number of such elements can be harmful. The present results did not reveal any morphological and/or anatomical difference between the individual with the extra element and the one with a normal karyotype. Again, lack of data on the C-banding pattern, cannot unequivocally establish this extra chromosome as a supernumerary. This acrocentric extra chromosome must necessarily owe its origin through pericentric inversion or, independently from the cumulative effect of the naturally occurring radioactive elements in the area from where the specimens were collected.

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CLASTOGENIC EFFECT OF WR-2721 ON MOUSE CHROMOSOMES

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THE protective action of the drug WR-2721 is reported to increase with dose, giving a DRF (dose reduction factor) of 2.7 at 500 mg/kg body weight, the maximum tolerated dose in mice[1], which is very near its toxic LD_{50} of 550–780 mg/kg for different mouse strains[2]. This drug has been recommended for use in human protection for both military and clinical purposes at the maximum tolerated dose[3]. However, Phillips[4] listed a variety of toxic effects of WR-2721 in various species including man. Earlier studies from this laboratory[5] also showed weight loss and peripheral lymphopenia in mice receiving 400 mg/kg and 500 mg/kg WR-2721 i.p., while 200 mg/kg and 300 mg/kg did not produce such effects. Therefore, to determine a safe and effective dose which can be administered in clinical radioprotection without untoward side effects, the effect of this drug at different doses was studied on the mouse bone marrow chromosomes.

Six to eight week-old male Swiss albino mice were given single i p. injections of 200, 300, 350 or 400 mg/kg b.wt. of WR-2721 (phosphorylated, lot no. H-12, YM 08310), obtained from Yamanouchi Co., Tokyo, Japan, and dissolved in double-distilled water. The bone marrow chromosomes were studied at different post-injection periods from 1 day to 28 days, after arresting mitosis by a prior colchicine treatment and preparing metaphase plates by the modified air-drying method of Kilian[6]. Animals were sacrificed by cervical dislocation. The chromosome aberrations were scored under oil immersion and the different types of aberrations enumerated separately.

The results are presented in table 1. The lowest drug dose used (200 mg/kg) showed identical values of aberration frequency as in the normal untreated animals at all the autopsies. Hence the values are not given in the table. With increase in the drug dose, there was a corresponding rise in the number of aberrations which was most evident on day 7 post-injection. With 300 mg/kg, there was a slight increase in the percentage of aberrant cells which further increased in the 350 mg/kg and 400 mg/kg groups, where the number was significantly higher than the normal at this interval. In all the groups, the values came down after day 7, but in the higher dose groups the aberrant cell frequency was maintained higher than normal throughout the experiment (table 1). In the 400 mg/kg group a significant increase in aberrant cells was seen as early as day 1 after the drug injection; thereafter the number came down to normal, to rise again on day 7. This increase in all the groups was mainly due to stable aberrations of which chromatid breaks dominated. In the higher dose groups, unstable aberrations, represented mainly by fragments and polypolidy also contributed at the later intervals, *i.e.* from day 7 on.

Table 1 *Frequency of different types of chromosome aberration in Swiss albino mice treated with 300, 350 or 400 mg*kg b. wt. of WR-2721. No. of cells studied for each interval = 600. (Percent abberrant cells in untreated control* 0.33 ± 0.21)

