



Allozyme variation in house fly populations, *Musca domestica* from Allahabad, India

Tripathi M.^{1*}, Agrawal U.R.² and Tripathi J.¹

¹Department of Zoology, Iswar Saran Degree College, University of Allahabad, Allahabad-211004, INDIA

²Department of Zoology, CMP Degree College, University of Allahabad, Allahabad-211002, INDIA

Available online at: www.isca.in, www.isca.me

Received 5th July 2013, revised 16th August 2013, accepted 17th September 2013

Abstract

Allozyme variation was assessed in the four populations of the common house fly *Musca domestica*. Allozymes at three gene enzyme system unraveled four loci which revealed nine alleles. F statistics revealed that except XDH all the other loci show inbreeding ($F_{is} > F_{st}$). Very little genetic differences have been found among the populations of *M. domestica*.

Keywords: *Musca domestica*, allozyme variation, inbreeding, genetic identity, genetic distance.

Introduction

Enzyme polymorphism in natural population are useful tool for estimating genetic variation¹. The level of genetic variation within populations of a species indicate species vitality and potential for evolutionary responses to environmental changes².

Musca domestica Linnaeus, common house fly with cosmopolitan distribution is of great medical and sanitary importance as these are vectors of many diseases causing organism³⁻¹¹.

Genetic variations among house flies population have been analyzed using allozyme in different parts of the world¹²⁻¹⁹. However such studies have been sporadically carried out in Indian subcontinent²⁰⁻²². In the present study allozyme variation in house fly, *M. domestica* from four different localities of Allahabad (U.P.) India has been analyzed to determine population diversity.

Material and Methods

The house flies *Musca domestica* L. were collected using sweep nets from four different locations with qualitative difference in the food resources i.e., Meat shop (MS), Vegetable market (VM), Dairy farm (DF) and Solid food waste (SW).

Fifty individuals were assayed for enzyme activity at three gene enzyme systems viz., malic enzyme (ME), aldehyde oxidase

(AO) and xanthine dehydrogenase (XDH). Sample preparation and electrophoretic procedures were according to the method of Tripathi et al 2010²⁰. The enzyme systems studied, staining solutions, gel and electrode buffer are given in table-1. Genotype information, genetic identity and genetic distance were calculated as described earlier²¹.

Results and Discussion

The three gene enzyme systems analyzed in present study viz., malic enzyme (ME), aldehyde oxidase (AO) and xanthine dehydrogenase (XDH) resolved four loci among the four populations. The activity of ME and XDH were confined to single locus figure-1 and 3 while the activity of AO was confined to two loci figure-2. All the four loci analyzed were polymorphic, which revealed nine alleles. Allele frequencies and Chi-square values are presented in table-2. Three loci viz., ME, AO-1 and AO-2 showed significant departure from Hardy-Weinberg equilibrium.

Flies from meat shop (MS) and dairy farm (DF) shared all the alleles while flies from vegetable market (VM) shared alleles at two locus i.e., ME and XDH with these two populations. The flies from solid food waste (SW) revealed fixed differences at ME and XDH, where it is monomorphic, however these flies shared VM at locus AO-1 and MS and DF at locus AO-2 table-2.

Table-1
Summary of electrophoresis and staining protocols followed in the present study

Enzyme	Gel/ electrode Buffer	Staining buffer	Substrate/Coenzyme	Dyes	Reference
ME (E.C.1.1.1.40)	0.1M Tris-HCl (pH 8.5)	0.1M Tris-HCl (pH 7.4)	Malic acid / NADP	NBT PMS	Tsukamoto (1989) ²³
AO (E.C.1.2.3.1)	0.1M Tris-HCl (pH 8.5)	0.1M Tris-HCl (pH 7.4)	Benzaldehyde	NBT PMS	Tsukamoto (1989) ²³
XDH (E.C.1.2.1.37)	0.1M Tris-HCl (pH 8.5)	0.1M Tris-HCl (pH 7.4)	Hypoxanthine / NADP	NBT PMS	Tsukamoto (1989) ²³

Table-2
Allele frequencies and Chi-square values in different collections of *M. domestica*

Locus	Allele	MS	VM	DF	SW
ME (n=50)	a	0.60	0.43	0.39	1.00
	b	0.40	0.57	0.61	-
	χ^2	8.17*	6.72*	3.26	-
AO-1 (n=50)	a	-	0.60	-	0.55
	b	-	0.40	-	0.45
	χ^2	-	8.17*	-	7.76*
AO-2 (n=50)	a	0.44	-	0.35	0.48
	b	0.56	-	0.65	0.52
	χ^2	7.45*	-	0.79	6.40*
XDH (n=50)	a	0.20	0.40	0.29	1.00
	b	0.42	0.60	0.45	-
	c	0.38	-	0.26	-
	χ^2	0.62	1.61	2.62	-

