a density of only 1.03: it is spherical, having a diameter of 185 A and a molecular weight of 1,300,000. Only 23 per cent of the molecule is composed of polypeptide components, the remainder being phopholipid and cholesterol. In another paper Mr. Baldwin and J. W. Williams (Wisconsin) presented a general method for the determination of sedimentation constant distributions. The distribution completely defines the heterogeneity of the preparation with respect to sedimentation behavior, and the method is applicable even when the diffusion constant varies widely among the molecules. The theory does not apply to concentration-dependent systems, and all quantities must be extrapolated to infinite dilution. The standard deviation of the sedimentation constant distribution, as well as the weight average diffusion coefficient, can be calculated independently.

The organization of this conference was carried out by E. R. Blout and K. G. Stern. The success has led to plans for a similar conference during the last week in August 1951. This conference will be under the chairmanship of R. W. G. Wyckoff.

So you

Technical Papers

The Effect of X-Irradiation in Oxygen and in Hydrogen at Normal and Positive Pressures on Chromosome Aberration Frequency in *Tradescantia* Microspores¹

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Investigations by Giles and Riley (1, 2) demonstrated a marked effect of oxygen in increasing the frequency of x-ray-induced chromosomal aberrations in microspores of Tradescantia paludosa. In certain experiments (2), the relation between aberration frequency and percentage of oxygen present during irradiation was studied. Although there was an almost linear increase in aberration frequency between 2% and 21% oxygen (air), no significant difference was noted between 0% oxygen (pure helium) and 2% oxygen (plus 98% helium). Additional experiments have now been performed to reinvestigate the effect of low percentages of oxygen. In addition, studies have been made of the effect on aberration frequency of irradiation in various percentages of oxygen under pressure. Preliminary experiments have also been carried out on the effects of irradiation in hydrogen at normal and positive pressures. The general methods utilized for x-irradiation of inflorescences and subsequent cytological analysis have been described previously (\mathcal{Z}) .

In the experiments designed to reinvestigate the effects of low percentages of oxygen, special attempts were made to remove as much oxygen as possible from the inflorescences before irradiation. The following procedure was utilized: (1) The inflorescences, after being placed in the gas-tight exposure chamber, were subjected to prolonged evacuation. (2) Helium, which had been especially freed of any residual oxygen, was then introduced

¹Work performed under Contract No. W-7405-Eng-26 for the Atomic Energy Commission. into the evacuated exposure chamber. To remove any oxygen, the helium was passed slowly through a coil in liquid nitrogen and then over copper foil heated to 500° C before entering the exposure chamber. (3) Finally, the inflorescences were kept in the dark for an hour before irradiation to facilitate the removal by respiration of any further oxygen. In addition to the exposures in helium, irradiations were performed in atmospheres containing 2%, 5%, and 10% oxygen (plus helium). The percentages of oxygen in these gas mixtures were accurate (on the basis of analyses furnished by the vendor, the Ohio Chemical and Surgical Equipment Co.) to $\pm 0.2\%$. All experiments were carried out at room temperature (approximately 25° C). To increase the statistical reliability of the determinations, a larger number of cells was scored than previously. The data are presented in Table They are also plotted in Fig. 1, together with the 1.

TABLE 1

THE EFFECT OF IRRADIATION IN VARIOUS PERCENTAGES
OF OXYGEN ON THE FREQUENCY OF CHROMOSOMAL
ABERRATIONS IN Tradescantia MICROSPORES
(All X-Ray Exposures of 400 r at 50 r/min)

Oxygen per- centage	No. cells	No. inter- changes	Inter- changes /cell	Inter- stitial dele- tions	I.D./cell
0	1,000	234	0.234 ± 0.015	287	0.287 ± 0.017
2	1,200	347	0.289 ± 0.016	394	0.328 ± 0.017
5	1,000	427	0.427 ± 0.021	501	0.501 ± 0.022
10	600	364	0.607 ± 0.032	453	0.755 ± 0.035

averages of points at higher percentages of oxygen, which were obtained in previous experiments (2). These results indicate that there is still a substantial yield of aberrations even in the complete (or nearly complete) absence of oxygen. When oxygen is present during irradiation, there is a rapid rise in aberration frequency above this base level. This increase is linear between 0% and 10% oxygen, after which the rise is apparently more gradual, and shows a definite leveling off at around 20% oxygen.



