



GENETIC DIVERSITY BETWEEN BALADI CHICKEN BREED AND SASSO AND ROSS STRAINS USING MICROSATELLITE MARKERS

Saadeya S. Mekky³, M. A. El-Sayed^{1*}, Heba A.E.M. Assi² and Zaky³H.I.,

¹. Anim. Gene. Reso. dept., Nat. Gene Bank, Agric. Res. Center, Giza, Egypt

². Anim. Prod. Res. Inst., Agric. Res. Center, Giza, Egypt.

³. Anim. and Poult. Breed. Dep., Desert Res. Center, Matariya, Cairo, Egypt.

Corresponding author: M. A. El-Sayed¹ Email: mohammed.el-Sayed@arc.sci.eg

Received: 01/09/2021

Accepted: 22 /09/2021

ABSTRACT:Sixty individuals from two commercial strains Sasso (n=20), Ross (n=20), and one local chicken breed Baladi (n=20) were amplified by 12 microsatellite markers. Over 12 loci in three chicken populations, we found a total of 96 alleles with an average of 8 alleles per locus. There were 15 alleles in locus ADL0315; and only 3 alleles in locus MCW0210. In specific alleles, overall loci (12 microsatellite loci) found 39 out of 96 alleles (40.63 %) with three populations. Twenty distinct alleles were found for the Baladi breed and 5 alleles for the Sasso strain, while 14 were found for the Ross strain. Most frequencies of alleles were distributed with low frequency, but the 14 allele frequency was higher than 0.5. The *PIC* and *He* averages in the Baladi breed were 0.679 and 0.70, respectively, while those in the Sasso strain were 0.588 and 0.61, respectively, which were the lowest. In Baladi chicken breed has highly informative *PIC* values exceed 0.5 except one marker of MCW0210 had reasonably informative markers. Also, in Sasso chicken strain has highly informative *PIC* values exceed 0.5 except five markers of MCW0210, ADL0037, MCW0206, MCW0002, and ADL0158 had reasonably informative markers. Finally, in Ross chicken strain has highly informative *PIC* values exceed 0.5 except three markers of MCW0210, ADL0037 and MCW0016 had reasonably informative markers. The mean values of *IC* obtained estimate 0.23 and 0.28 for Baladi breed and Sasso strain respectively, indicating the high level of inbreeding indicating low variability in their populations. While the mean value of 0.1 for the Ross breed indicating high variability in this breed. These findings could be attributed to a more recent divergence of Ross than Baladi breed and Sasso strain.

Keywords: Microsatellite, Molecular markers, Baladi local breed, Sasso and Ross strains.

INTRODUCTION

The chicken genome contains several unique characteristics when compared to other vertebrates. Because microsatellite markers are highly polymorphic, they can be used to track the inheritance of alternative alleles in a family tree. The development of DNA markers has made it possible to create effective and reliable genetic linkage maps in the production of livestock species. Linkage maps are primarily used to identify genes that regulate the expression of economically significant traits. Microsatellites are useful indicators for determining gene flow, effective population size (Nm), dispersal and migration concerns, and parentage and relatedness (e.g. Goldstein *et al.*, 1999, Luikart and England, 1999). Microsatellites can also be used to investigate the consequences and level of inbreeding (Pemberton *et al.*, 1999 and Sweigart *et al.*, 1999). As a result of the crossbreeding of native and standard breeds, more than 15 local Egyptian strains of chickens have been established throughout. It is vital to grade and improve the economic features of native strains in Egypt by using commercial foreign breeds (Mohamed, 2003). For crossbreeding with Baladi, a unique breed such as the Sasso from France is acquired. For the next generation, Baladi propagation farms (Ekthar) commonly use enhanced local breeds and on-farm animal selection. For crossbreeding and artificial insemination with hefty breeds like Hubbard males, some people adopt the unusual Sasso breed. Only a few farms use pure native breeds, and each farmer has his exclusive breeding program. More than 450 million eggs were produced by Baladi

propagation farms (Ekthar) in 2004 and 475 million eggs in 2005. Also included in this estimate is an unknown quantity (estimated to be 100 million) generated by commercial hatcheries, as well as poultry produced by broody poultry and the birds leftover from the previous year (FAO (2008). FAO (2009) reported that most of the surveyed modern hatcheries incubate either the improved Baladi breeds that are used in egg production in the rural sector, or the first generation Shamort type used for meat production. This hybrid is a result of crossbreeding between the improved Baladi breed and exotic Sasso and/or Shaver breeds. The crossing between Sasso sires (exotic standard meat-type strain) and dams from (developed local breed) improved significantly body weight at different ages (Amin, 2015 and Soliman *et al.* 2016). It has become more necessary to choose strains that are suitable for raising broilers at lower ambient temperatures in order to reduce the heating costs in broiler farms, especially during the fall and winter seasons in Egypt due to the severe changes in climate during these seasons (Abo ghanema, 2020). The understanding of the population's origins and genetic variability, which could help influence selection decisions (Habimana *et al.* 2020)

The study's objectives were to investigate the genetic diversity between the Baladi chicken breed and two commercial chicken strains (Sasso, Ross), to define the molecular descriptions for these strains using molecular genetics, and to provide a novel avian model for future breeding studies.

Microsatellite, Molecular markers, Baladi local breed, Sasso and Ross strains.

