cortical steroids on CO_2 evolution or fatty acid synthesis by mammary gland slices(11), on fatty acid synthesis by rat adipose tissue (12), or on glucose uptake by rat diaphragm (13,14,15).

Summary. The stimulation of glucose uptake or CO_2 evolution produced by 20, 50, or 10^4 μu insulin per ml *in vitro* to rat epididymal adipose tissue was markedly reduced by prior subcutaneous injection of the rat tissue donor with 7 mg cortisone acetate in divided doses over a period of 3½ days.

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Action of Mucoproteins *in vivo* and in Tissue Culture on Poliomyelitis Virus. (25642)

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The present report contains our findings on the protective effect of various mucoprotein fractions obtained from hog gastric mucosa in experimental poliomyelitis infection of mice (Lansing strain II) and the influence of the same proteins on development of the virus in monkey kidney cells.

Materials and methods. Three hog gastric mucosa fractions (A,B,C) received from Prof. V. Capraro (Milano, Italy) and 2 extracts (D,E) prepared in the Armour Laboratories by E. Samsa, V. Snider and P. Van Melle were used for study. The characteristics of these substances as determined by V. Capraro are: (A) A and H group specific substance, 95% pure by electrophoretic determination (free phase). Anti-agglutinating potency 10^7. (B) "B_{12} binding factor"(1) 100% pure. B_{12} binding titer, 22 μg/mg in the *E. coli* assay. (C) Acid aminopolysaccharide. Electrophoretically 95% pure, uronic acid content 35%. (D) Armour preparation KO-45-102-1. Uronic acid content 1.62%. (E) Armour preparation KO-45-124. B_{12} binding titer 228 mμg/mg, uronic acid content 1.6%. Doses of 1 and 2 μg showed A, respectively H substance activity, inhibiting 10 minimal agglutinating doses in the systems A red cells/anti A serum and O cells/anguilla anti H serum.

Infection. Swiss mice of 14-18 g, 4-5 weeks of age, were inoculated intracerebrally with 0.03 ml Lansing strain (Type II) virus, diluted 1:3000. Standard procedure was to infect mice Friday and begin treatment Monday; however on 4 groups of mice treatment was started prior to inoculation. Ten treatments were given, 2 daily for a period of 5 days, except when otherwise stated. Experimental animals were observed daily for a pe-

TABLE I. Action of Mucoproteins in Experimental Poliomyelitis (Lansing II) Infection of Mice.

Exp. No.	Preparation	Dose, mg/kg × No. treatments	Route	Survivors/total mice after: 10 days	20 days	30 days	60 days
I	A	10 × 10	Subcut.	10/10	7/10	4/10	3/10
		2.5 "		"	9/10	6/10	"
	B	10 "		6/10	4/10	2/10	2/10
		2.5 "		9/10	7/10	3/10	3/10
	C	10 "		"	4/10	2/10	2/10
		2.5 "		"	"	"	"
	E	10 "		10/10	6/10	5/10	4/10
		2.5 "		9/10	"	4/10	3/10
		Controls		"	4/10	0/10	
II	E	20 × 10	Oral	10/10	2/10	1/10	1/10
		Controls		"	"	0/10	
III	D	20 × 10	Oral	14/15	8/15	8/15	7/15
		10 "		13/15	5/15	2/15	2/15
	E	20 "		12/15	11/15	9/15	8/15
	γ-globulin	20 "		"	"	7/15	2/15
		Controls		24/30	7/30	3/30	1/30
IV	E	40 × 2	Oral*	9/10	4/10	2/10	2/10
		40 × 1	*	"	5/10	1/10	1/10
		20 × 2	Subcut.*	8/10	4/10	"	"
		10 "	*	10/10	8/10	5/10	5/10
		20 × 10		8/10	6/10	4/10	3/10
		10 "		10/10	8/10	"	"
		2.5 "		"	7/10	5/10	4/10
		Controls		7/10	4/10	0/10	

* Treatment started prior to inoculation.

riod of 60 days. Death followed appearance of paralysis by 3 to 6 days. For experiments on isolated cells, monkey kidney (*Macacca mulatta*) was prepared according to Bodian's technic(2) and suspended in Hank's solution. The medium was changed to Earle's solution when the cells showed a completed sheet (after 5-6 days). The latter solution was used for dilution of the mucoprotein also. Virus growth has been estimated by cytopathological observation. Moreover, the infectivity of the test tube aliquot, after completed cell infection, was measured by transfer to new tissue cultures.

