## Chromosomal Polymorphism in the Guinea-Pig, Cavia porcellus<sup>1</sup>

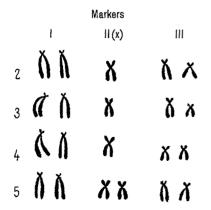
Barring human materials, a number of cases of chromosomal polymorphisms has been discovered in mammals<sup>2-6</sup> and the present report adds one more to the mammalian group.

In the course of investigations on the chromosome number and morphology of the guinea-pig, Cavia porcellus, for the study of the state of chromosomes during differentiation, we discovered the occurrence of natural heterozygotes with respect to some 'marker' chromosomes in some individuals of this species. As far as we are aware, the cytological investigation on the Indian guinea-pig, C. porcellus, has not been carried out. Ten guinea-pigs, seven males and three females of non-pedigree stock obtained from local dealers, have been examined cytologically employing special techniques 7.8 with some modifications made by us (Manna and Talukdar, unpublished).

The diploid chromosome number in *C. porcellus*, like *C. cobaya*<sup>9</sup>, is 64 (Figure) in various tissues of both sexes. The chromosomes can be arranged in 31 homologous



Fig. 1. Mitotic metaphase with 64 chromosomes in the corneal cell of a homozygous male (ca. ×1600).



Figs. 2-5. Only the 'marker' chromosomes from the full somatic metaphase complements in different types of individuals are drawn, Y chromosome in males not shown (ca. ×2600). Fig. 2. Male, homozygous for two large 'marker III'. Fig. 3. Male, heterozygous for 'marker III'. Fig. 4. Male, homozygous for two small 'marker III'. Fig. 5. Marker chromosomes in a homozygous female individual.

autosomal pairs and a pair of sex-chromosomes which is homomorphic XX in the female and heteromorphic XY in the male, the X being appreciably larger than Y. Three pairs of chromosomes stand out distinct and are designated as 'marker' chromosomes (Figures 2–5). Among them, 'marker I' consists of a pair of acrocentric autosomes which are largest of all. No marked size difference exists between 'markers II and III' but they can easily be distinguished from the position of their centromeres. Marker II represents the X chromosome. Therefore, it is single in males (Figures 2–4) and paired in females (Figure 5). The remaining marker chromosomes, designated as 'marker III', are a homologous pair of sub-metacentric autosomes.

Out of ten guinea-pigs examined, three males were heterozygous for the 'marker III' (Figure 3). The size difference between two components of the heteromorphic pair of 'marker III' was appreciable and the smaller member of the heteromorphic pair was sometimes confused with the next lower size group. The point of dissimilarity between the two members of 'marker III' was the relative difference in the length of the two chromosomes. The position of centromeres in the two chromosomes of 'marker III' in heterozygous individuals also appeared to be dissimilar in some cases. As a result of heteromorphism of 'marker III', the chromosome complements of different individuals of the species, with random mating, were found to be of three kinds: (i) homozygous for two large 'marker III' chromosomes (Figure 2), which may be represented symbolically as AA, (ii) heterozygous for one large and one small or Aa chromosome (Figure 3), and (iii) homozygous for two smaller third marker chromosomes or aa (Figure 4). However, out of the three expected types, only two, namely, the heterozygous and the homozygous individuals, could be identified with surety. It was rather difficult to distinguish accurately the two types of homozygotes in all cases, e.g. AA and aa types. A critical analysis is now in progress to overcome this difficulty concerning the need for accurate determination of the number of the two types of homozygotes in the study of their frequency in a population. Since out of ten guinea-pigs, three were heterozygous for 'marker III', the frequency of natural heterozygotes seems to be quite high. The determination of the frequency of natural heterozygotes in different domestic colonies is in progress.

In the guinea-pig which was heterozygous for 'marker III', cells of bone-marrow from tibia and femur and of cornea were found to contain the heteromorphic pair in the metaphase chromosome complements.

Besides the heteromorphism shown with regard to 'marker III', sporadic occurrences of heteromorphic ap-

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pearance of other chromosomes in some cells of the same individual were also observed. The significance of the various types of chromosomal polymorphism will be published elsewhere.

Zusammenfassung. Bei der karyologischen Analyse von Metaphasen in Knochenmark, Cornea und andern Geweben des indischen Meerschweinchens, Cavia porcellus

(7 ♂♂, 3 ♀♀) wurde festgestellt, dass drei männliche Individuen in einem autosomalen Chromosom heterozygot waren. Das Chromosom wird als Marker III bezeichnet.

