1) gene partial cds				-										-						
13. NM 007464.3 Mus musculus baculoviral IAP repeat-containing 3 (Birc3) mRNA	VΜV	/ G	* G	A	G N	LE	ΞM	ΤP	G۷	V N <mark>I</mark>	ΜP	S G	FQ	G١	/ S 1	ТС	S E	SK	(<mark>A</mark> I	K N I

Fig 2c

Fig 2 a, b, c: Shows amino acid changes between different breeds and species in the present study

The genetic distance result showed no dissimilarity between Poonchi chicken and Kadaknath and Leghorn breed. Genetic distance of 0.0097 was seen with Fayoumi breed. The highest genetic distance was observed with *Tympanuchus phasianellus* with value of 1.4851. phylogenic tree and Neighbor Joining method was used to infer evolutionary history. In order to compute evolutionary distance Maximum Composite Likelihood method was used. Deveraux and Reed (1999) constructed phylogenetic tree for amino acid sequence of *IAP*-1 gene.

BioEdit (Hall, 1999)^[8] software was used to construct

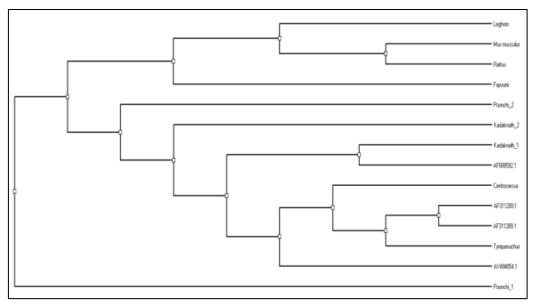


Fig 3: Phylogenetic tree of IAP-1 gene

4. Conclusion

Genetic characterization for *IAP*-1 gene in Poonchi chicken showed no within population variation or any variation with Kadaknath breed but it showed genetic variability between breed and species. Maximum divergence from Poonchi chicken was observed with *Tympanuchus phasianellus*. It can be concluded from the study that there is sustainable variation between the different species and but less variation within the breeds. Genetic characterization on large populations may explore the genetic variability of *IAP*-1 gene in the Poonchi population to establish the disease resistance population for improving the health status of native chicken population.

5. Acknowledgements

Authors are thankful to Farmers of Poonch region rearing the native birds for providing facilities for the present study.

6. References

- 1. Baelmans R, Parmentier HK, Nieuwland MGB, Dorny P, Demey F, Berkvens D. Haemolytic complement activity and humoral immune responses to sheep red blood cells in indigenous chickens and in eight German Dahlem Red chicken lines with different combinations of major genes (dwarf, naked neck and frizzled) of tropical interest. Tropical Animal Health and Production. 2005;37(3):173-186.
- 2. Bacon LD, Hunt HD, Cheng HH. A review of the development of chicken lines to resolve genes determining resistance to diseases. Poult. Sci. 2000;79:1082-1093.
- Bates P, Rong L, Varmus HE, Young JAT, Crittenden LB. Genetic mapping of the cloned subgroup A avian sarcoma and leukosis virus receptor gene to the TVA locus. J. Virol. 1998;72:2505-2508.
- 4. Bearse GE, McClary CF, Miller MW. The results of eight years' selection for disease resistance and susceptibility in White Leghorns. Poult. Sci. 1939;18:4000-4401.
- 5. Deveraux QL, Takahashi R, Salvesen GS, Reed JC. Xlinked *IAP* is a direct inhibitor of cell death proteases. Nature. 1997;388:300–303.
- 6. Deveraux QL, Roy N, Stennicke HR, Van Arsdale T, Zhou Q, Srinivasula M. *IAPs* block apoptotic events induced by caspase-8 and cytochrome c by direct inhibition of distinct caspases. EMBO J. 1998;17:2215–2223.
- Goodenbour JM, Kaiser MG, Lamont SJ. Linkage mapping of inhibitor of apoptosis protein-1 (*IAP* 1) to chicken chromosome 1. Animal Genetics. 2004;35(2):158-159.
- Hall TA. BioEdit: A user-friendly biological sequence alignment editor and analysis program for Windows 95&98NT. Nucleic acid symposium series. 1999;41:95-98.
- 9. Haunshi S, Sharma D. Immunocompetence in native and exotic chicken populations and their crosses developed for rural farming. Indian Journal of Poultry Science. 2002;37(1);10-15.
- 10. Lamont SJ. Impact of genetics on disease resistance. Poultry Science. 1998;77(8):1111-1118.
- 11. Lambert WV. Natural resistance to disease in the chicken: I. The effect of selective breeding on natural resistance to fowl typhoid. The Journal of Immunology. 1932;23(3):229-240.
- 12. Pinard MH, Janss LL, Maatman R, Noordhuizen JP and van der Zijpp AJ.. Effect of divergent selection for immune responsiveness and of major histocompatibility complex on resistance to Marek's disease in chickens. Poult. Sci. 1993;72:391-402.
- 13. Roy N, Deveraux QL, Takashashi R, Salvesen GS, Reed JC.. The c-*IAP*-1 and c-*IAP*-2 proteins are direct inhibitors of specific caspases. EMBO J. 1997;16:6914–6925.
- Saitou N, Nei M. The neighbor-joining method: a new method for reconstructing phylogenetic trees. Molecular Biology and Evolution. 1987;4:406- 425.
- Tamura K, Nei M, Kumar S. Prospects for inferring very large phylogenies by using the neighbor-joining method. Proceedings of the National Academy of Sciences (USA). 2004;101(30):11030-11035.
- 16. Takahashi R, Deveraux Q, Tamm I, Welsh K, Assa-Munt N, Salvesen G, Reed J. A single BIR domain of *XIAP*

sufficient for inhibiting caspases. J. Biol. Chem. 1998;273:7787-7790.

- 17. Tamm IY, Want ES, Scudiero DA, Vigna N, Oltersdorf T, Reed JC. *IAP*-family protein surviving inhibits caspase activity and apoptosis induced by Fas(CD95), Bax, caspases, and anticancer drugs. Cancer Res. 1998;59:5315-5320.
- Zhou H, Liu W, Lamont SJ. Genetic variation among chicken lines and mammalian species in specific genes. Poult. Sci. 2001;80:284-288.