9 HEAVY ION INDUCED MUTATIONS IN MAMMALIAN CELLS: CROSS SECTIONS AND MOLECULAR ANALYSIS (STATUS REPORT)

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Our investigations of heavy ion-induced mutations in mammalian cells, which had been begun a few years ago, were systematically continued. For the first time, it was possible to cover a large LET range with a few kinds of ions (Fig. 1). To do this, both UNILAC and SIS were used to yield comparable data for a large energy range. This is a necessary condition for a comprehensive description of the influence of such ion parameters as energy and LET. In these experiments, the induced resistance against the poison 6-thioguanin (6-TG), which is linked to the HPRT locus on the genome, is being used as mutation system.

The cells used so far are V-79 Chinese Hamster cells, but recently considerable efforts have been made to find a suitable human system. Preliminary experiments were performed with the P3 cell line originally isolated from a teratocarcinoma of a woman and the MGH-U1 cell line derived from a bladder carcinoma of a man. All data presented in this part of the report were obtained with V-79. Table 1 lists the data collected so far.

The LET dependence of mutation induction is displayed in fig. 1 for a few selected ions. The course of the curves for the various ions seems to be qualitatively similar; a systematic relation, however, between the cross sections σ_m and the LET does not seem to exist. Each ion appears to have its own specific curve. This confirms earlier observations made with other systems and end points, namely that the track structure of a specific type of ionising radiation plays a very important role, and the LET can not serve as a unifying parameter. On the basis of these and future data, it is planned to develop a theoretical description of the atomic mechanisms underlying the biological action of ionising radiation.

In addition to the mutation-induction cross-section measurements, the molecular changes of the DNA are being investigated by means of Multiplex PCR ("Polymerase Chain Reaction") gene amplification. From these experiments we expect further elucidation of the mutation-inducing mechanisms composing the biological action of heavy-ion radiation. First experiments have been performed at the Department of Clinical Genetics at the University of Ulm (Prof. G. Speith), but by now this method is being used regularly in our own laboratory as well. Contrary to what one might expect, first results suggest that heavy-ion radiation does not only produce deletions of larger parts of the gene, but also "point mutations" meaning that single bases are lost or altered. Even in this context, track-structure parameters appear to play an important role. Heavy ions of relatively low kinetic energy and correspondingly small penumbra radii seem to result mainly in large deletions, as opposed to high-energy heavy ions, which yield a large portion of small, localised alterations of the DNA.

Table 1: Physical parameters and biological results from the various heavy ion exposures.
* : Data from Kranert *et al.*, (1990).

Ion	E/M at cell surface (MeV/u)	LET (H_2O) ($keV/\mu m$)	Z^{*2}/β^2	rp (μm)	σ_i (μm^2)	σ_m ($10^{-4}\mu m^2$)	σ_m/σ_i (10^{-5})
O	1.9	754	11455	0.18	71.2±4.7	11.4±0.6	1.6±0.2
	* 8.8	276	3182	2.5	50±4	20.0±4.0	4.0± 0.8
	10.7	238	2663	3.5	49.5±2.7	9.5 ±1.1	1.91±0.2
	88	46	387	125	4.3±0.2	1.22±0.12	2.88±0.4
	396.0	18	126	1606	1.3±0.1	0.17±0.01	1.3±0.2
Ne	8.0	452	5293	2.11	45±4	21.3±3.0	4.75±1.1
	10.7	366	4070	3.5	52±4	15.7±5.0	3.03±1.2
	* 12.0	335	3666	4.2	42±3	15.0±5.5	3.5 ±1.3
	14.3	294	3127	5.7	33±4	7.7±2	2.32±0.9
	65	91	792	74	12.5±1	2.7±0.5	2.18±0.5
	191	42	321	465	4.7±0.2	1.1±0.1	2.26±0.3
	395	28	197	1599	2.1±0.2	1.0±0.1	4.73±0.7
Ar	5.6	1611	19633	1.15	50±3.5		
Ca	14.1	1088	11509	5.5	46±7	7.0±0.4	1.5±0.25
Ti	4.8	2414	29779	0.89	54±3	14.0±1.0	2.6±0.3
	15.0	1238	12997	6.15	50±6	8.6±1.6	1.7±0.38
Ni	6.0	3190	37205	1.3	61±6	9.1±0.8	1.5±0.3
	* 9.5	2517	27580	2.8	65±2	8.3±1.2	1.3±0.18
	* 14.3	1961	20535	5.7	87±5	5.7±1.8	0.65±0.05
	136	407	3285	261	52±2	5.6±1.1	1.1±0.2
	387	218	1565	1544	39±3	5.5±0.5	1.4±0.32
	630	180	1225	3536	38.5±1	6.2±0.6	1.6±0.2
Xe	9.7	7126	72739	2.9	70	12.0±2.5	1.7±0.36
Au	2.2	12895	193900	0.24	57±2	4.1±0.7	0.7±0.2
	8.7	12568	126411	2.44	90±6	8.3±2.1	0.9±0.3
Pb	11.6	11948	116633	4.0		8.7±0.6	
	* 15.2	10800	102501	6.3	88±8	9.2±2.6	1.1±0.33
	150	3090	23064	308	97±5	14.5±2.5	1.5±0.38
	500	1630	11489	2387	68±3	8.9±1.3	0.9±0.2
	980	1325	8829	7493	52±5	8.3±1.2	1.2±0.23
U	3.9	15817	195911	0.62	71±5	15.0±2.0	2.1±0.37
	10.8	15220	139202	3.5	105±10	8.5±1.5	0.8±0.16
	12.7	13468	129809	4.6	90±9	4.5±2.0	0.5±0.20
α	0.85	163			42	11.9	2.8