The irradiation experiments in atmospheres of increasing oxygen concentration just outlined were all performed at normal atmospheric pressure (ca. 740 mm of Hg at Oak Ridge). Since the amount of dissolved oxygen is directly proportional to pressure, an experiment was designed to test for an oxygen effect in another way-by carrying out exposures in which different sets of inflorescences were irradiated in the same oxygen-helium gas mixtures, but at pressures of 1, 2, and 3 atm above normal pressure. The same exposure chamber was used as previously, and the pressure inside the chamber was measured by means of a standard gauge on the gas cylinder, the accuracy of which was checked against a mercury manometer over the range of the first 2 atm above normal. The data for exposures in 5% and 10% oxygen (plus helium) are presented in Table 2, together with results obtained in air and



in pure (ca. 99.8%) helium. The values for two separate experiments in 5% oxygen at normal pressure have been averaged in the table. During the exposure in compressed air at 3 atm above normal a small leak developed in the exposure chamber, and although the pressure gauge did not drop, it is felt that the value for this exposure may not be as reliable as for the other observations.

These results show that there is a marked increase in aberration frequency accompanying increasing pressure in those experiments in which oxygen was present during irradiation. When exposures occurred in the absence of oxygen (in pure helium), however, no increase resulted, indicating that pressure alone, at least within this range, did not influence the yield of aberrations. In Fig. 2 the

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Pressure in exposure chamber		No. cells	No. inter- changes	Interchanges /cell	No. interstitial deletions	Interstitial deletions /cell
	Irradiated	l in 10%	oxygen (plu	s 90% helium)		
Normal atmospheric (ca. 740 mm	Hg)	800	474	0.59 ± 0.027	599	0.75 ± 0.031
One atm above normal		550	436	0.79 ± 0.038	563	1.02 ± 0.043
Two atm above normal		700	584	0.83 ± 0.035	875	1.25 ± 0.042
Three atm above normal		700	584	0.83 ± 0.035	732	1.05 ± 0.039
	Irrada	lated in ;	pure helium	$(\sim 99.8\%)$		
Normal atmospheric (ca. 740 mm	Hg)	9 50	229	0.24 ± 0.016	256	0.27 ± 0.017
Three atm above normal		1,000	223	0.22 ± 0.015	259	0.26 ± 0.016
		Irra	diated in air			
Normal atmospheric (ca. 740 mm	Hg)	1.000	713	0.71 + 0.027	780	0.78 + 0.028
Three atm above normal		950	786	0.83 ± 0.030	994	1.05 ± 0.033
	Irradiate	d in 5%	oxygen (plus	95% helium)		
Normal atmospheric (ca. 740 mm	Hg)	1.850	903	0.49 + 0.016	954	0.52 ± 0.017
One atm above normal	5.	950	583	0.61 ± 0.025	671	0.71 ± 0.027
Two atm above normal		1,000	683	0.68 ± 0.026	802	0.80 ± 0.028
Three atm above normal		840	657	0.78 ± 0.031	783	0.93 ± 0.033

TABLE 2

THE EFFECT OF PRESSURE ON THE FREQUENCY OF X-RAY-INDUCED CHROMOSOMAL ABERRATIONS IN Tradescantia MICROSPORES IRRADIATED IN ATMOSPHERES CONTAINING VARIOUS PERCENTAGES OF OXYGEN. (All Exposures of 400 r at 50 r/min)

TABLE 3

THE EFFECT OF IRRADIATION IN HYDROGEN ON CHROMOSOME ABERRATION FREQUENCY IN *Tradescantia* MICROSPORES (400 r at 50 r/min)