Dose of drug (mg/kg b. wt.) WR-2721.	Post-treatment time (days)	Total No. of aberrant cells	Percent abberrant cells ± S E.	Types of abberrations				
				Chromatid breaks	Chromosome breaks	Fragments	Multiple breaks	Poly ploidy
300	1 d	2	0.33 ± 0.21	2	–	–	–	–
	2 d	2	0.33 ± 0.21	2	–	–	–	–
	4 d	2	0.33 ± 0.21	1	–	1	–	–
	7 d	4	0.66 ± 0.42	2	–	2	–	–
	10 d	2	0.33 ± 0.21	1	–	1	–	–
	14 d	2	0.33 ± 0.21	1	–	–	–	1
	21 d	3	0.5 ± 0.13	2	–	1	–	–
	28 d	2	0.33 ± 0.21	2	–	–	–	–
350	1 d	3	0.5 ± 0.13	2	1	–	–	–
	2 d	2	0.33 ± 0.21	1	–	1	–	–
	4 d	2	0.33 ± 0.21	1	1	–	–	–
	7 d	6	1.00 ± 0.26 $P < 0.05$	4	1	1	–	–
	10 d	3	0.5 ± 0.13	2	–	1	–	–
	14 d	4	0.66 ± 0.42	2	–	1	–	1
	21 d	3	0.5 ± 0.13	1	1	–	–	1
	28 d	3	0.5 ± 0.13	2	–	–	–	1
400	1 d	6	1.00 ± 0.26 $P < 0.05$	10	–	–	1	–
	2 d	2	0.33 ± 0.21	1	1	–	–	–
	4 d	2	0.33 ± 0.21	1	–	1	–	–
	7 d	9	1.50 ± 0.59 $P < 0.05$	11	–	1	–	1
	10 d	3	0.5 ± 0.13	1	–	1	–	1
	14 d	4	0.66 ± 0.42	2	–	1	–	1
	21 d	4	0.66 ± 0.42	6	–	–	–	–
	28 d	4	0.66 ± 0.42	2	1	–	–	1

The action of WR-2721 in inducing the chromatid breaks in the present study appears to be similar to the effect of ionizing radiation, and shows a dose-dependent increase. In the 400 mg drug-treated animals, there was an early induction of breaks which was evident by day 1, followed by a fall to normal, only to reappear on day 7. This leads to the assumption that further division of the affected cell is delayed considerably and the anomalies are expressed when the cells again enter division. This is followed by an elimination through cell death, resulting in a reduction in the number of aberrant cells at later intervals. This elimination may result from an increase in the injury due to the expression of latent breaks (multiple fragments) and/or an effect on the membrane which prevents cytokinesis, leading either to cell death or polyploidy. In the 350 and 400 mg/kg groups, persistence of polyploidy is seen at later intervals and its appearance is earlier when the drug dose is higher. The occurrence of polyploidy may also be attributed to a toxic effect on the spindle. Jackson and Lindahl-Kiessling[7] observed that cysteamine, the parent compound of WR-2721, induces polyploidy in human leucocyte cultures stimulated to divide by phytohaemagglutinin. They stated that the changes induced by sulphydryl compounds are remarkably similar to changes in neoplasia. Therefore the suitability of WR-2721 for use in clinical studies as a safe radioprotector at the maximum tolerated dose may not be assessed only on the basis of LD_{50} toxicity studies.

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SCANNING ELECTRON MICROSCOPE OBSERVATIONS ON THE COPULATORY AND PENIAL SETAE OF THE EARTHWORM *OCTOCHAETONA PATTONI* (MICHAELSEN)

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COPULATORY and penial setae in earthworms are of significance as characters of taxonomic importance offering good means of identification. *Octochaetona pattoni* has four pairs of penial setae, a pair in each of the four male openings in segments 17 and 19; and copulatory setae on 8 and 9. But for the ultrastructure of the penial setae of *Lampito mauritii*[1] there is no available detailed description of the setal structures of tropical earthworms.

Penial and copulatory setae were processed as described earlier for *L. mauritii*[1]. The setae were scanned in a JEOL 100S transmission-cum-scanning electron microscope, at the Tata Institute of Fundamental Research, Bombay.

The penial setae of *O. pattoni* were earlier described by Stephenson[2] as 1.7 to 2 mm long, 17 μ thick, slightly but regularly curved the distal fourth having sharp lateral edges, which become expanded at the tip forming a sort of shovel, the shaft being ornamented with rings of teeth.

The tip of the seta under SEM reveals the shovel to have distinct extensions (figure 1). The curvature of the shovel is 25 μ and the distance across the tips of the lateral edges is about 17.5 μ. That the shovel is medial in origin is evident from the borken tip in figure 2. The presence of pores at the base of the shovel cannot be overlooked, but this needs confirmation through ultrasectioning of the penial setae.

