

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^{3,4}.

The assays of poliovirus cytopathic effect (CPE) were performed by the plaque method of DULBECCO⁵; those on vaccinia virus CPE were previously described by us⁶.

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% NaHCO₃ solution.

Table I shows a clear-cut inhibition of polio 1 and vaccinia virus CPE for concentrations of hydrazine up to 8 µg/ml and up to 16 µg/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, pre-incubating polio and vaccinia virus in a medium containing 100 µg/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

for 10 h with hydrazine (67 µg/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 ^a	Hydrazine sulphate µg/ml ^b	CPU detected after 36 h (mean and range)
Polio 1	10 ⁴	- (4)	1.4 × 10 ⁷ (7 × 10 ⁶ -3 × 10 ⁷)
Polio 1	10 ⁴	66.6 (4)	2.4 × 10 ⁵ (10 ⁵ -4.2 × 10 ⁵)
Polio 1	10 ⁴	33.3 (2)	2.6 × 10 ⁶ (2.2 × 10 ⁶ -3 × 10 ⁶)
Polio 1	10 ⁴	16.6 (2)	5.4 × 10 ⁶ (5.3 × 10 ⁶ -5.6 × 10 ⁶)
Polio 1	10 ⁴	8.3 (2)	10 ⁷ (9.4 × 10 ⁶ -1.1 × 10 ⁷)
Vaccinia	5 × 10 ³	- (3)	10 ⁶ (3 × 10 ⁵ -8 × 10 ⁵)
Vaccinia	5 × 10 ³	66.6 (3)	10 ⁴ (7 × 10 ³ -3 × 10 ⁴)
Vaccinia	5 × 10 ³	33.33 (3)	1.8 × 10 ⁵ (9 × 10 ⁴ -3 × 10 ⁵)

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

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

Virus	Hydrazine sulphate µg/ml ^a	% 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	33.3 (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)

^a 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 vaccinnico. Tale azione non deriva nè da un effetto sulla particella virale nè da un danno cellulare.

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Istituti di Farmacologia, Igiene e Microbiologia dell'Università, Cagliari (Italy), February 11, 1964.

¹ B. LODDO, Boll. Soc. ital. Biol. sper. 37, 395 (1961).

² B. LODDO, W. FERRARI, G. BROZZU, and A. SPANEDDA, Nature 193, 97 (1962).

³ B. LODDO, W. FERRARI, G. BROZZU, and A. SPANEDDA, Boll. Ist. sioroter. milan. 41, 111 (1962).

⁴ B. LODDO and C. E. ZANDA, Arch. int. Pharmacodyn. Théor. 133, 1 (1961).

⁵ R. DULBECCO, Proc. Nat. Acad. Sci. 38, 747 (1952).

⁶ B. LODDO, M. L. SCHIVO, and W. FERRARI, Lancet 1963ii, 914.