Table 3. Effects of CGA-112913 (IKI-7899 or UC-62644) on the reversion rate of several histidine auxotrophs of *Salmonella typhimurium*

Concentration	Average number of revertants per plate either with or without the S-9 metabolic activation system in the following strains									
	TA98		TA100		TA1535		TA1537		TA1538	
(µg/plate)	(−) S-9	(+) S-9	(−) S-9	(+) S-9	(−) S-9	(+) S-9	(−) S-9	(+) S-9	(−) S-9	(+) S-9
0.9	56	24	143	110	4	5	4	14	20	10
2.0	70	30	132	132	5	12	5	14	17	10
20.0	44	24	115	127	7	6	3	13	16	12
200.0	43	23	122	119	6	5	3	9	16	15
500.0	29	26	117	111	4	5	4	9	21	10
− control	45	23	118	114	7	4	4	10	24	14
+ control	254	3318	303	268	40	23	66	46	217	97

flected in the rest of the data and can be attributed to experimental variation.

CGA-112913 has been tested in the laboratory, greenhouse and field against the spruce budworm, *Choristoneura fumiferana*, the major pest of Canadian forests, and found to be very effective at extremely low dosages[10,11]. Its acute toxicity in mammals is very low with an LD_{50} of > 4000 mg/kg[12]. Our tests indicate that this material is non-mutagenic in the Ames system and therefore warrants further tests including long-term chronic toxicology studies.

1 The authors thank Professor Bruce N. Ames for supplying the tester strains together with all the relevant information for conducting the bioassays and Ms. Norma Charlebois for her meticulous technical assistance.

2 Maas, W., van Hes, R., Grosscurt, A.C., and Deul, D.H., in: Chemie der Pflanzenschutz- und Schädlingsbekämpfungsmittel, vol. 6, p. 423. Ed. R. Wegler. Springer-Verlag, Heidelberg 1981.
3 Retnakaran, A., Granett, J., and Ennis, T., in: Comprehensive Insect Physiology, Biochemistry and Pharmacology, vol. 12, cap. 14. Eds G.A. Kerkut and L.I. Gilbert. Pergamon Press, Oxford 1984, in press.
4 Casida, J.E., in: Advances in Pesticide Science, part I, p. 45. Ed. H. Geissbühler. Pergamon Press, Oxford 1979.
5 Retnakaran, A., Bull. ent. soc. Am. *28* (1982) 146.
6 Ames, B.N., McCann, J., and Yamasaki, E., Mutation Res. *31* (1975) 347.
7 Maron, M.D., and Ames, B.N., Mutation Res. *113* (1983) 173.
8 Levin, D.E., Yamasaki, E., and Ames, B.N., Mutation Res. *94* (1982) 315.
9 McCann, J., Springarn, N.E., Kobori, J., and Ames, B.N., Proc. natn. Acad. Sci. USA *72* (1975) 979.
10 Retnakaran, A., Canadian Forestry Service Information Report, FPM-X-45 (1981).
11 Retnakaran, A., Can. Ent. *114* (1982) 523.
12 Ciba-Geigy Technical Data Sheet on CGA-112913 (1982).

0014-4754/85/111464-02$1.50 + 0.20/0

Genetic studies on gynandromorphism *(sgm, gm)* in *Culex pipiens fatigans*[1]

W. Ahmad, A. Ara and U.M. Adhami

Section of Genetics and Cytogenetics, Department of Zoology, Aligarh Muslim University, Aligarh-202001 (India), 2 March 1983

Summary. Individuals showing anterior-posterior gynandromorphism or somatic mosaicism of palpi respectively have been found in a laboratory strain of *Culex pipiens fatigans*. The origin of the gynanders on the basis of binucleated eggs bearing an *m* factor and independently fertilized by male gametes of *M* and *m* genotype respectively has been suggested.
Key words. Mosquito; *Culex pipiens fatigans;* gynandromorphism.

Gynandromorphism and intersexuality are frequent in mosquitoes and have been reported in *Culex pipiens fatigans*[2,3], *Culex pipiens*[4,5], *Aedes aegypti*[6,7], *Aedes albopictus*[8] and in *Aedes nigripes*[9]. Laven[4] described an autosomal recessive intersex gene in *Culex pipiens* which feminizes genetic males and McCleland[6] showed that intersexuality is under genetic control in *Aedes aegypti* too. He also isolated a strain where males show sex conversion under certain temperature condition but appear normal when reared at lower temperatures. Craig[10] isolated a different temperature sensitive intersex producing strain. Amir Skanian[11] was successful in inducing gynandromorphism and sex mosaicism by the mutagenic agent thalimide in *Culex pipiens molestus*.