MATERIAL AND METHODS

Chicken Sample Collection and DNA isolation

A total of 60 individuals as 20 blood samples for each of two commercial strains Sasso and Ross and one local breed; Baladi under Sinai environmental conditions, were assayed in the present study which was collected from South Sinai Research Station located at Ras Sudr, Desert Research Center, Matariya, Cairo, Egypt. DNA isolation was carried out as previously described by Ibrahim *et al* (2021) and conserved in National Gene Bank, Agricultural Research Center, Egypt.

Microsatellite loci, PCR and amplification conditions

Twelve microsatellite loci were selected based on the degree of polymorphism (Table 1) according to (Cheng 1996); (Cheng and Crittenden 1994); (Cheng *et al.* 1995); (Crooijmans *et al.* 1996); (Crooijmans *et al.* 1997); (Nahashon *et al* 2008); (Soltan *et al* 2016) and (Cheng 1994) for application in diversity study. The PCR reactions were carried out in 20 μ l as a total volume as described by Roushdy *et al.* (2013a).

Microsatellite and genetic analysis:

All resulted from polyacrylamide gels electrophoresis were visualized and scored with Alphasoft 2200 software (Version 4.0.1) All scored microsatellite data were firstly corrected to estimate each allele size according to its number of repeats. All possible extracted population figures were carried out employing an Arlequin 3.51 software package after data conversion using the Convert program. The absence of PCR products in the samples is manipulated as missing data. As a

consequence, the analysis program accounts for them as null (unknown) alleles not exceeded 0.10 of data as our default analysis and Using POPGEN v.1.32 for calculating private alleles.

RESULTS AND DISCUSSION

For the microsatellite markers used in the study, Table 1 lists the locus, chromosome number, Genbank accession number, Reference, annealing temperatures and band size. MCW (Crooijmans *et al.*, 1996; Groenen *et al.*, 1997), LEI (Gibbs *et al.*, 1997), and ADL markers Cheng *et al.*, (1995) were chosen. According to PIC values, the current study included a set of twelve microsatellite loci with highly polymorphic SSR markers. The genetic diversity within the samples of the three examined chickens (Baladi breed, Sasso and Ross strains) the number of alleles per locus, the mean expected and observed heterozygosity was used to define the genetic diversity within the three chicken populations (Baladi breed, Sasso, and Ross strains).

Ninety-six alleles were detected across 12 loci in three chicken strains/breeds with a mean number of 8 alleles per locus (Table 2). Locus ADL0315 was highly polymorphic with 15 alleles, while locus MCW0210 had the lowest polymorphism of 3 alleles. Our results are similar to Soltan *et al* (2016) who detected that a total of 59 distinct alleles were identified from 10 microsatellite markers in 29 birds with the average number of alleles per locus 5.9. Also, Roushdy *et al* (2013a) reported that the total number of alleles detected across the 9 microsatellites markers was 115 with an average of 4.2 alleles per locus. Ding *et al.* (2010) investigated that a total of 166

alleles were detected at the 29 microsatellite markers in 220 birds. Habimana *et al* (2020) reported that 305 alleles were observed at the 28 microsatellite loci with an average of 10.89 alleles per microsatellite marker. Finally, Ibrahim *et al* (2021) investigated that a total of 62 alleles out of 97 specific alleles (63.92%) was detected overall loci (14 microsatellite loci) for the two strains.

According to Barker (1994), who indicated that the average number of alleles per locus in studies of genetic distances should be > 4 to lower the standard error in the calculation of genetic distances.

As a result, only three locations had an allele number < 4 as MCW0210 (2), MCW0002 (3), MCW0016 (3) as shown in table (2).

Specific alleles there were 39 out of 96 alleles (40.63%) found across all loci (12 microsatellite loci) with three breed genotypes. Twenty specific alleles were found in the Baladi breed, in addition, 5 in the Sasso strain, while 14 were found in the Ross breed. MCW0210 and ADL0024 markers produced no specific alleles either for the Baladi breed and Sasso strain. Also, ADL0106, ADL0315, MCW0206, MCW0002 and ADL0267 produced no specific alleles either for Sasso and Ross strains.

The most specific alleles were found in ADL0315, which has nine different specific alleles. The mean number of alleles per breed varied from 1.67, 0.42 and 1.17 in Baladi, Sasso and Ross populations respectively, (Table 2). As a result, these private alleles would be used as a breed fingerprint, even if only one allele was used for one locus. On the other hand, our values were smallest than reported by Ding *et al* (2010) who

detected Bian chicken had the most private alleles with 15 (46.9%), followed by Jinghai chicken with 12 (37.5%), and Youxi chicken had only 5 private alleles. There were 15 loci with private alleles out of the 29 microsatellite markers (51.7 %). Also, Roushdy *et al* (2013a) reported that there were a total of 30 out of 138 alleles (21.74 %) found in the three strains studied. According to Roushdy *et al* (2013b) analysed the three strains and found a total of 21 out of 115 alleles (18.26%), there were (5) private alleles found in Dokki4, whereas the GM and SM strains contained (14) and (11) respectively. There were seven, five, and nine private number alleles found, respectively. Finally, Habimana *et al* (2020) reported that ADL0112 indicated the highest number of private alleles (60%) out of the total number of alleles (6). The total number of common alleles was 57 with a mean of 4.75. The highest number of common alleles was detected in LEI0166 which had 7 common alleles, while, the lowest in marker MCW0210 which had 2 alleles. Similar results were reported by Roushdy *et al.* (2013a), who investigated the value of 43 common alleles were detected versus 10 microsatellite loci overall strains. On the other hand, Roushdy *et al* (2013b) reported that a total of 40 common alleles were found compared to 9 microsatellite loci.