MS= Meat shop, VM = Vegetable market, DF= Dairy farm, SW= Solid food waste (in all the tables); n= number of individuals in each sample; *=Populations not in Hardy Weinberg equilibrium

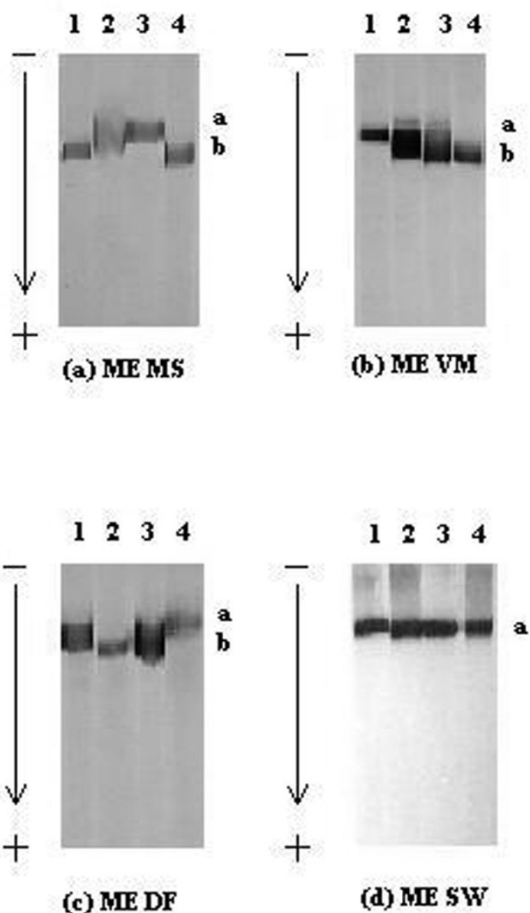


Figure-1

Electrophoretic phenotypes of Malic Enzyme (a) ME MS (b) ME VM (c) ME DF (d) ME SW in *Musca domestica*. The regions of electromorphs are indicated on the right

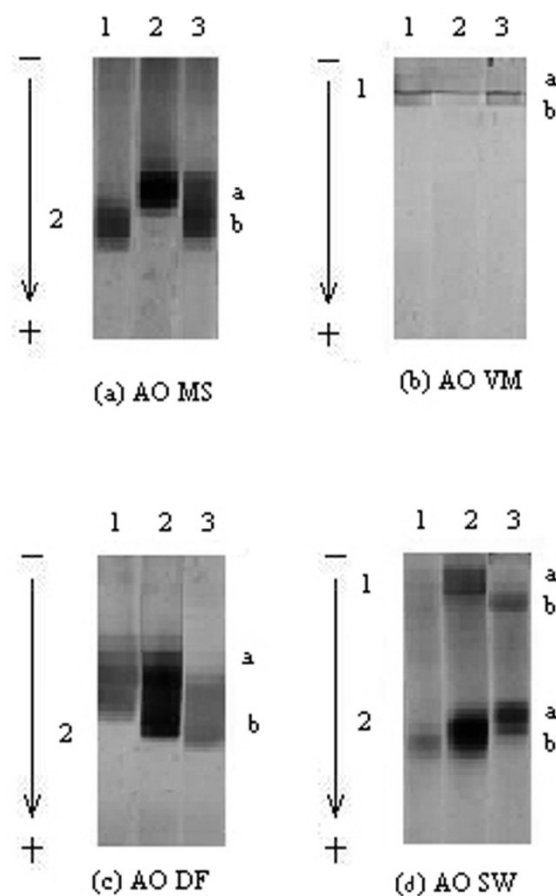


Figure-2

Electrophoretic phenotypes of Aldehyde oxidase (a) AO MS (b) AO VM (c) AO DF (d) AO SW in *Musca domestica*. The regions of activity are indicated on the left and electromorphs are indicated on the right

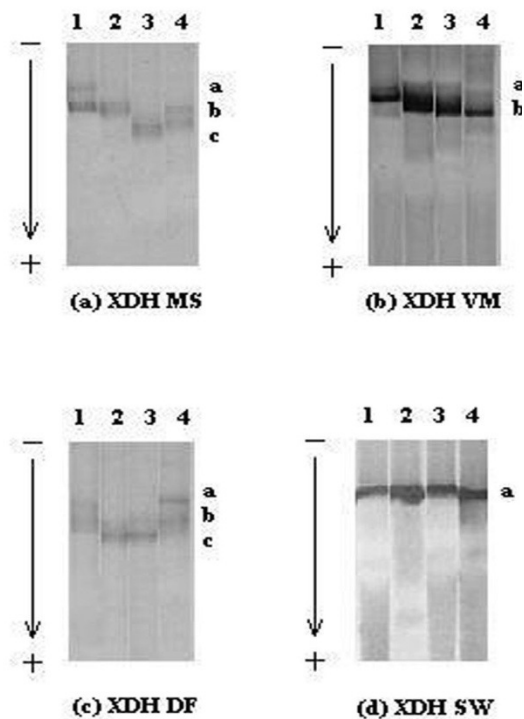


Figure-3

Electrophoretic phenotypes of Xanthine dehydrogenase (a) XDH MS (b) XDH VM (c) XDH DF (d) XDH SW in *Musca domestica*. The regions of electromorphs are indicated on the right