The present study reports on two kinds of gynandromorphism from an inbreeding laboratory strain of *Culex pipiens fatigans*. Attempts have been made to investigate the possible genetic basis and the probable mechanism of origin of these abnormalities.

Materials and method. In a laboratory strain of *Culex pipiens fatigans* which was being searched for genetic variations, two kinds of gynandromorphs were discovered. One type was partly female and partly male, the anterior part being that of a male while the posterior was typical of a female. On the anterior part, i.e., on the head, feathery antennae, long palpi and a sucking type proboscis, all characteristics of a male, were present. The posterior part showed a broad abdomen with membranous pleurae and long wings, characteristics of female last abdominal segment or slightly further, and the last abdominal segment had female genitalia. This appearance is designated as simple gynandromorph *(sgm)* (fig. 1). The second type of gynander was similar to *sgm* in all respects except palpi, where it showed somatic mosaicism. The palp of one side was always longer, extending beyond the proboscis, whereas the palp of the other side was very short (fig. 2). This type was called gynandromosaic *(gm)*.

In spite of repeated efforts these gynanders did not take a blood meal. With mouth parts being typically those of a male (the proboscis being the sucking type) this was to be expected. Techniques and details of rearing, breeding experiments and crossings were similar to those described earlier[12]. All experi-

Experientia 41 (1985), Birkhäuser Verlag, CH-4010 Basel/Switzerland

Table 1. Results of crosses made to recover gynandromorphs (*sgm*) in *Culex pipiens fatigans*

S. No.	Types of cross		Progeny* Total	Female	Male	Gynanders
1)	Gynander	× normal ♂	–	–	–	–
2)	Normal ♀ (gynander sib)	× normal ♂ (gynander sib)	454	228	210	16
3)	Normal ♀ (gynander sib)	× normal ♂ (out crossed)	382	197	185	–
4)	Normal ♀ (out crossed)	× normal ♂ (gynander sib)	319	168	151	–

* Data based on five generations of single-pair matings for each type of cross and then pooled together.

Table 2. Results of crosses made to recover gynandromosaics (*gm*) in *Culex pipiens fatigans*

S. No.	Type of cross		Progeny* Total	Female	Male	Gynanders
1)	Gynander	× normal ♂	–	–	–	–
2)	Normal ♀ (gynander sib)	× normal ♂ (gynander sib)	347	158	180	9
3)	Normal ♀ (gynander sib)	× normal ♂ (out crossed)	482	258	224	–
4)	Normal ♀ (out crossed)	× normal ♂ (gynander sib)	419	218	201	–

* Data based on five generations of single-pair matings for each type of cross and then pooled together.

ments were conducted in an environmental control room with constant conditions of 27 ± 1 °C and 75 ± 5% relative humidity. The control room was illuminated for 12 h.

Results. Crossing experiments for *sgm* and *gm* are summarized separately in tables 1 and 2. The crossing of the gynander with a normal male did not yield any progeny (tables 1 and 2, No. 1). Another experiment was made by just taking pairs of normal sibs of the gynanders. Out of a total of 454 or 347 individuals 16 *sgm* or 9 *gm* respectively were recovered (tables 1 and 2, No. 2). Females and males from the line yielding the gynanders were then outcrossed with individuals of the opposite sex from non-gynander lines. None of these crosses produced any gynanders in the progeny (tables 1 and 2, Nos 3 and 4).

Discussion. It may reasonably be assumed that the gynander was produced by inbreeding. The frequency of *sgm* and *gm* is 3.5% and 2.6% respectively and well below the corresponding value of 5–8% in the 'Mainz strain'[5]. The fact that gynanders appeared in successive generations indicates that some type of inheritance does exist. Since both kinds of gynanders are sterile and unable to reproduce, the factors responsible for this disturbance cannot be transmitted through the gynandromorphs themselves. It is quite probable that the factor responsible for it are transmitted through sexually normal individuals of the same stock. Most likely the trait is recessive. It therefore seems to be different from *gyn* which was suggested to be the expression of a dominant gene[5].

Had the sex in mosquitoes been dependent on an XX–XY mechanism as in *Drosophila*, the origin of the gynander could have been explained simply as a result of the loss of one X chromosome in the cells of the male parts. Since this is not so, one has to look for alternative mechanisms. Gilchrist and Haldane suggested the factors *M* and *m* for the determination of sex, the factor for maleness, *M* being dominant[13]. A male has the factors, *M*/*m*, the females are homozygous *m*/*m*. If one assumes the origin of the gynander on this basis, the genetic constitution of the male and female parts should be *M*/*m* and *m*/*m* respectively for *sgm*. This could be possible if the egg had two nuclei (both *m*) and each one of them was fertilized by a different sperm, one having *M* and the other *m*. The two parts of the individual would then develop differently, the portion having the *M*/*m* constitution would give rise to typical male characteristics while the portion with *m*/*m* genes would differentiate into typical female structures.

