as those observed with vesicular exanthema of swine virus. Were the correlation only between plaque morphology and type, it would be interesting but not particularly meaningful in a biological sense. However, the fact that differences in plaque morphology of a virus were correlated with extremes of pathogenicity in a natural host lends more than academic interest to the observations. The findings with polio viruses previously mentioned and those with the virus described in this report (*5*) suggest that the correlation of physiological and morphological plaque variations with host pathogenicity may reflect a phenomenon common to other species.

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Absence of Albuminlike Serum Proteins in Turtles

The differentiation of species of turtles of the genus *Pseudemys* by paper electrophoresis of serum proteins was reported recently (*1*). However, no turtle serum proteins comparable in electrophoretic behavior to human serum albumin were observed. Other workers (*2*) have reported marked differences between the electrophoretic patterns of turtles and snakes. A biochemical comparison of the total protein and albumin content of reptilian sera (*3*) revealed notably lower albumin values in turtles than in snakes. However, the particular salting out procedure of that study did not exclude alpha globulins from the albumin fraction.

Table 1. Rat, alligator, and turtle serum proteins (grams per 100 ml of serum). For this study, serum of one male specimen of Holtzman albino rat and of one male specimen of *Alligator mississipiensis* and pooled sera of three specimens of *Chelydra serpentina* were used. The "albumin" fraction from the serum of another specimen of *Chelydra* did give a faint biuret reaction.

Protein	Rat (g/100 ml)	Alligator (g/100 ml)	Turtle (g/100 ml)
Total protein	5.95	5.80	2.20
Albumin plus alpha globulins	3.12	1.80	2.03
Albumin	2.14	0.70	0.00
Alpha globulins	0.98	1.10	2.03
Other globulins	2.83	4.00	0.17

During our studies (*4*) with serum proteins of human beings and lower vertebrates with neoplasia, paper electrophoresis techniques did not reveal albuminlike components in sera of normal representatives of three major families of turtles. Turtle families and species studied were Chelydridae (*Chelydra serpentina*), Dermochelidae (*Dermochelys coriacea*), and Testudinidae (*Clemmys insculpta* and *Testudo gigantea*).

Sera were collected from clotted samples of blood obtained by cardiac puncture. Our paper electrophoresis was done with a Spinco apparatus, at 5 ma constant current for 16 hours. Paper strips were stained with bromphenol blue and were photoscanned and analyzed by means of the Spinco-Analytrol instrument. Specimens of human serum were included in each run. Rat and alligator serum proteins were compared with those of a turtle by a biuret procedure following a modified salt-ether fractionation (*5*) of the blood sera.

The turtle sera examined appear free of a human-like albumin serum protein component, according to electrophoretic analyses (Fig. 1). Ether-salt fractionation and biuret analysis did not consistently reveal albuminlike protein in the serum of *Chelydra* (Table 1).

The findings are provocative from the viewpoints of comparative biochemistry, physiology, and systematics. Albumin synthesis is a function that has long been ascribed to the parenchymal cells of the liver (*6*). Such cells are reported to be structurally cirrhotic-like in the liver of fish, amphibians, and reptiles (*7*). Interestingly enough, paper electrophoresis of the blood serum of Elasmobranchii (*8*) has revealed no component with the mobility of albumin. Correlations between liver histology and protein biochemistry are not available for reptiles. Such studies might be of phylogenetic value. Boyden and Paulsen (*9*) have emphasized the value of electrophoretic studies of serum proteins as a step toward understanding the biochemical evolution of the vertebrates. However, physical chemical criteria, in addition to paper electrophoresis, and protein analyses of greater sensitivity than the biuret reaction are necessary before one can satisfactorily define the nature of the presence or absence of "albumin" in the sera of turtles or other lower vertebrates.

The results reported here suggest an absence of a human-like serum protein with electrophoretic properties of albumin in more than one genus and family of turtles. Similar independent observations of Zweig and Crenshaw (*1*) are supported by our work.

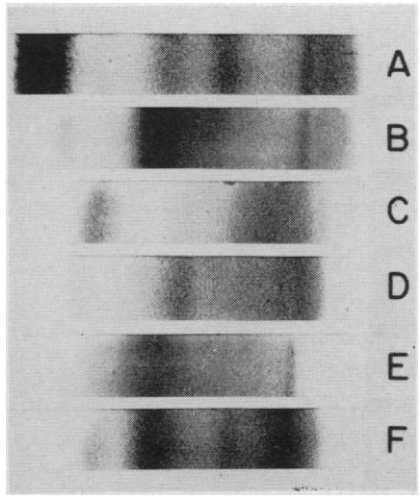

Fig. 1. Paper electrophoresis patterns of serum proteins of turtles and of the serum proteins of a human being (*A*) and an alligator (*B*) for comparison. Other patterns represent the turtle species *Dermochelys coriacea* (*C*), *Clemmys insculpta* (*D*), *Testudo gigantea* (*E*), and *Chelydra serpentina* (*F*).

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Inhibition of Ribonuclease by Polyacids

Heparin and other sulfated polysaccharides have been reported to act as competitive inhibitors of beef pancreas and rodent liver ribonucleases (*1, 2*). Two well-recognized effects of heparin have been reproduced with the polyacids

silicotungstic, phosphotungstic, and phosphomolybdic. These are (i) anticoagulant activity in vitro and in vivo (*3*); (ii) the ability to elicit the appearance of plasma clearing factor activity (*3*). The work discussed in this report demonstrates that these inorganic substances also resemble heparin in their ability to inhibit crystalline beef pancreas ribonuclease. In addition, dextran sulfate (*4*) and two sulfated pectic acid derivatives which possess anticoagulant activity (*5*) were also found to be potent ribonuclease inhibitors.