Number of alleles ranged from 1.85 to 2.07 with a mean of 1.97. The percentage of polymorphic loci ranged from 50.00% to 75.00% with a mean of 68.75% and the mean observed heterozygosity ranged from 0.250 to 0.447, with a mean of

0.356. The mean expected heterozygosity ranged from 0.483 to 0.534, with a mean of 0.510 table-3. Nei's genetic identity values were highest between the samples collected from MS and DF (I=0.955) and the Nei's genetic distance values were lowest between these two populations (D=0.046) table-4. As revealed by the Wright's F statistics very little genetic variation seems to have occurred among house fly populations analyzed in the present study. Except XDH all the other loci reveal inbreeding table-5.

Conclusion

It seems that the flies collected from MS and DF are genetically very similar. This may be due to the fact that the larval food substrates from the two collection sites were more or less similar as suggested by Thomas and Barker²⁴. As well as the house fly population analyzed in the present study are characterized by a high level of inbreeding. Thus the present findings support the tenet that very little genetic differentiation has accompanied the population differentiation among house flies in Allahabad region of India²².

Acknowledgement

We are grateful to Professor Raghav Ram Tewari, Cytogenetics Research laboratory, Department of Zoology, University of Allahabad for their useful suggestions, encouragement and providing laboratory facilities throughout the course of present work. Thanks are due to, Head, Department of Zoology (UGC-SAP and DST-FIST scheme sponsored), University of Allahabad, for providing all the necessary facilities.

Table-3
Genetic variability in different collections of house flies populations

Population	Sample Size	Number of loci	Mean Effective no. of alleles	Percentage of polymorphic loci	Mean observed heterozygosity (H _o)	Mean expected heterozygosity (H _E)
MS	50	4	1.85	75.00%	0.447	0.534
VM	50	4	2.07	75.00%	0.287	0.483
DF	50	4	1.95	75.00%	0.440	0.526
SW	50	4	2.01	50.00%	0.250	0.497
Mean	50	4	1.97	68.75%	0.356	0.510

H_o = No. of heterozygotes / Total no. of individuals, H_E = 1 - Σx_i² (Nei, 1972), where x_i is the frequency of ith allele at a locus

Table-4
Genetic identity (I) and genetic distance (D) among house flies in different collections

(I)					
(D)	Population	MS	VM	DF	SW
	MS	-	0.558	0.955	0.636
	VM	0.584	-	0.607	0.621
	DF	0.046	0.499	-	0.572
	SW	0.452	0.477	0.559	-

I = J_{xy} / √J_x J_y, D = -ln I, Where J_{x y} is the arithmetic mean of J_{x y} = Σ x_i y_i over all loci, J_x is the arithmetic mean of i_x = Σ i_x² over all loci, and x_i (or y_i) is the frequency of the ith allele in the first (or second) population.

Table-5
Wright's F statistics for all the variable loci.

Loci	F _{is}	F _{st}
ME	0.474	0.243
AO-1	0.461	0.001
AO-2	0.267	0.010
XDH	0.055	0.284
Mean	0.314	0.135

