

*Poultry Genetic Resources of India*

# ASEEL



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**(Indian Council of Agricultural Research)**

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## Introduction

The domestication of chicken dates back to 1400 B.C. Since time immemorial the chicken were reared in small flocks for home consumption until 20<sup>th</sup> Century when poultry farming was developed on commercial lines. The chickens were subsequently bred for egg/meat production. In India, the poultry is traditionally reared as backyard poultry on zero input system. The poultry is reared for meat, egg and as game birds especially in the tribal belts. Rearing of poultry in the distant regions is culturally associated with the tribes. Widespread geographical distribution and varied climatic regions have led to the development of different population structure/breeds. Aseel is among the several breeds found in India.

“Aseel” is an Arabic word meaning “pure” or “thoroughbred”. The Aseel gamefowl breed might well be 3,500 years old as cockfighting has been mentioned in an Indian manuscript “Manusriti” of the same antiquity ([www.calcuttaaseelclub](http://www.calcuttaaseelclub)). Probably descended from the Indian red Jungle fowl it has been moulded through countless generations of special selection, always on the same lines, into a magnificent warrior. Aseel or Malay fowl



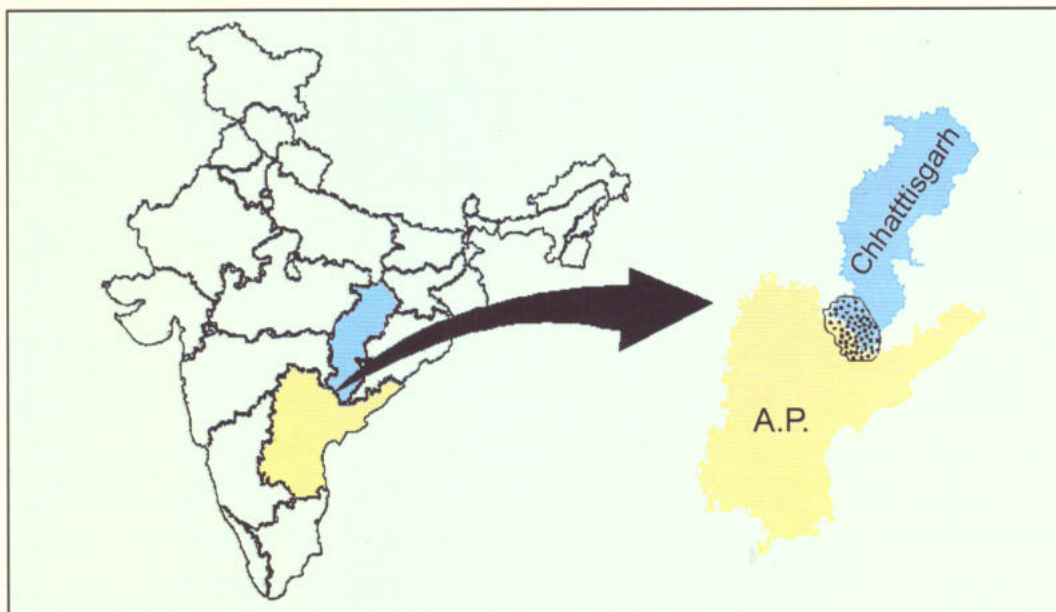
*A typical Aseel bird*

have given rise to all the present day breeds of poultry. European breeds have been developed from this fowl. Aseel is an important indigenous breed of poultry reputed for its fighting tendency and delicious meat quality. It has an excellent power for heat tolerance and disease resistance.

The unique characteristics of the breed are result of evolutionary forces and their interactions over longer period of time. However, these adaptation and unique characteristics might have been diluted due to intermixing, sub-structuring and/or consequent genetic drift in the population over time. There is a worldwide recognition of the need for the conservation of livestock diversity (FAO, 1995) and for characterization of breeds and populations including their genetic differentiation and relationships.

### Distribution

Main breeding tract of Aseel includes North Bastar and South Bastar of Chattisgarh and Khammam district of Andhra Pradesh. Average land



*Distribution of Aseel*



holding of farmers is 4.67 acres in its breeding tract. Out of five one member of the family is engaged in poultry farming.

## Climate

Elevation of land in the breeding tract is 550-760m above mean sea level.