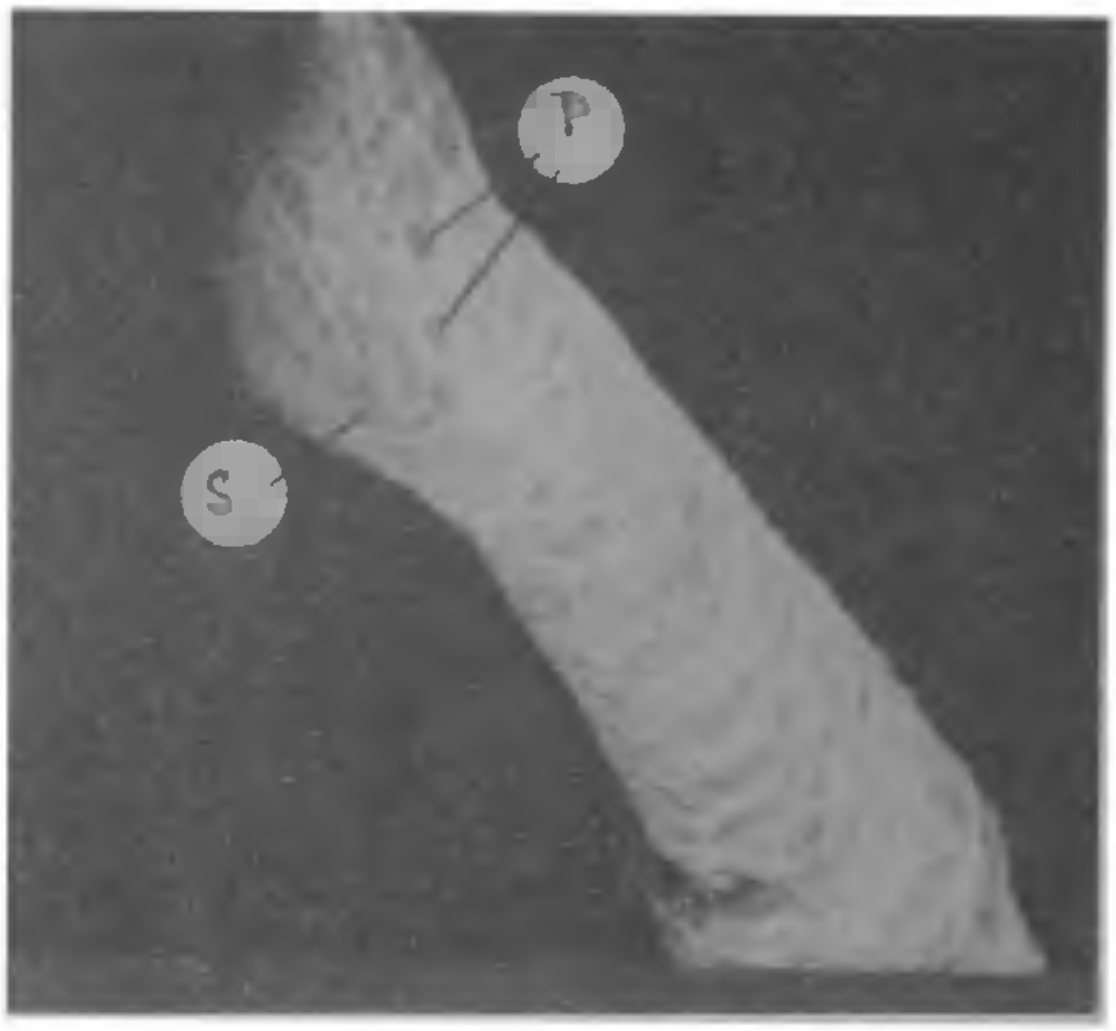

Figure 1. Distal end of penial seta. P—pores, S—shovel (× 2000)

Figure 2. Medial origin of the shovel in a penial seta. MO—medial origin (× 2500)