Allele frequency was ranged from 0.025 to 0.750 as shown in table (3). Most frequencies of alleles were distributed with low frequency, but the 14 allele frequency was higher than 0.5. Three frequencies in Baladi breed with locus (MCW0210, MCW0016 and ADL0267), six in Sasso strain with locus (MCW0210, ADL0037,

Microsatellite, Molecular markers, Baladi local breed, Sasso and Ross strains.

ADL0158, MCW0206 and MCW0002 in two alleles), and five in Ross chicken strain with locus (MCW0210, ADL0037, ADL0024, MCW0016 and ADL0267). Ding *et al.* (2010) reported that Bian chicken had the lowest average observed heterozygote frequency (0.4320), whereas Youxi chicken had the highest (0.4736). Roushdy and El-Sayed (2017) reported that the Silver Montazah and Mandara chicken strains, as well as Beige and Grey quail lines, had mean allele frequencies ranging from 0.13 (UBC005) to 0.50 (GUJ0029) for Silver Montazah and from 0.17 (MCW0167 and UBC002) to 0.50 (MCW0126, MCW0114, and GUJ0029) for Mandara chicken strains, while Beige and Grey quail lines had mean allele frequencies ranging from 0.10 (UBC001) to 0.50 (MCW0127 and MCW0200) for Beige and from 0.13 (MCW0114, GUJ0028 and UBC002) to 1.00 (MCW0200) for Grey quail lines. Ibrahim *et al.* (2021) detected that 1.00 of white jumbo quail strains (WJQS's) highest allele frequencies was 80 at GUJ0048, and the lowest was 65 at GUJ0051. Also, the loci GUJ0013 and GUJ0053 had the highest estimated average allele frequency of 0.33. The lowest was 0.04 at UBC001.

Table 4 summarises the polymorphism information content (*PIC*) and heterozygosity (*He* and *Ho*) for the three chicken populations. Baladi chicken breed had the highest average polymorphism information content (0.679) and an average of heterozygote *He* (0.6973), whereas the Sasso chicken strain had the lowest (0.588 and 0.6059, respectively). Sasso chicken strain had the lowest average *Ho* (0.4445), whereas the Ross chicken strain had the

highest (0.5721). The slight difference between the mean observed compared to the expected heterozygosity (*He*) detected in the present study may reflect slight inbreeding/ and or crossbreed and selection against heterozygotes. Similar results were reported by Roushdy *et al.* (2013b) who investigated that ADL176 had the highest polymorphism information content (*PIC*) value (0.70), while MCW43 had the lowest (0.41). Also, Roushdy *et al.* (2013a) reported that the *PIC* varied from 0.271 for locus MCW73 to 0.7162 for locus ADL176, with a mean of 0.5545 for all loci. Ding *et al.* (2010) reported that the Bian chicken had the highest polymorphism information content (0.5168) and heterozygote frequency (0.5750), while Jinghai chicken had the lowest at 0.4915 and 0.5505. Ibrahim *et al.* (2021) reported that in GUJ0053 and UBC001, the *PIC* for WJQS ranged from 0.19 to 0.89, with a mean of 0.58 in white jumbo quail strain WJQS. In GUJ0053 and UBC001, it varied from 0.32 to 0.89, with a mean of 0.62 in GJQS.

The effective number of alleles (*Ne*) used to corollary the expected heterozygosity (when the heterozygosity is high, the effective number of alleles will be highest), MCW0210 had the lowest *Ne* of 2.24, with *He* of 0.40, 0.45, and 0.43 for the Baladi, Sasso, and Ross strains, respectively. While the highest *Ne* was 8.90 for ADL0315 when *He* were 0.92, 0.80 and 0.79 with Baladi breed, Sasso and Ross strains respectively (Table 4). The same trend was reported by Roushdy and El-Sayed (2017) for the lowest mean of *Ne* was 2.72 and 2.46 for GUJ0029 and MCW0200 when *He* was 0.67 and 0.49 with chicken strains and

quail liens, respectively, while the highest means of N_e were 7.95 and 9.32 for GUJ0028 and UBC001 when H_e were 1.21 and 1.26 with chicken strains and quail lien.

According to Botstein *et al.* (1980), highly informative markers have PIC values greater than 0.50, moderately informative markers have PIC values between 0.25-0.50, and slightly informative markers have PIC values less than 0.25. In Baladi chicken breed has highly informative PIC values of 0.73, 0.70, 68, 90, 0.65, 0.80, 0.73, 0.79, 0.55, 0.61 and 0.63 for ADL0106, ADL0037, ADL0024, ADL0315, MCW0206, LEI0166, MCW0002, MCW0014, MCW0016, ADL0158 and ADL0267 respectively, except one marker of MCW0210 had reasonably informative markers. Also, in Sasso chicken strain has highly informative PIC values of 0.62, 0.64, 0.77, 0.72, 0.76, 0.66 and 0.61 for ADL0106, ADL0024, ADL0315, LEI0166, MCW0014, MCW0016, and ADL0267 respectively, except five markers of MCW0210, ADL0037, MCW0206, MCW0002 and ADL0158 had reasonably informative markers. Finally, in Ross chicken strain has highly informative PIC values of 0.59, 0.66, 0.77, 0.75, 0.69, 0.55, 0.81, 0.69 and 0.56 for ADL0106, ADL0024, ADL0315, MCW0206, LEI0166, MCW0002, MCW0014, ADL0158 and ADL0267 respectively, except three marker of MCW0210, ADL0037 and MCW0016 had reasonably informative markers as shown in table (4). Similar results were obtained by Roushdy *et al.* (2013b) who found that the informative PIC values 0.44, 0.42, 0.41 and 0.44 for ADL136, ADL171,

