MOHAMED A. H., SMITH J. D. and APPLEGATE H. G. (1966b) Cytological effects of hydrogen fluoride on tomato chromosomes. *Can. J. Genet. Cytol.* **8**, 575–583.
MUELLER H. J. (1961) Do air pollutants act as mutagens? In: Symposium on Emphysema and the Chronic Bronchitis Syndrome. (Aspen, Colorado, June 13–15, 1958). *Am. Rev. Resp. Dis.* **83**, 571–572.

1971

Atmospheric Environment Pergamon Press 1971. Vol. 5, pp. 117–122. Printed in Great Britain.

THE EFFECTS OF ATMOSPHERIC HYDROGEN FLUORIDE UPON *DROSOPHILA MELANOGASTER*—II

FECUNDITY, HATCHABILITY AND FERTILITY*

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Abstract—Two strains of *Drosophila melanogaster* were treated with sub-lethal levels of gaseous hydrogen fluoride for six weeks. Egg samples were collected at various times for hatchability determinations. Adults reared from these samples were evaluated for fecundity and fertility. Treatment with HF caused a marked reduction in hatchability and fecundity in the more sensitive strain. Male fertility was depressed but female fertility remained stable over the test period. The reduction of these parameters in the offspring of populations subjected to low levels of atmospheric HF contamination for prolonged periods suggests that HF causes genetic damage.

INTRODUCTION

THE DIFFERENTIAL responses of four genetic lines of *Drosophila melanogaster* to hydrogen fluoride contamination of the atmosphere were described in a previous paper (GERDES, *et al.*, 1971). Concentrations of 5.5 ppm HF were lethal for all populations tested, but different degrees of tolerance were observed at lower concentrations.

This study represents an attempt to evaluate changes which took place in biological parameters related to survival ability of *D. melanogaster* populations when these populations were exposed to different levels of hydrogen fluoride in the atmosphere. The parameters, fecundity, hatchability and fertility, relate directly to the reproductive capacity of a population. HF induced variations among these parameters are an indication of the accumulation of sub-vital genetic factors.

METHOD AND MATERIALS

The two strains of *Drosophila melanogaster* used in this work, Oregon-r and yellow white (*yw*), were propagated from stocks received from the Genetics Foundation, the University of Texas at Austin. These strains were chosen because of their disparate response to hydrogen fluoride (GERDES *et al.*, 1971).

The flies were treated and grown as described previously (GERDES and SMITH, 1967; GERDES *et al.*, 1971). Each population cage was permitted to equilibrate in the environmental chamber prior to administration of fluoride in the atmosphere. The cages were then individually subjected to average concentrations of 0, 1.3 and 2.9 ppm HF in the atmosphere.

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The use of exchangeable food vials in the outer wall of the styrofoam cages provided a continuous food supply, and, by changing them in sequence over a 14-day cycle, this permitted emergence of sufficient adults to maintain the populations at the desired levels. Additional food vials provided the necessary egg collection sites.

Egg collections were made at the initiation of each experiment and after 3 and 6 week exposure periods. These eggs were collected and hatched, and the adults were used in the determination of the various parameters.

The fecundity determinations were made using female progeny from the egg collections. Ten pairs of flies were mated with two replications for each treatment combination. Their eggs were collected and scored on the basis of the total number of eggs laid in a 24-hr period.

Hatchability was estimated as the percentage of eggs collected that yielded adults, which were classified according to sex. These adults were later used in the fertility determinations, as it was considered necessary that these tests be run on flies of the same age.

Fertility was determined by making 50 single pair matings among the adults from each hatchability test. If no larvae were observed by the eighth day, the male and female were separated and each was remated to a tester. The flies were scored individually as being sterile or fertile.

A split–split plot experimental design was used, with HF concentration being the whole plot treatment, strains representing sub-plots and treatment duration being the sub–sub plots.