The origin of gynander showing mosaicism of palpi can be explained on the same lines. Denoting the gene for short palp by *Sp*, the genetic constitution of a *gm* phenotype may be as follows:

$$\text{♀ part } \frac{m}{m}\,\frac{+}{+} \text{ and ♂ part } \frac{M}{m}\,\frac{Sp}{+}$$

It comes perhaps from a mother *m*/*m*, +/+ and a father *M*/*m*, *Sp*/ + . The mother can produce only one kind of gamete, namely *m* + . The father produces four kinds of gamete: *M Sp*, *m Sp*, *M* + *and m* + . The offspring expected of such parents would be:

$$\frac{M}{m}\,\frac{Sp}{+} \text{ or } \frac{M}{m}\,\frac{+}{+} \text{ or } \frac{m}{m}\,\frac{Sp}{+} \text{ or } \frac{m}{m}\,\frac{+}{+}$$

Assuming the presence of two egg nuclei the female part (posterior one) must have originated from an egg nucleus *m* + and a sperm *m* + and the male part from an egg nucleus *m* + and a sperm *M Sp*, otherwise it could not be male and could not have a short palp $\frac{M}{m}\,\frac{Sp}{+}$, heterozygous .

Alternatively, a disturbance in the genic balance may be responsible for the production of gynanders. In that case some other mechanism like the sterility factor reported in *Culex pipiens* will have to be considered[14].

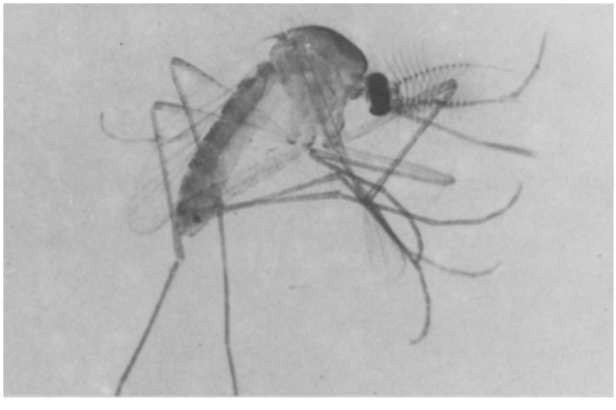

Figure 1. Showing anterior male and posterior female part *(sgm)*.

Figure 2. Showing gynandromorphism and somatic mosaicism of palpi *(gm)*.

1 This study was supported by the grant (No. 7/112/919/81 EMR-1), Council of Scientific and Industrial Research Govt., of India.
2 Shetty, N.J., and Chowdaiah, B.N., Curr. Sci. *44* (1975) 702.
3 Shetty, N.J., and Chowdaiah, B.N., Ind. J. Hered. *7* (1975) 1514.
4 Laven, H., Z. Naturforsch. *10b* (1955) 320.
5 Laven, H., Z. Vererbungsl. *88* (1957) 443.
6 McCleland, G.A.H., Bull. ent. Soc. Am. *9* (1963) 173.
7 VandeHey, R.C., and Craig, G.B. Jr, Bull. ent. Soc. Am. *7* (1961) 174.
8 Bat-Miriam, M., and Craig, G.B. Jr, Mosq. News *26* (1966) 13.
9 Brust, R.A., Can. J. Zool. *44* (1966) 911.
10 Craig, G.B. Jr, Proc. 12th Int. Cong. Ent. (1965) 263.
11 Amir Khanian, J.D., Experientia *26* (1970) 796.
12 Waseem, A., Genetic studies on morphological variability in *Culex pipiens fatigans*, p. 157. Ph. D. thesis, Aligarh Muslim University 1980.
13 Gilchrist, B.M., and Haldane, J.B.S., Hereditas *33* (1947) 175.
14 Laven, H., Z. Vererbungsl. *85* (1953) 118.