MCW43 and MCW49, respectively, the rest of markers had highly informative the markers. Colombo *et al.* (2014) reported that the breed had four PIC values greater than 0.5, which are regarded as extremely informative, and nine PIC values between 0.25 and 0.5, which are considered medium informative; the mean PIC value was 0.313, which is considered moderate. Also, Seo *et al.* (2013) investigated that the LEI0251 had the largest number of alleles, H_e , H_o , and PIC for 12, 0.882, 0.852, and 0.865, respectively, while MCW0264 had the lowest H_e and PIC values of 0.709 and 0.648, respectively, but the H_o value was the lowest in GCT0016 marker. Lujiang *et al.* (2006) detected it was found that the average H and PIC values within the 78 populations over-all 27 loci were 0.622 and 0.573, respectively, and the average number of alleles was 6.00 Most polymorphic population was Shuanglian in central China with H and PIC values of 0.678 and 0.633, respectively. Soltan *et al.* (2016) reported that six selected microsatellite loci had an average PIC of over 0.70 and could be employed as genetic markers for the Norfa chicken's genetic diversity research. The F_{is} was measured between -0.626 (MCW0002) and 0.896. (ADL 0106). As indicated in table 4, the P -value ranged from 0.000 (ADL0106, ADL0037, ADL 0024, and MCW0206) to 1.000 (MCW 0002, MCW0014, and MCW0016). The mean values of inbreeding coefficient IC obtained estimate 0.23 and 0.28 for Baladi breed and Sasso strain respectively, indicating the high level of inbreeding indicating low variability in their populations. While, the mean value of 0.1 for the Ross strain indicating high

Microsatellite, Molecular markers, Baladi local breed, Sasso and Ross strains.

variability in these populations. These findings might be due to more recent divergence of Ross strain than Baladi breed and Sasso strain. On the other hand, our value was lower than reported by Roushdy *et al* (2013b) who indicating the high level of inbreeding in strains studied confirming by IC which is equal to 0.48, 0.50 and 0.52, respectively. Habimana *et al* (2020) reported that only 10% of the loci in the IC populations differed significantly from the Hardy-Weinberg equilibrium HWE ($p > 0.05$).

In table 5, presents Analyses of molecular variance (AMOVA), Within-individual genetic diversity accounted for 68.26% of the genetic diversity in the current study. The fixation indices provide information about the population structure, such as the inbreeding coefficient, such there was a range of 0.31744 in the population fixation indexes when comparing individual variance to the total variance (F_{IT}). There was a minimal level of population differentiation, as shown by the lowest fixation indices ($F_{ST} = 0.19818$) across populations. These results are in agreement with Roushdy *et al* (2013a) for Dokki4, GM and SM breeds. On the other hand, this study value was higher than reported by Roushdy *et al* (2013b) for Mamourah, Mandara and Baheij strains and lower than reported by Roushdy *et al* (2013c) for Matrouh, El-salam and Bandarah strains.

Based on Nei's genetic distance, cluster analysis revealed that the studied populations were divided into two major groups (Fig. 1). The 1st group included the Baladi breed and Sasso strain our result agrees with (Mohamed, 2003) who reported that a special breed such as the Sasso from France is imported for crossing with Baladi. And the 2nd

group harbored Ross chicken strain. Chicken breed diversity can be revealed through genetic analysis (Nassiri *et al.*, 2007; Semik and Krawczyk, 2011) to make an accurate assessment of each breed when deciding on conservation and breeding programmes, further information on special adaptations, distinguishing morphologies, performance level, demography (including effective population size, and geographical distribution) and descriptive are needed (Groeneveld *et al.* 2010).

CONCLUSION

It could be concluded that the 12 microsatellite markers were suggested to serve genetic diversity on multiple levels, including conservation of such genetic resources, future improvements for these three poultry species. Results confirmed that microsatellite markers could be strongly utilized as a molecular tool in fingerprint analysis for two commercial strains and one local chicken. The current study proposes using a large genome scan analysis based on more approved microsatellites to cover the entire chicken genome, which could be useful in future MAS (marker-assisted selection) systems. The best utilization of such data can aid in the preservation of allelic diversity and genetic variation. The acquired results appear to be promising in terms of defining and controlling the continuing conservation effort for animal genetic resources, improvements in the future for these groups.

Table (1): Information on Locus, chromosome number, Genbank accession number, Reference, annealing temperatures and band size for microsatellite markers used in the study.

Locus	Chromosome	GenBank accession number	Reference (a)	Tm	band size
MCW0210	5	G31987	5	58	140-204
ADL0106	10	G01550	3	55	135-180
ADL0037	1	L23912	2	57	160-224
ADL0024	3	L23906	2	58	140-188
ADL0315	7	G16117	1	58	254-399
MCW0206	--	AF030579	7	60	205-268
LEI0166	--	X85531	7	60	300-498
MCW0002	C4	---	4	50	140-176
MCW0014	E11	---	4	55	180-260
MCW0016	E2	---	4	50	85-181
ADL0158	--	G01582	6	52	170-230
ADL0267	--	---	8	55	105-153

a) Reference papers: 1(Cheng 1996); 2(Cheng and Crittenden 1994); 3(Cheng et al. 1995); 4(Crooijmans et al.1996); 5 (Crooijmans et al. 1997); 6(Nahashon et al 2008); 7 (Soltan et al 2016) and 8 (Cheng 1994).