RESULTS AND DISCUSSION

As a check on the homogeneity of the initial populations, replicated egg collections were taken from all cages after they had adjusted to the environmental chamber and before HF treatments were started. TABLE 1 gives the mean values of all the biological parameters in response to the HF treatments. TABLE 2 shows no significant differences among the cages which would receive different treatments, but a highly significant difference in fecundity existed between the two genetic strains. These analyses indicate homogeneity within the base populations from which each experimental sequence originated.

TABLE 1. TABLE OF MEAN VALUES OF BIOLOGICAL PARAMETER RESPONSE TO HF ATMOSPHERIC CONTAMINATION

Treatment duration	HF level (ppm)	Fecundity		Hatchability		Male fertility	
		Oregon-r	Yellow white	Oregon-r	Yellow white	Oregon-r	Yellow white
0	0	280	262	82	79.5	100	100
	1.3	280	271	79	79	100	99+
	2.9	293	268	78	75.5	99+	99+
3 weeks	0	286	261	79.5	76	100	99+
	1.3	242	241	71	70.5	98.5	97
	2.9	222	225	73	71	97	93
6 weeks	0	289	270	79	78	100	100
	1.3	196	212	74	71	99	96
	2.9	192	198	65.5	61	92	91

Fecundity: mean number of eggs from seven egg collections (days 2, 3, 6, 7, 10, 11, and 14).
Hatchability: mean percentage hatchability from four egg collections (days 2, 6, 10, and 14).
Male fertility: mean percentage male fertility.

TABLE 2. ANALYSES OF VARIANCE FOR HATCHABILITY, FECUNDITY AND FERTILITY FROM EGG COLLECTIONS TAKEN BEFORE TREATMENT INITIATION

Sources of variation	d.f.	Fecundity mean squares	Hatchability mean squares	Percentage fertility mean squares
Replication	1	11.69	0.36	20.80
Level	2	10.74	119.79	8.12
Error *a*	2	13.67	15.98	7.85
Strain	1	601.10*	80.62	2.79
Level × strain	2	34.18	37.97	2.02
Error *b*	3	27.02	26.66	10.99

*$P \leq 0.01$

The parameter determinations were made using adults hatched from eggs collected from the treated populations. This point is emphasized as it provides the basis for the genetic interpretations. Since the flies used to obtain the data were not exposed to HF, differences related to treatments are presumed to reflect degrees of genetic damage induced by treatment of their parents.

Hatchability data were obtained from four of the population egg collections, and the analyses of variance are presented in TABLE 3. Although these data were quite

TABLE 3. HATCHABILITY ANALYSES OF EGGS COLLECTED FROM *Drosophila* REARED FROM EGGS THAT WERE COLLECTED FROM THE HYDROGEN FLUORIDE TREATED POPULATIONS

Sources of variation	d.f.	Mean squares collection day*			
		2	6	10	14
Replication	1	35.20†	32.41	47.06	8.37
Level	2	214.17†	261.10	439.45	321.68‡
Error *a*	2	1.89	60.53	86.01	2.17
Strain	1	55.63	286.09	64.19	295.78†
Level–strain	2	67.85	98.96	72.67	166.12†
Error *b*	3	101.51	51.86	27.64	16.80
Duration	2	214.77‡	39.50	142.56†	398.92‡
Level–duration	4	89.81†	48.48	71.09	106.65†
Strain–duration	2	23.70	28.87	19.70	40.49
Level–strain–duration	4	14.34	19.73	23.21	7.00
Error *c*	12	23.33	23.67	28.18	23.74

*Number days after mating, eggs in the sample are from a 24-hr collection.
†$p \leq 0.05$.
‡$p \leq 0.01$.