References

- Murphy R.W., Sites J.W., Buth D.G. and Haufler C.H., Proteins: isozyme electrophoresis, In: Molecular systematic (eds. Hills D.M., Moritz C. and Mable B.K.), Sinauer Associates. Inc. Sunderland, Massachusetts, U.S.A., 655 (1996)
- Mateus R.P. and Sene F.M., Temporal and spatial allozyme variation in the South American cactophilic *Drosophila antonietae* (Diptera : Drosophilidae), *Biochem. Genet.*, **41**, 219- 233 (2003)
- West L., The housefly. Its natural history, medical importance and control, Ithaca NY: Comstock, (1951)
- Scott H.G. and Lettig K.S., Flies of Public Health Importance and their Control, Washington: U.S. Government Printing Office, (1962)
- Greenberg J.B., Flies and disease, Vol.1, Ecology, classification and biotic association, Princeton, NJ: Princeton University Press, (1973)
- Keiding J., The house fly: biology and control, WHO Vector Control Series: 63, (1986)
- Tan S.W., Yap K.L. and Lee H.I., Mechanical transport of rotavirus by the legs and wings of *Musca domestica.*, *J Med Entomol.*, **34**, 527-531 (1997)
- Grubel P., Huang L., Masubuchi N., Stutzenberger F.J. and Cave D.R., Detection of *Helicobacter pylori* DNA in houseflies (*Musca domestica*) on three continents, *Lancet.*, **352**, 788-792 (1998)
- Sasaki T., Kobayashi M. and Agui N., Epidemiological potential of excretion and regurgitation by *Musca domestica* (Diptera: muscidae) in the dissemination of *Escherichia coli* O157: H7 to food, *J Med Entomol.*, **37**, 945-949 (2000)
- Zurek L., Denning S.S., Schal C. and Watson D.W., Vector competence of *Musca domestica* (Diptera: Muscidae) for *Yersinia pseudotuberculosis*, *J Med Entomol.*, **38**, 333-336 (2001)
- Maisnier-Patin S. and Andersson D.I., Adaptation to the deleterious effects of antimicrobial drug resistance mutations by compensatory evolution, *Res Microbiol.*, **155**, 360-369 (2004)
- Stanger J., Preliminary observations on genetic variation in three colonies of *Musca domestica* (Diptera: Muscidae) isolated from central Alberta, *Quaest Entomol.*, **20**, 51-59 (1984)
- Black IV W.C. and Krafur E.S., Electrophoretic analysis of genetic variability in the house fly (*Musca domestica* L.) *Biochem Genet.* **23(3-4)**, 193-203 (1985)
- Black IV W.C. and Krafur E.S., Temporal and spatial trends in allozyme frequencies in house fly populations, *Musca domestica* L. *Theor. Appl. Genet.*, **71**, 673-681 (1986 a)
- Black IV W.C. and Krafur E.S., Seasonal breeding structure in house fly, *Musca domestica* L., Populations, *Heredity*, **56**, 289-298 (1986 b)
- Krafur E.S., Helm J.M. and Black IV W. C., Genetic diversity at electrophoretic loci in the house fly, *Musca domestica* L. *Biochem Genet.*, **30** , 317-328 (1992)
- Krafur E.S., Bryant N.L., Marquez J.G. and Griffith N.T., Genetic distances among North American, British and West African house fly populations *Musca domestica* L. *Biochem Genet.*, **38**, 75-284 (2000)
- Taskin B.G., Taskin V. and Kucukakyuz K., Electrophoretic analysis of genetic diversity in natural house fly (*Musca domestica* L.) populations from the Western and Southern coasts of Turkey, *Tubitak, Turk. J. Biol.* **35**, 337-346 (2011a)
- Taskin B.G., Taskin V. and Kucukakyuz K. and Kence M., Determination of esterase enzyme polymorphism in house fly (*Musca domestica* L.) populations in Turkey, *Tubitak, Turk. J. Zool.* **35(6)**, 896-877 (2011 b)
- Tripathi M., Tewari R.R. and Agrawal U.R., Genetic variations in house fly, *Musca domestica* L (Diptera :Mucidae) from Allahabad India . *Proc. Nat. Acad. Sciences, India (Biological Sciences)* **80 (1)**, 24-29 (2010)
- Tripathi M., Agrawal U.R. and Tewari R.R., Seasonal genetic variation in house fly populations, *Musca domestica* (Diptera: Mucidae), *Cell. Mol. Biol.*, **57**, 129-134 (2011)
- Tripathi M., Agrawal U.R., Tripathi J and Tewari R.R., Spatial genetic variation in house fly populations, *Musca domestica* (Diptera :Mucidae), *Int J Pharm Bio Sci.*, **3(4)**, 927 – 934 (2012)
- Tsukamoto M., Enhancement of Staining intensity of mosquito larva zymograms after electrophoresis, *J. UOEH.*, **11**, 461-479 (1989)
- Thomas R. and Barker J.S.F., Breeding structure of natural populations of *Drosophila buzzatii*: Effects of the distribution of larval substrates, *Heredity*, **64**, 355-361 (1990)
- Genetic variations among Ecologically diverse species of Anurans at the level of Genus based on ISSR Marker Santhosh Kumar K., Lingaiah Kusuma, Ramachandra N.B. and Nair Vijay Mala, *I. Res. J. Biological Sci.*, **1(7)**, 11-19(2012)
- Sodium Dodecyl sulphate Polyacrylamide gel Electrophoresis Pattern of Horse Gram Seed Storage Proteins during Germination Pek Geok Pang, Asrul Afandi, Rahman Shefiqur and Shaha Ranajit kumar, *I. Res. J. Biological Sci.*, **1(4)**, 39-50(2012)