Microsatellite, Molecular markers, Baladi local breed, Sasso and Ross strains.

Table (2): Number of alleles observed for each locus within each breed, Private alleles, Common alleles and total number of alleles.

L o c u s	Private alleles			Common alleles	Total number of alleles
	Baladi	Sasso	Ross		
MCW0210	-	-	1	2	3
ADL0106	1	-	-	5	6
ADL0037	2	-	2	5	9
ADL0024	-	-	3	6	9
ADL0315	9	-	-	6	15
MCW0206	2	-	-	6	8
LEI0166	2	-	3	7	12
MCW0002	1	-	-	3	4
MCW0014	1	4	2	6	13
MCW0016	1	-	2	3	6
ADL0158	-	1	1	4	6
ADL0267	1	-	-	4	5
Mean	1.67	0.42	1.17	4.75	8
Total	20	5	14	57	96

Table (3): Allele size in base pair, their frequencies for each locus and chicken breed as observed in the present study.

L o c u s	Alleles (bp)	Frequencies			L o c u s	Alleles (bp)	Frequencies		
		Baladi	Sasso	Ross			Baladi	Sasso	Ross
MCW0210	140	0.2500	0.6765	0.0556	LEI0166	300*	0.0500	-	-
	172	0.7500	0.3235	0.7222		318	0.1750	0.0278	-
	204*	-	-	0.2222		336*	0.0250	-	-
						354	0.3000	0.0278	-
						372	0.2500	0.2222	-
						390	0.0750	0.4444	0.2632
						408	0.0500	0.0556	0.0263
						426	0.0750	0.1667	0.4737
						444	-	0.0556	0.1053
						462*	-	-	0.0789
						480*	-	-	0.0263
				498*	-	-	0.0263		
ADL0106	135	0.1750	0.1765	-	MCW0002	140	0.3000	-	0.0526
	144	0.3250	0.3529	-		152	0.2250	0.5000	0.4737
	153	0.3500	0.4706	0.4211		164*	0.1500	-	-
	162	0.0750	-	0.4737		176	0.3250	0.5000	0.4737
	171	0.0250	-	0.1053					
	180*	0.0500	-	-					
ADL0037	160*	-	-	0.0526	MCW0014	180*	-	0.0526	-
	168*	-	-	0.1053		188*	-	0.1316	-
	176	-	0.2778	0.7368		196*	-	0.3684	-
	184	0.1500	0.6667	-		204	0.3250	-	0.2500
	192	0.2500	-	0.0789		212	0.1750	0.0789	0.1944
	200	0.4500	0.0278	0.0263		220	-	0.1316	0.0833
	208	0.0500	0.0278	-		228*	-	0.2368	-
	216*	0.0750	-	-		236	0.1250	-	0.0278
	224*	0.0250	-	-		244	0.1750	-	0.2222
						246*	-	-	0.0278
						248*	0.0250	-	-
				252	0.1750	-	0.1667		
				260*	-	-	0.0278		
ADL0024	140*	-	-	0.0500	MCW0016	85*	-	-	0.3000
	146*	-	-	0.5250		101*	-	-	0.7000
	152	0.0526	-	0.2250		133	0.0250	0.3056	-
	158	0.3947	0.4444	-		149	0.5000	0.3889	-
	164	0.0789	0.3611	-		165	0.4500	0.3056	-
	170	0.3947	0.1667	0.1000		181*	0.0250	-	-
	176*	-	-	0.0250					
	182	0.0263	-	0.0750					

Microsatellite, Molecular markers, Baladi local breed, Sasso and Ross strains.

Continue table (3):

	188	0.0526	0.0278	-					
ADL0315	255	-	0.0937	0.1842	ADL0158	170*	-	-	0.0500
	264	-	0.3438	0.3421		182	0.4737	-	0.4500
	273	-	0.2188	0.0263		194	0.3947	0.2105	0.1500
	291*	0.0250	-	-		206	0.1053	0.7368	0.2750
	300	0.0250	-	0.1842		218	0.0263	-	0.0750
	309	0.1000	-	0.2105		230*	-	0.0526	-
	318	0.0750	0.0625	0.0526					
	327	0.1250	0.2188	-					
	336	0.1500	0.0625	-					
	354*	0.0500	-	-					
	363*	0.1000	-	-					
	372*	0.0500	-	-					
	381*	0.0500	-	-					
	390*	0.1500	-	-					
	399*	0.1000	-	-					
MCW0206	205*	0.0500	-	-	ADL0267	105	0.0263	0.1579	0.5263
	214*	0.4000	-	-		117	0.5263	0.4737	0.3947
	223	0.4250	-	0.0263		129	0.2632	0.3684	0.0263
	232	0.0250	0.0500	0.2632		141	0.1579	-	0.0526
	241	0.1000	0.7250	0.3421		153*	0.0263	-	-
	250	-	0.1500	0.1316					
	259	-	0.0500	0.2105					
	268	-	0.0250	0.0263					

* Private alleles

Table (4): Polymorphism information content (PIC), heterozygosity (He and Ho) for three chicken populations