TABLE 4. FECUNDITY ANALYSES OF *Drosophila* FEMALES REARED FROM EGGS THAT WERE COLLECTED FROM THE HYDROGEN FLUORIDE TREATED POPULATIONS

Sources of variation	d.f.	Mean squares collection day*						
		2	3	6	7	10	11	14
Replication	1	12.72	1.14	0.74	49.71	0.36	7.02	0.94‡
Level	2	40.65†	16.07	426.70†	375.99†	66.68	33.25‡	1.55‡
Error *a*	2	2.13	14.20	12.42	15.99	6.63	0.70	0.01
Strain	1	225.00‡	40.96	159.20†	640.94‡	308.59‡	128.07‡	1.14
Level–strain	2	4.73	8.58	114.85	155.51†	34.70‡	15.92†	1.39
Error *b*	3	5.90	4.39	14.21	10.09	0.63	0.75	0.23
Duration	2	39.72‡	56.58‡	471.16‡	247.35‡	79.63‡	63.14‡	5.66‡
Level–duration	4	14.56‡	22.30†	130.55‡	245.80‡	29.80‡	25.45‡	1.21‡
Strain–duration	2	1.16	69.02‡	172.19‡	59.36	30.21‡	24.58‡	0.94‡
Level–strain–duration	4	2.17	4.92	66.02	53.51	7.37	2.95	0.36
Error	12	0.74	4.91	22.20	29.88	2.61	0.99	0.12

*Number days after mating.
†$p \leq 0.05$.
‡$p \leq 0.01$.

variable, hatchability was depressed by increasing the HF concentration or by prolonging the duration of the treatment. The analyses suggested that the strain, level × strain and level × duration effects might be real, but the principal causes of variability were treatment level and duration.

The samples used for the hatchability tests were maintained until the adults could be sexed. Although a few significant deviations from a 1:1 sex ratio were found, there was no obvious pattern associated with them. Thus, the reduction in hatchability caused by HF treatment of the parents appeared to be independent of the sex of the embryo.

Fecundity was measured as the number eggs laid per female per day. Seven 24-hr egg collections were evaluated at periods ranging from two to 14 days after mating. Differences existed in the fecundity of the two strains and a natural decrease in fecundity as the flies aged was observed. However, treatment level, treatment duration and the interactions of duration with level and strain were all statistically significant (TABLE 4) at most collection dates. The depressing effect that prolonging the exposure to sub-lethal concentrations of HF had upon fecundity is of special interest, since the duration effect suggests genetic damage more explicitly than does treatment level. The females that were tested for fecundity were obtained from egg collections gathered from the various experimental populations, but all eggs were collected in a 24-hr period. There was one collection day (day 14) which exhibited significant differences between replicas. The reduction in fecundity was most pronounced in those females derived from the population treated longest. Thus, within a treatment level all eggs

TABLE 5. FERTILITY ANALYSIS OF ADULT *Drosophila* REARED FROM EGGS THAT WERE COLLECTED FROM THE HYDROGEN FLUORIDE TREATED POPULATIONS

Sources of variation	d.f.	Mean squares	F
Total	71		
Replication	1	35.30	14.16
Level	2	7.01	2.81
Error *a*	2	2.49	0.25
Strain	1	.11	0.01
Level–strain	2	3.00	0.30
Error *b*	3	10.10	1.28
Sex	1	35.65	4.53
Level–sex	2	8.39	1.07
Strain–sex	1	15.26	1.94
Level–strain–sex	2	13.25	1.68
Error *c*	6	7.88	1.16
Duration	2	25.82	3.81*
Level–duration	4	15.74	2.32
Strain–duration	2	2.95	0.43
Sex–duration	2	20.83	3.07
Level–strain–duration	4	5.68	0.84
Level–sex–duration	4	11.39	1.68
Level–strain–sex–duration	6	29.09	4.29†
Error *d*	24	6.78	

*$p \leqslant 0.05$.
†$p \leqslant 0.01$.

received the same exposure to HF, but the parent population exposure varied. The inverse relationship observed between the treatment duration of the parents and the fecundity of their female offspring suggests that exposure of *Drosophila* to low HF concentrations can cause genetic damage and that this genetic damage is accumulative as the exposure period is increased.