	Average	Range
Temperature (°C)	27	11-40
Relative humidity (%)	51	25-86
Annual rainfall (cm)	130	120-160

## Farmer's Background

Aseel is mainly reared by tribals of Bastar and adjoining Andhra Pradesh. This breed flourished under the patronage of Nawabs, Jamindars and kings of this area. The contribution of King of Vijaynagram is worth mentioning for the development of this breed.

## Flock Structure

Majority of the birds are kept as small (<10) flocks (63%). Medium flocks (11-20) are 29% where as large (>20) flocks are 8% only. Aseel rearing is not developed as organized farming yet. Each household keeps few birds with them.

## Housing

Birds are kept in kutcha and thatched houses only during night. In day time birds are kept in free ranging system.

## Management and Feeding

It can thrive well under free ranging condition with little supplementation of kitchen waste and grains like paddy, kanki, jowar, finger millet etc. Adequate water is provided to the birds.



*Free ranging system*

### **Breed Characteristics**

Aseel is well known for its endurance, power and fighting qualities. Large variations are found in morphological traits of this breed.



*White Aseel*



### **Plumage colour**

Most common colour is brownish-red followed by reddish-brown and black. However some white and golden colour plumage is also seen.



*Most common colour of plumage*

### **Pattern**

The patchy followed by solid and stripped pattern are generally observed. However, few dull and spotted birds are also found.

### **Skin colour**

Yellow skin is most common. White colour is also found in few.

### **Shank colour**

Yellow shank is prevalent.

### **Earlobe colour**

Earlobe is of red colour.

### Comb colour and type

Various type of comb like single, pea and rose have been observed. Pea comb is commonly found, whereas, single and rose comb are present in few. In some exceptional cases V-shaped comb is observed in adult males and yellow colour comb in pullets.

### Eye colour

Brown eyes are prevalent. However, black colour is also observed in both the sexes in field.

### Performance

#### Body weight

Birds attain half kg body weight during 14<sup>th</sup> week. One Kg between 20-24 weeks, one and half Kg in 32 weeks and by the time they complete one year of age, weight reach around 2.5 Kg.

Age (week)	Mean Body Weight (g)
0	29.0 ± 0.05
8	234.0 ± 0.14
20	934.0 ± 0.61
40	1964.0 ± 12.25

#### Feed efficiency

Feed efficiency (feed consumed divided by gain in body weight) during different weeks range from 2.45-5.29 upto 10<sup>th</sup> week and feed efficiency decreases after 10<sup>th</sup> week. Feed efficiency ranges from 4.08 - 8.57 after 10<sup>th</sup> week of age.

### **Age at first egg**

Aseel bird lay first egg at 27-29 weeks of age. In field condition, a hen lays for a period of 15 to 20 days. Then it incubates the eggs for 21-22 days. After chicks come out of eggs it broods the chicks for 8-12 weeks before laying again.

### **Egg production**

In 3 laying cycle it lays around 30-36 eggs (10-12 egg per laying cycle) per year. Eggshell colour is predominantly brown.

### **Meat production**

There is no fixed age for slaughter of Aseel birds in field. The tribals generally slaughter the birds after 28-32 weeks that too during some festival, religious event or marriages only. Dressing percentage is quite high (75%) when the birds are slaughtered during 27-50 weeks of age.

### **Fertility & hatchability**

Aseel is highly fertile (84.28%). Hatchability on total egg set basis is 74% and fertile egg set basis is 85%.

### **Broodiness**

This bird has natural instinct of broodiness which makes it natural incubator and hatcher, a desirable trait for the villagers. Birds go for brooding for a long period and usually thrice a year and sometimes twice a year.

### **Socio-cultural Aspect**

Aseel birds are integral part of the cultural heritage of tribal community in Bastar and Andhra Pradesh border areas.





*Ready to fight ...*

This Socio-cultural integrity is the savior for this breed till now. Traditional bird fighting is one of the main attractions of festivals and fairs.



*In the mid of fighting*

## **Disease and Parasites**

Ranikhet, fowl pox, bacillary white diarrhea are common. Ascariasis is common parasitic disease, taeniasis is also seen. Among external parasites, lice and tick infestation is prevalent . Due to tribal area few farmers avail the

facility of vaccination; almost all the birds remain unvaccinated. Treatment facility is also not available in the area.

### **Mortality**

Average mortality is quite high (40%) up to one week of age. In adults it is upto 13%.