Loci	Baladi			Sasso			Ross			Fis	P-value	Effective number of alleles (Ne)*
	PIC	He	Ho	PIC	He	Ho	PIC	He	Ho			
MCW0210	0.38	0.4000	0.0000	0.44	0.4510	0.5294	0.43	0.4381	0.4444	0.097	0.327	2.2371
ADL0106	0.73	0.7513	0.2000	0.62	0.6417	0.0000	0.59	0.6031	0.0000	0.896	0.000	3.7112
ADL0037	0.70	0.7218	0.2000	0.48	0.4905	0.1111	0.44	0.4481	0.2105	0.691	0.000	4.4024
ADL0024	0.68	0.6942	0.2105	0.64	0.6619	0.0556	0.66	0.6718	0.5000	0.617	0.000	5.3394
ADL0315	0.90	0.9179	1.0000	0.77	0.7984	0.6875	0.77	0.7881	0.8947	-0.042	0.837	8.8971
MCW0206	0.65	0.6628	0.2000	0.45	0.4577	0.0500	0.75	0.7710	0.7895	0.467	0.000	4.5003
LEI0166	0.80	0.8205	0.9000	0.72	0.7381	0.5000	0.69	0.7055	0.2632	0.263	0.001	5.8966
MCW0002	0.73	0.7641	1.0000	0.50	0.5128	1.0000	0.55	0.5633	0.9474	-0.626	1.000	2.7759
MCW0014	0.79	0.8064	1.0000	0.76	0.7852	0.8947	0.81	0.8365	0.9444	-0.177	1.000	8.2462
MCW0016	0.55	0.5603	1.0000	0.66	0.6810	0.6111	0.42	0.4308	0.6000	-0.353	1.000	4.3831
ADL0158	0.61	0.6245	0.2632	0.41	0.4211	0.1053	0.69	0.7090	0.8500	0.301	0.001	3.3556
ADL0267	0.63	0.6444	0.5263	0.61	0.6316	0.7895	0.56	0.5789	0.4211	0.065	0.299	3.0738
Mean	0.679	0.70	0.54	0.588	0.61	0.44	0.613	0.63	0.57			4.7349
St. Dev		0.1349	0.4045		0.1347	0.3641		0.1410	0.3172			2.0768
IC	0.23			0.28			0.1					

PIC = Polymorphism information content; He = Expected heterozygosity; Ho = Observed heterozygosity.

IC: Inbreeding Coefficient (IC= (He- Ho)/He).

*Ne = Effective number of alleles [Kimura and Crow (1964)].

Microsatellite, Molecular markers, Baladi local breed, Sasso and Ross strains.

Table (5): AMOVA analysis of Baladi breed, Sasso and Ross chicken strains based on 12 microsatellite markers.

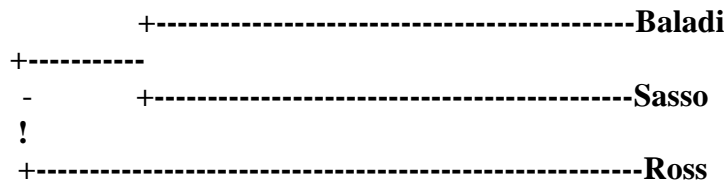
Source of variation	d.f.	S.S.	Percentage variation	Fixation indices
Among populations	2	73.675	19.82	FIS : 0.14873
within populations	57	218.575	11.93	FST : 0.19818
Within individuals	60	170.500	68.26	FIT : 0.31744
Total	119	462.750		

F_{IS}: Fixation indices (Among populations)

F_{IT}: Fixation indices (Within individuals)

F_{ST}: Fixation indices (Among individuals within populations)

Figure (1): Dendrogram Based Nei's (1978) Genetic distance of three chicken breeds produced by UPGMA clustering based on Nei's genetic distance using 12 microsatellite loci.