Results. The therapeutic effects observed *in vivo* are presented in Table I. It would seem that there is no dose-effect relationship in the ranges studied and no significant difference between repeated oral and subcutaneous administration of the mucoprotein. Therefore, and in view of the differences among experiments, it is justifiable to evaluate the results in their totality: by adding survival rate at the 30 day observation period (as generally done), administration of 10 doses of preparation E gave 42.5% protection (32 out of 75), of preparation D, 33.3% (10 out of 30), of preparation A (blood group substance) 50% (10 out of 20). Considering all animals treated, regardless of preparation and dose, and including groups treated once or twice only, survival rate amounts to 33.5% (70 out of 205) compared to 5% in the untreated groups (3 out of 60). The difference between total survivors of treated groups in comparison to controls was just as marked at the 60 day observation period: 28.2% (59/205) in treated and 1.7% (1/60) in control groups. In one experiment, 20 mg/kg gammaglobulin administered orally 10 times resulted in a protective effect of the same order as with mucoprotein.

In tissue cultures, toxicity of the mucoprotein (preparation E) for monkey kidney cell was first determined: 200 μg/tube induced slight changes, while no effect was noticed with lower (2-100 μg/tube) doses after 4 days of growth. In the tubes used for toxicity experiments (10-200 μg mucoprotein), inoculated the fifth day with 100 TCD_{50}

(10^{-4}) virus, growth seemed to be slighter than in the controls 2 days after inoculation. However, one day later, virus growth in all 45 tubes was the same as in the controls. In a second experiment, to control more closely the possible retarding effect of mucoprotein on the infection, virus inocula decreasing to the limit of infectiveness (from 10^{-4} to 10^{-8}) were used, with a fixed dose, 100 μg/tube, of mucoprotein; in one series of tubes the mucoprotein was kept in contact with the cells for 24 hours, then washed (before inoculation), in another, it was present for the duration of the experiment. The mucoprotein induced no measurable effect on virus development and infectivity in this experiment including 2 series of 5 replicate tubes for each of the 5 virus dilutions used. In another experiment of the same size, previous exposure of the cells for 4 days to 100 μg mucoprotein or even its continued presence after inoculation did not influence growth and infectivity. Nor did mixing 10^{-4} to 10^{-8} inocula with 100 μg mucoprotein for one hour at room temperature change virus growth. In neutralization test, using mice for test of virulence, up to 2500 μg/ml mucoprotein had no direct effect on the virus: all 50 mice inoculated died at the same time as the controls. (In the same test, the minimal neutralizing dose of gammaglobulin, 625 μg/ml, gave 100% survival.)

Conclusions. It appears that certain mucoproteins exert a protective action against experimental poliomyelitis infection of mice and, interestingly, by oral administration also. This action does not seem to be related to virus neutralization, nor to an interference of virus uptake by the cell, or to virus multiplication and infectiveness, according to the results of tissue culture experiments. Excluding the direct action of mucoprotein on the virus, on its cellular uptake and multiplication, the hypothesis of a "nonspecific" effect, whereby treatment would increase in some manner the nonspecific resistance of the organism, becomes suggestive. The lack of dose-effect relationship is favorable to such an hypothesis, though it is quite possible that the dose range was above the minimal required to elicit protective action. The acid mucoprotein (C) and the B_{12} binding substance (B) appeared to be less effective than the blood group substance (A) in the single experiment in which they were compared. However, our data are not sufficient for quantitative comparison between different types of preparations. Whether the action noted is an integral property of this and perhaps other types of mucoproteins, or related to a single, more simple component or subunit, such as a mucopolysaccharide, requires further investigation.

Summary. Mucoproteins obtained from hog gastric mucosa exert a protective effect in experimental poliomyelitis (Type II) infection of mice, while they have no action on the virus in tissue cultures. The protective action is attributed to some mechanism increasing nonspecific resistance of the host.

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Transfer of Radioiodinated Human Serum Albumin (RISA®) from Cerebrospinal Fluid to Blood Plasma.* (25643)

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There have been many conflicting reports regarding the site of reabsorption of cerebrospinal fluid. Cerebrospinal fluid protein plays a significant role in association with many pathological states of the CNS; however, little is known about the dynamics involved. Behring(1), Sweet(2), and others(3) have described transfer of various ions and non-

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