0014-4754/85/111465-03$1.50 + 0.20/0

Torus shaped bands at the 2R telomere region and at the region 68 of the salivary gland chromosomes of *Drosophila auraria*[1]

Z.G. Scouras and C.D. Kastritsis

Department of Biology, School of Sciences, Aristotelian University of Thessaloniki (Greece), 10 August 1984

Summary. The telomere of the 2R arm of the salivary gland chromosomes of *D. auraria* exhibits a definite toroidal structure in routine squashed preparations, stained either by propionic orcein-carmine or by fluorescent dyes. There is evidence that a band (or bands) of region 68 (possibly homologous to that of *D. melanogaster)* of the 3L chromosome arm also exhibits a toroidal structure. These toroids are associated with heterochromatin, but it is not certain that they are themselves heterochromatic.
Key words. Toroidal bands; *Drosophila*; polytene chromosomes.

Polytene chromosomes are an ideal system to study the interphase chromosomal structure. Since these chromosomes are the results of 10 rounds (2^{10}–1024 C) of DNA replication[2], they have a large size, visible under the light microscope, exhibiting a characteristic banding pattern, which represents a cytological map of the genome. Many genes can be placed on the map and a large number of these exhibit, during their activity, the phenomenon of puff formation. Looking at the literature concerning the structure of the bands, we can see that the main idea which has been proposed is that the band has a disk-like structure[3]. Recently, studies have been published proposing a toroidal structure for bands of these chromosomes in *Drosophila melanogaster*. These studies were performed by using thin sections examined under the light or the electron microscope after specific treatments[4,5].

In this report we show that certain bands of the polytene chromosomes of *D. auraria,* when observed under the light microscope in routinely fixed squashed preparations, exhibit torus-shaped configurations.

Materials and methods. A stock of *Drosophila auraria,* originally collected in Kirishima, Japan, and maintained in the Department of Zoology, University of Texas, Austin, as stock no. 3040.11b, and three sublines (nos. 17, 8 and 2) derived from the original stock by sibling matings[6] were used in this study.

The routine squashed preparation method was used for the observation of salivary gland polytene nuclei of animals at various developmental stages. Salivary glands were dissected in a Ringer-type solution[7], fixed in 3:1 ethanol:propionic acid, stained in propionic orcein-carmine and observed under a phase contrast microscope. Furthermore, the chromosomes were stained with fluorescent dyes (Quinacrine dihydrochloride[8] and Hoechst 33258[9]) and observed using a BG 12 excitation filter and a 53 barrier filter under a Zeiss fluorescence microscope.

For the detection of late replicating patterns of the genetic materials, a routine ^{3}H-thymidine (250 μCi/ml, s.a. 2 Ci/mM, The Radiochemical Centre Amersham) autoradiographic method[10] was used during several developmental stages.

Results and discussion. During the examination of the salivary gland polytene chromosomes of all strains of *D. auraria* used in this study, we observed that the bands which form the telomere of the 2R chromosome arm (two heavy plus a few fainter bands) exhibit a definite resemblance to a toroidal structure; this is the only region of the polytene complement which shows this structure more often than not, under the conditions described above. Figure 1 illustrates the torus shaped telomere of the 2R chromosome arm.

Beyond the 2R telomere, a band (or bands) of region 68 of the polytene chromosomes of the stocks examined also seems to show a toroidal structure (figs 2b, 2c). This region of the *D. aura-*

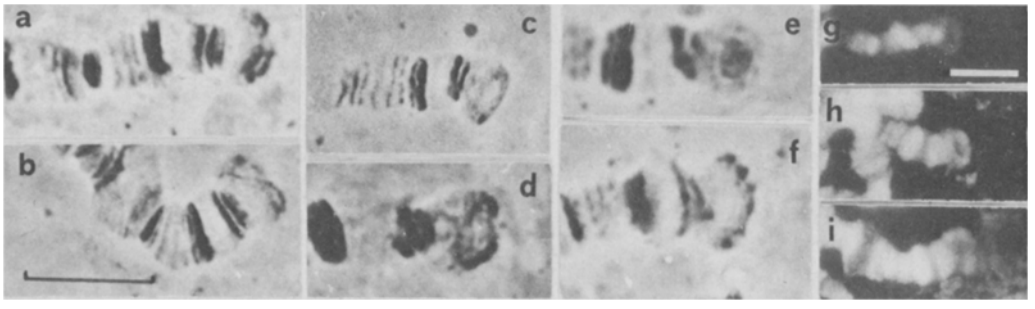

Figure 1. Toroidal structure of the 2R chromosome telomere of *D. auraria. a–f* Stained with propionic orcein-carmine; *g* stained with Quinocrine dihydrochloride; *h–i* stained with Hoechst 33258. *a* Asynapsed chromatids indicate toroidal structure; *b* figure eight type appearance of torus shaped bands; *c, d, g, h* toroidal structure of the telomere; *e* tight configuration of the telomere; *f, i* distorted configuration. Bars in all figures represent 10 μm.