The fertility data (TABLE 5) show the least effect, there is a rather large but non-significant sex effect and a significant duration effect. It is probable that the experimental design used masked the nature of the fertility response. The fertility levels of the females were relatively stable for all treatment combinations. The effect on male fertility (TABLE 1) does not appear important except for those males derived from populations exposed 6 weeks. The *yw* strain was affected more than was the Oregon-r strain, but the male fertility of both strains was depressed at the highest HF level after 6 weeks exposure of the parent populations. This is reflected by the significance of the four-way interaction.

The two strains of *Drosophila* were selected because of their differential responses to the sub-lethal levels of atmospheric hydrogen fluoride which were used. Although many other factors may also be involved, the reduced hatchability and fecundity observed for the *yw* strain following exposure to HF can account for a major portion of the HF sensitivity of this strain.

REFERENCES

GERDES R. A. and SMITH J. D. (1967) A population cage. *Texas J. Sci.* **19**, 394–396.

GERDES R. A., SMITH J. D. and APPLEGATE H. G. (1971) The effects of atmospheric hydrogen fluoride upon *Drosophila melanogaster*—I. Differential genotypic response. *Atmospheric Environment* **5**, 113–116.

nospheric Environment Pergamon Press 1971. Vol. 5, pp. 123–135. Printed in Great Britain

THE CAPTURE OF SUB-MICRON AEROSOL PARTICLES BY SINGLE FINE FIBRES

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Abstract—Apparatus is described for the direct observation of the capturing of aerosol particles by single, very thin fibres. Particles of sub-micron diameters, and fibres of similar diameters were used. Other equipment described makes possible the direct assessment of deposits on individual fibres by use of an electron microscope.

By these means the nature of the flow around fibres as it depends on various aerosol and fibre parameters was studied. Also measurements were made of the capturing power of various fibres, and it was found that this may vary widely for what appear to be identical conditions.

These experiments indicate that current aerosol theory for the submicron region based on simplified geometrical ideas like diffusion and direct interception is unlikely to represent the facts of fibrous aerosol filtration. Factors such as surface condition of the fibres and electrostatic effects must be included before the theory can be considered realistic.

Résumé—Un appareil et un procédé pour surveiller directement la prise des particules d'aérosol par des fibres, individuelles et très ténues, sont décrits. Les diamètres des particules et aussi des fibres qui ont été utilisés étaient généralement de moins q'un micron. De plus, on a décrit un appareillage pour l'évaluation directe des sédiments sur une fibre unique en utilisant un microscope électronique.

Par ces moyens, on a étudié la nature de l'écoulement autour des fibres et la manière dont elle est sensible aux paramètres divers de l'aérosol et des fibres. En outre, on a mesuré la capacité des fibres diverses pour la prise des particules et les résultats peuvent être bien différents même sous des conditions qui semblaient être identiques.

Les résultats de ces essais ont mis en évidence que la théorie d'usage courante fondée sur les idées géometriques et simples par exemple ceux de la diffusion et de l'interception directe est peu probable représenter les faits de la filtration d'aerosols par des fibres. Il est nécessaire y comprendre d'autres facteurs notamment les effets électrostatiques et l'état de la surface des fibres.

INTRODUCTION

IN THE development of high efficiency fibrous filters, particularly for the filtration of aerosols of sub-micron particles, perhaps the most important factor has been the incorporation of very fine fibres, a few microns or less in diameter. Such filters have a very considerable capability of stopping highly dispersed aerosols, and in some cases a 99.98 per cent filtration efficiency is routine, though even this may be insufficient when highly toxic materials are involved.

In order to improve these filters, it is of great importance to know in detail the mechanisms of capture of a particle by a fibre. Recent books (DAVIES, 1966; FUCHS, 1964) deal at length with the theory of these mechanisms, and attempt to relate predicted results with experimental data. However, the theory considers highly idealized situations, as is stated in some cases (PICH, 1966, p. 224), and obviously cannot accurately predict the results in an actual physical case. For example, the flow of aerosol around perfect right circular cylinders is usually assumed, whereas it is very obvious to the experimenter that practical filters are not composed of such cylinders, and, even if they were, the initial deposition of particles would speedily alter these conditions drastically. It is further assumed that particles touching such

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