### **Molecular Characterization**

Information on genetic variation within the breed, and its structure may help to evaluate how likely various factors responsible for change in its foundation genetic structure are operating, and generated information can be used in the conservation and improvement of this unique breed of goat. Of the many genetic markers now available, microsatellite loci are best suited to analyze the degree and pattern of genetic variability within and differences between- populations because of their high variability, high mutation rate, large number, distribution through out the genome, codominant inheritance and neutrality with respect to selection (Boyce et al., 1996). Thus, during analysis most of the assumptions for population genetics theory are fulfilled.

The random blood samples of Aseel poultry were collected from Baster (Chhattisgarh) and Khammam (Andhra Pradesh) districts, which is the breeding tract for this breed. DNA was extracted from whole blood using standard protocol (Sambrook et al., 1989). The DNA isolation procedure encompassed lysis of RBC's, digestion of protein using Proteinase K and precipitation of protein using phenol: Chloroform: isoamylalchol. After diluting the DNA, quality of DNA was checked on 0.8% agarose gel. Twenty six microsatellite primers were taken for the study (Table 1).



**Table1: Primer sequence, chromosomal location, annealing temperature and MgCl<sub>2</sub> concentration**

S. No.	Primer	Primer Sequence	Chromosomal Location	Annealing Temp.(°C)	MgCl <sub>2</sub> Concentration
1.	ADL176	TTGTGGATTCTGGTGGTAGC TTCTCCCGTAACACTCGTCA	E6	50°	1.5
2.	ADL102	TTCCACCTTTCTTTTTTATT GCTCCACTCCCTTCTAACC	C30 E29	46°	1.5
3.	ADL136	TGTCAGCCCATCGTATCAC CCACCTCCTCCTGTTCA	E5 C10	52°	1.5
4.	ADL267	AAACCTCGATCAGGAAGCAT GTTATTCAAAGCCCCACCAC	C3 E6	50°	2.0
5.	MCW014	AAAATATTGGCTCTAGGAAGTGC ACCGGAAATGAAGGTAAGACTAGC	E11	52°	1.5
6.	ADL210	ACAGGAGGATAGTCACACAT GCCAAAAAGATGAATGAGTA	E30	45°	1.5
7.	MCW049	AGCGGCGTTGAGTGAGAGGAGCGAT CCCCAACCCGCGGAGCGCTAT	C6	55°	1.5
8.	MCW007	AGCAAAGAAGTGTCTCTGTTTCAT ACCCTGCAAAGTGAAGGGTCTCA	1	60°	0.75
9.	MCW005	ACCTCCTGCTGCAAATAAATTGC TCACTTTAGCTCCATCAGGATTCA	C11 E5	55°	1.5
10.	MCW041	CCCATGTGCTTGAATAACTTGGG CCAGATTCTCAATAACAATGGCAG	C3	55°	1.5
11.	ADL158	TGGCATGGTTGAGGAATACA TAGGTGCTGCACTGGAAATC	C30 E29	50°	1.5
12.	ADL020	GCACTCAAAGAAAACAAAT TAGATAAAAATCCTTCCCTT	1	55°	3.0
13.	MCW059	AAGTGCCTTTGCTATCCTGATTGG AACTCCTATTGTGCAGCAGCTTAT	C1 E2	55°	1.5
14.	ADL171	ACAGGATTCTTGAGATTTTT GGTCTTAGCAGTGTGTTGTTT	E43	45°	1.5
15.	HUJ008	CTTAATATGTGTGAGGTGGC GTTCTACAATTGCATTAGC	C21	60°	1.5
16.	HUJ012	GTCTCATGCTATGAGAGTGG CCTCTGGTTGAATCAGTCTG	8	60°	1.5
17.	HUJ002	CATCTCACAGAGCAGCAGTG GAATCCTGGATGTCAAAGCC	17	60°	1.5
18.	HUJ006	GGAACATGTAGACAAAAGCA AGCAGTCCATTTACAGCCA	3	60°	1.5
19.	HUJ007	CATAAACTAAAGTCTCAACAC TTCTTCCACCACTTTGCTA	5	60°	1.5
20.	MCW016	ATGGCGCAGAAGGCAAAGCGATAT TGGCTTCTGAAGCAGTTGCTATGG	E2	50°	0.75
21.	ADL040	TTTCCCAGATTTACAACCTT GCCAGTGATACTCCAGCAGC	6	49°	1.5
22.	ADL034	AACCTAAAACTCCTGCTGC GGGAACCTGTGGGCTGAAAG	E47 W24	50°	1.5
23.	ADL023	CTTCTATCCTGGGCTTCTGA CCTGGCTGTGTATGTGTTGC	5	60°	1.5
24.	ADL039	GCTACAACGCTTCAAACCTG ACAAACAAACCAAAAAACCT	15	55°	0.75
25.	ADL044	AAGTGGTTTATTGAAGTAGA CTGTGGTGTGCGTTAGTTG	12	60°	0.75
26.	MCW002	TCCAGAGACAGTTGCTCCACATTC GCAAGTTAGTTATTGTAGGGGCTC	C4	50°	1.5