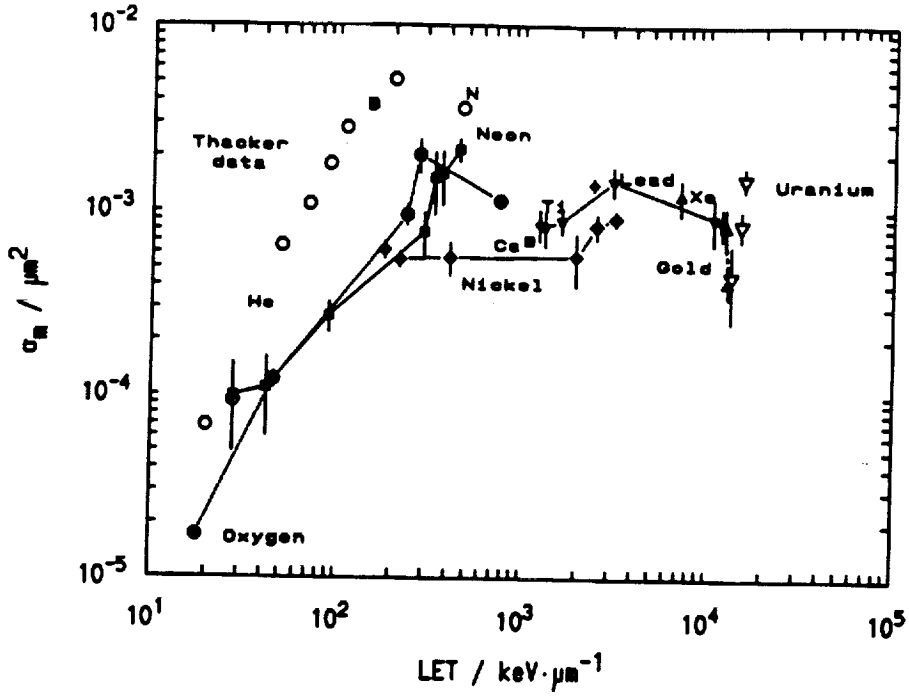

Figure 1: Mutation induction cross sections plotted versus LET together with data from Thacker *et al.*, (1979) (open circles) and 3 uranium experiments from Kranert *et al.*, (1990).

References

[1] T. Kranert, E. Schneider, and J. Kiefer. Mutation induction in V79 Chinese hamster cells by very heavy ions. *Int. J. Radiat. Biol.*, 58:975–987, 1990.

[2] J. Thacker, A. Stretch, and M. A. Stephens. Mutation and inactivation of cultured mammalian cells exposed to beams of accelerated heavy ions. II. Chinese hamster V79 cells. *Int. J. Radiat. Biol.*, 36:137–148, 1979.