Pressure in exposure chamber	No. cells	No. inter- changes	Inter- changes c/cell	Inter- stitial dele- tions	I.D./cell
Normal atmos- pheric (ca. 740 mm Hg)	707	149	0.21 ± 0.017	166	0.23 ± 0.018
Three atm above normal	498	100	0.20 ± 0.020	124	0.25 ± 0.022

frequencies of chromosomal interchanges at various pressures in 5% and 10% oxygen have been plotted against the standard curve from Fig. 1, on the assumption that the amount of dissolved oxygen in the microspores is directly proportional to the pressure, and that this value determines the aberration yield at the standard dose of 400 r at 50 r/min. As can be seen from the graph, the agreement with previous results at comparable partial pressures of oxygen is very good. These results thus support earlier views that the amount of dissolved oxygen present in cells is an important factor in determining aberration frequency. They indicate that pressure alone, at least within the range up to 3 atm above normal, does not influence this type of radiation-induced change.

Several different types of evidence (3) support the view of Thoday and Read (4) that the effect of oxygen in increasing chromosome aberration frequency may result from an indirect action of x-radiation to decompose water with the production of hydrogen peroxide. However, although very little hydrogen peroxide appears to be produced by x-rays in oxygen-free water (5, 6), it will be recalled (Table 1) that there is still a substantial aberration frequency in inflorescences exposed in purified helium to 400 r. The question naturally arises as to the mechanism by which these aberrations are produced. It seems quite probable (3) that the effect of a substance such as hydrogen peroxide on aberration frequency would arise from an increased production of chromosome breaks, rather than from an influence on the reunion behavior of broken ends. If this is the case, aberrations induced by x-rays in the absence of oxygen might result (at least in part) from breaks produced by another substance, the OH radical, which is formed by the radiodecomposition of oxygen-free water. On the other hand, all the aberrations produced in the absence of oxygen might arise from the direct action of the radiation in ionizing the molecules of the chromosomes themselves.

A preliminary attempt has been made to distinguish between these two possibilities by means of experiments designed to remove the OH radical by promoting, during x-irradiation, the back reaction to form water. Allen (6) has shown that the presence of molecular hydrogen markedly accelerates this back reaction. Consequently, inflorescences were irradiated in pure hydrogen at normal atmospheric pressure and at 3 atm above normal. These data are presented in Table 3. Although there is some decrease in aberration frequency compared to earlier results obtained with irradiation in helium, the differences for the most reliable observations, those of interchanges, are not significant. In this experiment it was not possible to obtain evidence that hydrogen was actually present in the cells during irradiation. However, previous experiments with oxygen (2) have indicated that under similar conditions this gas diffuses into microspores very Thus, despite the fact that hydrogen is less rapidly. soluble in water than is oxygen, it seems reasonable to conclude that a considerable amount of hydrogen would be present in the microspores during irradiation, and that this should react to remove the OH radical. Consequently, the failure to find a significant decrease in aberration frequency when x-radiation is performed in hydrogen may mean that all the residual aberrations induced by x-rays in the absence of oxygen arise as a result of a direct effect of the radiation on the chromosomes. Tt. should be pointed out, however, that this conclusion is based on the assumption that reactions leading to H_2O_2 production or suppression in cells from which oxygen has been removed as completely as possible are similar to those occurring in oxygen-free pure water. There is as yet little experimental evidence on this point, and it is quite possible that the complexity of the cellular environment may cause very different reactions to occur.

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The Detection of Internally-borne Bacterial Pathogens of Beans by a Rapid Phage-Plaque Count Technique¹

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The use of specific bacteriophages for identifying bacterial species or strains is a valuable aid in medical and epidemiological work; phage typing of enteric pathogens, staphylococci, and streptococci is extensively employed (1). Phage has also been widely used to separate closely related groups, species, and strains of bacteria for purposes of classification (1, 2). Within recent years bacterial viruses have been recognized as very useful tools in the rapid identification of plant

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