REFERENCES

- Abo ghanema, Mahmoud M. 2020.** Effect of cold stress on growth performance, carcass traits, blood parameters and antioxidant enzymes in different broilers strains. *Damanhour Journal of Veterinary Sciences* 3 (1), (2020) 17–22.
- Amin, E. M. 2015.** Genetic components and heterotic effect of growth traits in 3x3 diallel crossing experiment in chickens. *Journal of American Science*: 11(1).
- Barker. J.S.F. 1994.** A global protocol for determining genetic distance among domestic livestock breeds. *Proceedings of the 5th World Congress on Genetics Applied to Livestock Production, (GALP'94)*. Guelph. Ontario. Canada. pp: 501-508.
- Botstein, D.; R.L.White; M.Skolnick; R.W. Davis. 1980.** Construction of a genetic linkage map in man using restriction fragment length polymorphisms. *American Journal of Human Genetics*, v.32, p. 314-331.
- Cheng H.H., Levin I., Vallejo R.L., Khatib H., Dodgson J.B., Crittenden L.B., Hillel J. 1995.** Development of a genetic map of the chicken with markers of high utility. *Poultry Sci.*, 74, 1855–1874.
- Cheng, Hans H. 1994.** Microsatellite Markers for Genetic Mapping in the Chicken. *Poultry Science* 73:539-546
- Colombo, Elena, Maria G. Strillacci, Maria C. Cozzi, Manuela Madeddu, Maria G. Mangiagalli, Fabio Mosca, Luisa Zaniboni, Alessandro Bagnato, Silvia Cerolini. 2014.** Feasibility study on the FAO chicken microsatellite panel to assess genetic variability in the turkey (*Meleagris gallopavo*). *Italian Journal of Animal Science* 2014; 13:3334 doi:10.4081/ijas.2014.3334.
- Crooijmans, R. P. M. A., A. F. Groen, A. J. A. Vankampen, S. Van Derbeek, J. Van Derpoel and M. A. M. Groenen. 1996.** Microsatellite polymorphism in commercial broiler and layer lines estimated using pooled blood samples. *Poult. Sci.* 75: 904-909.
- Ding, Fu-Xiang, Gen-Xi Zhang, Jin-Yu Wang, Yuan Li1, Li-Jun Zhang, Yue Wei Hui-Hua Wang1, Li Zhang and Qi-Rui Hou. 2010.** Genetic Diversity of a Chinese Native Chicken Breed, Bian Chicken, Based on Twenty-nine Microsatellite Markers. *Asian-Aust. J. Anim. Sci.* Vol. 23, No. 2: 154 – 161.
- FAO. 2008.** Poultry sector country review. FAO Animal Production and Health Division, Food and Agriculture Organization of the United Nations. Prepared By Farid A. Hosny.
- FAO. 2009.** Mapping traditional poultry hatcheries in Egypt. Prepared by M. Ali Abd-Elhakim, Olaf Thieme, Karin Schwabenbauer and Zahra S. Ahmed. AHBL - Promoting strategies for prevention and control of HPAI. Rome.
- Gibbs M., Dawson D.A., McCamley C., Wardle A.F., Armour J.A.L., Burke T. 1997.** Chicken microsatellite markers isolated from libraries enriched for simple tandem repeats. *Anim. Genet.*, 28, 401–417.
- Goldstein, D. B.; Roemer, G. W.; Smith, D. A.; Reich, D. E. and Wayne, R. K. 1999.** The use of microsatellite variation to infer population structure and

Microsatellite, Molecular markers, Baladi local breed, Sasso and Ross strains.

- demographic history in a natural model system. *Genetics*, 151, 797–801.
- Groenen M.A.M., Crooijmans R.P.M.A., Veenendall A., Van Kaam J.B.C.H.M., Vereijken A.L.J., Van Arendonk J.A.M., Van der Poel J.J. 1997.** QTL mapping in chicken using three generation full sib family structure of the extreme broiler × broiler cross. *Anim. Biotech.*, 8, 41–46.
- Groeneveld LF, J.A, Lenstra, H. Eding M.A. Toro; B. Scherf; D. Pilling; R. Negrini; EK Finlay; H. Jianlin; E. Groeneveld et al. 2010.** Genetic diversity in farm animals – A review. *Anim Genet* 41(Suppl 1):6-31.
- Habimana R, Okeno TO, Ngeno K, Mboumba S, Assami P, Gbotto AA, et al. 2020.** Genetic diversity and population structure of indigenous chicken in Rwanda using microsatellite markers. *PLoS ONE* 15(4): e0225084. <https://doi.org/10.1371/journal.pone.0225084>.
- Ibrahim, Nashat Saeid, M.A. El-Sayed, Heba A.E.M. Assi, Enab, A.A and Abdel-Moneim E. Abdel-Moneim. 2021.** Genetic and physiological variation in two strains of Japanese quail *Journal of Genetic Engineering and Biotechnology*, 19-15, DOI: 10.1186/s43141-020-00100-3.
- Luikart, G. and England, P. R. 1999.** Statistical analysis of microsatellite DNA data. *Tree*, 14: 253-256.
- Lujiang, QU , LI Xianyao , XU Guifang , CHEN Kuanwei , YANG Hongjie , ZHANG Longchao , WU Guiqin, HOU Zhuocheng , XU Guiyun & YANG Ning. 2006.** Evaluation of genetic diversity in Chinese indigenous chicken breeds using microsatellite markers. *Science in China Series C: Life Sciences* 2006 Vol.49 No.4 332—341, DOI: 10.1007/s11427-006-2001-6.
- Mohamed, A.A. 2003.** Effect of diallel crosses on poultry performance. M. Sci. Thesis, Faculty of Agriculture, Alexandria University.
- Nahashon. Samuel N., Akuley Amenyenu, Callie Harris and Nathaniel Adefope. 2008.** Chicken and quail Microsatellite Markers Reveal Polymorphisms in Guinea Fowl *The Journal of Poultry Science*, 45: 249-254.
- Nassiri MTB; Z. Hamid and S. Tavakoli. 2007.** The investigation of genetic variation at microsatellite loci in Mazandranna- tive chickens. *J PoultSci* 6:675-678.
- Nei, M. 1978.** Estimation of average heterozygosity and genetic from a small number of individuals. *Genetics*, 89: 583-590.
- Pemberton, J. M.; Coltman, D. W.; Coulson, T. N. and Slate, J. 1999.** Using microsatellites to measure the fitness consequences of inbreeding and outbreeding. In *Microsatellites, Evolution and applications* (ed. Goldstein & Schlotterer), pp. 151-164. Oxford: Oxford University Press.
- Roushdy, Kh, M.A.El-Sayed and A.A.Bakir. 2013a.** Comparative chicken genome analysis of Egyptian local breeds and developed strains.3.The microsatellite discrimination between Dokki-4, Golden Montazah and Silver Montazah strains. *Egypt. poult. Sci.*33: 971-983.
- Roushdy, Kh, M.A.El-Sayed and A.A.Bakir. 2013b.** Comparative chicken