Most of the microsatellite loci used in the study are dinucleotide repeats, and they undergo mutation at a higher rate by slip strand mechanism. Out of the loci selected at least 11 loci (ADL 176, ADL 267, MCW14, ADL 210, ADL 158, HUI 8, HUI 12, HUI 2, HUI 6, MCW 16, MCW 2) were di-nucleotide repeats, and 2 loci (MCW 41, MCW 59) were mononucleotide repeats, (T)<sub>n</sub>, and MCW 49 is tri-nucleotide repeat (TCA). It is evident from the chromosomal location that the loci selected were evenly distributed through out the genome of poultry (Table 1). The primer ADL 102, ADL 136, ADL 158, ADL 171, ADL 176, ADL 210, ADL 267, MCW 14, MCW 41, MCW 59, MCW 16 and MCW 2 were from the recommended list of primers by FAO for Domestic Animal Diversity analysis (Barker et al., 1998). Some of the recommended primers did not give distinct PCR amplifications and were excluded from the study. The primer pairs MCW 7, MCW 5, MCW 49, ADL 20, ADL 23, ADL 34, ADL 39, ADL 40, ADL 44, HUI 2, HUI 6, HUI 7, and HUI 8, HUI 12 were taken from the published literature. The criterion for selection of the appropriate microsatellites loci was the same as taken by FAO viz; 1) the microsatellite loci are in the public domain, 2) the microsatellite loci have been mapped and are not in linkage disequilibrium, 3) the microsatellite loci have shown inheritance in Mendelian fashion, 4) there are at least 4 alleles reported in different populations or reference populations.

The PCR conditions were standardized for all of the 26 primer pairs selected for the study. The variables, which required standardization, included annealing temperature, MgCl<sub>2</sub> concentration, quantity of primer, Taq polymerase and dNTP's.

The PCR products were loaded on 6% denatured PAGE (Polyacrylamide gel electrophoresis). The standard DNA markers were

simultaneous run in the gel for sizing of the alleles. The molecular weights of the alleles were estimated by making a standard curve taking  $\log_{10}$  of the molecular weight of Standard marker on X-axis and mobility of the DNA on Y-axis. The sizes of the alleles were extrapolated from the standard curve.

The statistical analysis was carried out using POPGENE software (Yeh et al., 1999). The heterozygosity measures were calculated using the following formulae given by Nei (1978).

1. The heterozygosity was calculated as:-

$$1 - \sum_{i=1}^{k-1} X_i^2$$

2. The unbiased heterozygosity

$$H = \frac{2n}{2n-1} \left[ 1 - \sum_{i=1}^{k-1} X_i^2 \right]$$

$k$  = no. of alleles  
 $X_i$  = frequency of  $i^{\text{th}}$  allele  
 $X_j$  = frequency of  $j^{\text{th}}$  allele

The PIC values (Polymorphic Information Content) was calculated using the formula given by (Botstien et al., 1980)

$$\text{PIC} = 1 - \left( \sum_{i=1}^{k-1} X_i^2 \right) - \sum_{i=1}^{k-1} \sum_{j=i+1}^k 2X_i X_j$$

No of alleles revealed by different microsatellite loci along with PIC values are presented in Table 2. All the loci selected were polymorphic in the populations selected. HUI8, ADL40 and MCW 2 were least polymorphic with only 2 alleles; ADL 136 was highly polymorphic and revealed 9 alleles.