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## In vitro Antiviral Activity of Hydrazine Sulfate

In previous publications we have shown the inhibitory properties of guanidine on poliovirus replication 1.2.

During our efforts to elucidate the structure-activity relationships for this compound, we were able to demonstrate the antiviral activity of hydrazine sulphate.

In fact hydrazine sulphate was found to be able to inhibit polio and vaccinia virus multiplication in vitro.

The viral strains and the procedures used for the culture of human amnion cells (Mascoli's line) were as previously described <sup>3, 4</sup>.

The assays of poliovirus cytopathic effect (CPE) were performed by the plaque method of DULBECCO<sup>5</sup>; those on vaccinia virus CPE were previously described by us <sup>6</sup>.

The inhibition of polio and vaccinia virus replication was evaluated by counting the cytopathic units (CPU) present at various time intervals in the culture medium.

Since hydrazine lowers the pH of the medium, this was kept at 7.3 by adding a few drops of 1% NaHCO<sub>3</sub> solution.

Table I shows a clear-cut inhibition of polio 1 and vaccinia virus CPE for concentrations of hydrazine up to 8  $\mu$ g/ml and up to 16  $\mu$ g/ml respectively.

The protective effect against the CPE of either polio or vaccinia virus is accompanied by a marked inhibition of the virus replication, as is shown in Table II.

The inhibition of the viral replication exerted by hydrazine stems neither from a direct effect on viral particles nor from a detectable cell damage. Indeed, preincubating polio and vaccinia virus in a medium containing 100  $\mu$ g/ml of hydrazine does not significantly modify the viral infectivity; on the other hand, either polio or vaccinia viruses normally replicate in cells pre-incubated

Table I. Inhibition by hydrazine sulphate of polio 1 and vaccinia virus cytopathic effect

Virus	Hydrazine sulphate μg/ml*	% of inhibition (mean and range)	
Polio 1	66.6 (19)	93.6 (72.5-100)	
Polio 1	33.3 (16)	94 (66 -100)	
Polio 1	16.6 (13)	63 (22 -100)	
Polio 1	8.3 (7)	61 (14 -100)	
Polio 1	4.1 (3)	38 (12 - 75)	
Polio 1	1.0 (2)	13 (0 - 26)	
Vaccinia	66.6 (6)	89.7 (82 - 94)	
Vaccinia	93.3 (5)	70.8 (40.3- 95.9)	
Vaccinia	16.6 (3)	60.5 (49 - 73)	
Vaccinia	8.3 (3)	29 (0 - 51)	
Vaccinia	4.1 (3)	4 (0 - 12)	
Vaccinia	4.1 (3)	4 (0 - 12)	

<sup>&</sup>lt;sup>a</sup> In parentheses the number of trials.

for 10 h with hydrazine (67  $\mu$ g/ml) and then washing out hydrazine.

The mechanism by which hydrazine exerts its antiviral effect is not yet clear.

Table II. Inhibition by hydrazine sulphate of polio and vaccinia virus replication

Viral strain	Inoculum in CPU*	Hydrazine sulphate μg/ml¤	CPU detected after 36 h (mean and range)
Polio 1	104	- (4)	$1.4 \times 10^{7} $ $(7 \times 10^{6} - 3 \times 10^{7})$
Polio I	104	66.6 (4)	$2.4 \times 10^5$ $(10^5 - 4.2 \times 10^5)$
Polio 1	104	33.3 (2)	$2.6 \times 10^{6}$ $(2.2 \times 10^{6} - 3 \times 10^{6})$
Polio 1	104	16.6 (2)	$5.4 \times 10^{6}$ $(5.3 \times 10^{6} - 5.6 \times 10^{6})$
Polio 1	104	8.3 (2)	$10^{7}  (9.4 \times 10^{6} - 1.1 \times 10^{7})$
Vaccinia	$5 \times 10^{8}$	- (3)	$10^{6}$ $(3 \times 10^{6} - 8 \times 10^{5})$
Vaccinia	$5 \times 10^{3}$	66.6 (3)	$10^4  (7 \times 10^9 - 3 \times 10^4)$
Vaccinia	$5 \times 10^3$	33.33 (3)	$\begin{array}{c} 1.8 \times 10^{5} \\ (9 \times 10^{4}  3 \times 10^{5}) \end{array}$

a Cytopathic unities. b In parentheses the number of trials.

Riassunto. Il solfato di idrazina inibisce l'effetto citopatico e la moltiplicazione sia del virus polio che di quello vaccinico. Tale azione non deriva nè da un effetto sulla particella virale nè da un danno cellulare.

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