- genome analysis of Egyptian local breeds and developed strains.4.The microsatellite discrimination between Mamourah, Mandara and Bahij strains. Egypt. Poult. Sci.33: 985-997
- Roushdy, Kh, M.A.El-Sayed and A.A.Bakir. 2013c.** Comparative chicken genome analysis of Egyptian local breeds and developed strains.5.The microsatellite discrimination between Matrouh,El-Salam and Bandarah strains.J. Animal and Poultry Prod.,Mansoura Univ.4 (11) :701-713.
- Roushdy, Kh. and M.A. El-Sayed. 2017.** Genetic Assessment of Two Chicken Strains and Two Japanese quail Lines by Using Chicken and Quail Microsatellite Markers Reciprocally. Egypt. Poult. Sci. Vol (37) (I): (21-36).
- Semik, E. and J. Krawczyk. 2011.** The state of poultry genetic resources and genetic diversity of hen populations. Ann. Anim. Sci 11:181-191.
- Seo, D. W., M. R. Hoque, N. R. Choi, H. Sultana, H. B. Park^{1,2}, K. N. Heo³, B. S. Kang³, H. T. Lim^{1,2}, S. H. Lee⁴, C. Jo and J. H. Lee. 2013.** Discrimination of Korean Native Chicken Lines Using Fifteen Selected Microsatellite Markers. Asian-Aust. J. Anim. Sci. Vol. 26, No. 3: 316-322.
- Soliman, A.S.; M.A. Kosba; M. K. Sheb; M. Bahie El-Deen and T.S. Rabie. 2016.** Studing of some productive characters in across between Alexandria, Saso and Fayoumi chekens.Egyp.Poult. Sci. Vol (36) (II): 465 – 479.
- Soltan, M. E.; A. A. Enab; Eman M. Abou-Elewa and S. A. Farrag. 2016.** Genetic Characterization of a local Egyptian Chicken Strain Norfa Using Microsatellite Markers. 9th International Poultry Conference, 7-10 November 2016, Hurghada, Red Sea – Egypt.
- Sweigart, A.; Karoly, K.; Jones, A. and Willis, J. H. 1999.** The distribution of individual inbreeding coefficients and pairwise relatedness in a population of *mimulus guttatus*. Heredity, 83 (Pt 5):625-632.

الملخص العربي

التنوع الوراثي بين سلالة الدجاج البلدي وسلالات الساسو والروس باستخدام واسمات الميكروستالايت

سعدية سعد الدين مكي3 محمد احمد السيد حسن1 هبة عبد الوهاب محمود عاصي
2 حسن اسماعيل زكي3

1. قسم الموارد الوراثية الحيوانية - البنك القومي للجينات- مركز البحوث الزراعية
2. بقسم بحوث تربية الدواجن - معهد بحوث الانتاج الحيواني- مركز البحوث الزراعية
3. قسم الانتاج الحيواني و الداجني - مركز بحوث الصحراء

تم استخدام عدد 60 عينة من سلالتين تجاريتين (عدد = 20 الساسو) ، (وعدد = 20الروس) ، وسلالة محلية واحدة بلدي (بعدد = 20) باستخدام 12 واسما جزيئي. تم اكتشاف عدد 96 أليلاً من خلال الـ 12 موقعاً في ثلاث مجموعات دجاج بمتوسط عدد 8 أليلات لكل موضع. اظهرالكاشف الجزيئي ADL0315 تنوع بدرجة عالية 15 أليل ، بينما كان الكاشف MCW0210 أقل تنوعاً حيث كانت 3 أليلات فقط. تم اكتشاف عدد 39 اليل متخصص من أصل 96 أليلات بنسبة (40.63 %) من الـ (12 كاشف جزيئي) من الثلاث سلالات. بالنسبة لسلالة البلدي وسلالة ساسو لوحظ 20 اليل و 5 أليل مميز لكليهما بينما تم الحصول على 14 أليل مميز لسلالة الروس . تم توزيع معظم تكرار الأليلات بشكل منخفض ، حيث وجد ان هناك 14 أليل كان أعلى من 0.5. وكان متوسط PIC (0.679) ومتوسط He (0.6973) في سلالة الدجاج البلدي الأعلى ، وكانت المتوسطات لسلالة دجاج الساسو هي 0.588 و 0.6059 على التوالي حيث كان الأقل قيمة. قيم PIC غنية بالمعلومات تجاوزت 0.5 باستثناء كاشف جزيئي واحد- MCW0210 له علامات قيمة معقولة. أيضاً ، في سلالة الساسو تحتوي على قيم PIC مفيدة للغاية تتجاوز 0.5 باستثناء خمسة كواشف MCW0210 و ADL0037 و MCW0206 و MCW0002 و ADL0158 تحتوي على قيم معقولة. أخيراً ، في سلالة الروس لديها قيم PIC غنية بالمعلومات تتجاوز 0.5 باستثناء ثلاثة علامات MCW0210 و ADL0037 و MCW0016 لها قيم معقولة. تقدر القيم المتوسطة لـ IC التربية الداخلية لسلالة البلدي وسلالة الساسو 0.23 و 0.28 على التوالي ، مما يشير إلى ارتفاع مستوى التزاوج الداخلي مما يشير إلى انخفاض التباين في عشائرها. بينما يشير متوسط القيمة 0.1 لسلالة الروس إلى تباين كبير في هذه السلالة. قد تكون هذه النتائج بسبب الاختلاف الأخير بين سلالة الروس وكلا من سلالة البلدي وسلالة الساسو.