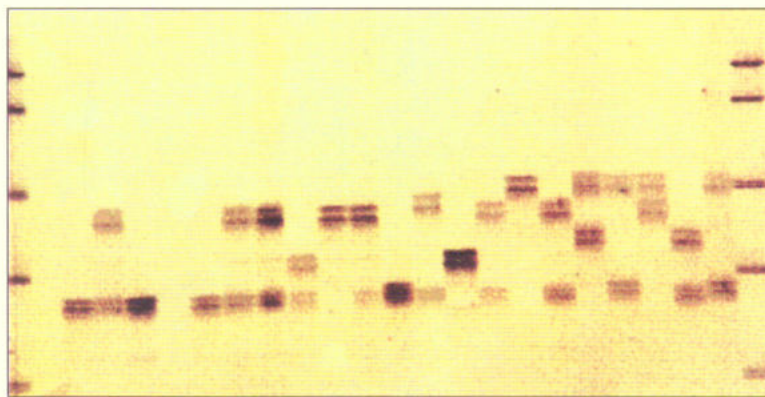
**Table 2: Estimates of genetic diversity in the Aseel poultry**

Locus	Observed no. of Alleles	Effective no. of Alleles	PIC	Observed Heterozygosity	Expected Heterozygosity
ADL176	8	4.72	0.7881	0.8529	0.7998
ADL102	3	2.61	0.6176	0.9714	0.6265
ADL136	9	6.22	0.8392	0.4286	0.8513
ADL267	3	1.39	0.5212	0.2571	0.5288
MCW14	4	2.01	0.5033	0.3714	0.5106
ADL210	5	2.44	0.5890	0.6000	0.5975
MCW49	4	1.96	0.4918	0.3714	0.4990
MCW7	3	2.80	0.6433	0.7429	0.6526
MCW5	5	3.74	0.7327	0.7714	0.7433
MCW41	4	3.03	0.6698	0.6000	0.6795
ADL158	4	2.47	0.5955	0.7429	0.6041
ADL20	4	2.34	0.5987	0.4483	0.6092
MCW59	4	3.37	0.7034	0.6970	0.7142
ADL171	6	3.36	0.7065	0.6000	0.7168
HUJ8	2	1.12	0.0820	0.0857	0.0832
HUJ12	4	2.85	0.6498	0.6571	0.6592
HUJ2	4	3.34	0.7011	0.7335	0.7116
HUJ6	4	2.78	0.6345	0.6765	0.6440
HUJ7	3	2.50	0.6012	0.5143	0.6099
MCW16	4	3.72	0.7318	0.7143	0.7424
ADL40	2	1.92	0.4800	0.4000	0.4870
ADL34	5	2.76	0.6246	0.4412	0.6339
ADL23	5	3.13	0.6805	0.4839	0.6917
ADL39	4	2.17	0.6122	0.4571	0.6211
ADL44	4	3.47	0.7114	0.4857	0.7217
MCW2	2	1.74	0.2130	0.1212	0.2163
Mean	4.2	2.85	0.6047	0.5472	0.6137

(Pandey et al., 2003)



Allele ranges from 2 to 9 with the mean of 4.2. All the loci except two (HUI 8 & MCW 2) were found to be highly polymorphic. PIC ranged from 0.0820 (HUI 8) to 0.8392 (ADL 136). The average PIC was 0.6047 indicating markers suitability for genetic diversity analysis. The observed heterozygosity ranged from 0.0857 (HUI 8) to 0.9714 (ADL 102) with the mean of 0.5472. Expected heterozygosity ranged from 0.0832 (HUI 8) to 0.7998 (ADL 176) with the mean of 0.6137. Population revealed higher values for observed and expected heterozygosity. The heterozygosity values are higher than observed in commercial breeds/strains of poultry.



*Gel showing different alleles at a locus*

Heterozygosity estimates within the population were based on a set of microsatellite loci showing substantial heterogeneity in the number of alleles and PIC values. The use of a mixture of highly variable and less variable microsatellites reduces the risk of overestimating genetic variability which might occur if only highly variable loci are used.

These selected loci are highly polymorphic and suitable for genetic diversity analysis. The PIC which gives the informativeness of the loci is also high and indicates their suitability for this kind of analysis. There are sufficient numbers of loci which can be used for diversity analysis as these cover almost the complete genome of the poultry. Since there are large

number of alleles in heterozygous condition there is little scope of losing alleles due to fixation. The population is not endangered and to maintain the present level of genetic diversity only *In Situ* conservation and improvement measures may be sufficient. This is more so because these birds are intricately associated with the cultural aspects of their respective regions.

### Future Strategies

Farmers should be educated and helped in :

1. Vaccination
2. Endo and ecto parasitic control
3. Organization of Aseel Exhibition /mela in the area and best birds of different category should be awarded and farmers should be encouraged
4. Disease diagnostic facilities should be created at Tehsil /block Level

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