Ribonuclease activity was determined by the spectrophotometric method of Kunitz (*6*) at *p*H 5.0. Each of the substances was brought to *p*H 5.0 before addition to the test system. The polyacids were commercial products.

The data given in Table 1 demonstrate that all five of the polyacidic substances inhibit ribonuclease activity under these conditions. Relative inhibitory activity seems to fall in the general order: dextran sulfate > sulfated pectic acid amide > sulfated pectic acid methyl ester > silicotungstate > phosphomolybdate. In general, the sulfated polysaccharides were more active than the inorganic polyacids.

Since pepsin has the lowest isoelectric point of any known protein (*7*), a sample was inactivated by alkali treatment (*8*), then adjusted to *p*H 5.0. This material showed very little ribonuclease inhibitory activity. This result is in marked contrast to the inhibition obtained by Vandendriessche (*2*) with polyaspartic acid and indicates that proper spacing of the polyacidic groups is also an important characteristic of this type of ribonuclease inhibitor (*9*).

Table 1. Inhibition of ribonuclease (RNase) by polyacids.

Amt. added (mg/4 ml)	RNA added (mg/4 ml)	Decrease in A_{300} between 1 and 5 min. after mixing	Relative RNase activity (%)
	Silicotungstate		
None	0.9	0.038	100
0.20	0.9	0.002	5
0.10	0.9	0.008	21
0.08	0.9	0.014	37
0.06	0.9	0.021	55
0.04	0.9	0.034	89
0.02	0.9	0.035	92
	Phosphomolybdate		
None	0.9	0.035	100
0.6	0.9	0.035	100
0.8	0.9	0.023	66
0.8	1.2	0.034	97
	Dextran sulfate		
0.2	0.9	0.001	3
0.1	0.9	0.004	11
0.02	0.9	0.012	34
	Sulfated pectic acid methyl ester		
None	1.2	0.036	100
0.10	1.2	0.012	34
0.05	1.2	0.025	71
	Sulfated pectic acid amide		
0.05	1.2	0.020	57
	Inactivated pepsin		
None	1.2	0.042	100
(1.0)	1.2	0.038	90

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Antihypertensive Effects of an Aldosterone Antagonist

A new synthetic steroid was recently described (*1*) that antagonized the renal excretory effects of aldosterone and deoxycorticosterone acetate (DCA) in rats (*2*) and man (*3*): 3-(3-oxo-17β-hydroxy-4-androsten-17α-yl)propionic acid γ-lactone (SC-5233). It was of interest to examine the ability of this compound to prevent and reverse the experimental hypertension produced in rats by DCA.

In the first experiment, two groups of eight 1-month-old Sprague-Dawley rats were implanted with a 20-mg DCA pellet and offered 0.86-percent NaCl as drinking fluid. In addition, one group received SC-5233, 10 mg/kg per day subcutaneously for 19 days, and the other group received the solvent (propylene glycol). Blood pressures (*4*) were taken periodically under standard "double-blind" conditions. No differences between the blood pressures of the two groups were observed after 1, 5, or 10 days of treatment. However, on the 15th and 18th days, the mean pressure of the treated group was 168 mm, while that of the untreated group was 177 and 181; these differences were significant (*5*) at the 5- and 1-percent levels, respectively. Because of these results, investigations are under way to determine whether SC-5233 will prevent the production of adrenal-regeneration hypertension (*6*), which may involve hyperreactivity to aldosterone (*7*).

Additional experiments were performed on metacorticoid hypertensive rats (*8*), which are no longer under the active metabolic influence of DCA (*9*). This form of experimental hypertension resembles essential hypertension in man, particularly with regard to the pharmacology, physiology, and pathology of the two diseases (*10*).

For acute studies, SC-5233 was injected into 12 metacorticoid rats, and the blood pressures were recorded before and 2, 4, and 6 hours after injection. Twelve hypertensive control rats received the solvent. It was found that the compound had a hypotensive action when it was administered by both the oral and parenteral routes (Table 1).

For chronic studies, two groups of six metacorticoid rats drinking saline were observed for 7 days and then injected with SC-5233 or the solvent for 19 days. The total dose of SC-5233 over this period was 800 mg/kg (subcutaneously). Blood pressures were recorded five times during the pretreatment week, 14 times during treatment, and twice after; SC-5233 caused a significant decrease of pressure, which was reversed after the cessation of therapy (Table 2). This experiment was subsequently repeated at a dosage of 280 mg/kg in 14 days, with the same results.

The ability of SC-5233 to block the pressor action of DCA is consistent with its ability to block the renal excretory actions of DCA and aldosterone (*2, 3*). However, its hypotensive action in metacorticoid hypertensive rats can hardly be due to the same process, since such rats are no longer being subjected to DCA overdosage (*9*). Additional evidence is provided by the finding that we have been unable to uncover any activity of the 19-nor derivative of SC-5233 in blocking the pressor action of DCA in saline-fed rats or in lowering the blood pressure of metacorticoid hypertensive rats, in spite of the fact that this deriva-

Table 1. Acute hypotensive action of SC-5233 in metacorticoid rats.

Time of treatment	Blood pressure (mm)	
	SC-5233	Controls
*Experiment A**		
Before injection	190	190
2 hr after injection	169†	194
4 hr after injection	170†	192
6 hr after injection	166†	193
Experiment B‡		
Before injection	188	188
2 hr after injection	166§	194
4 hr after injection	162§	192
6 hr after injection	160§	194

* A, 20 mg/kg subcutaneously (eight rats per group).
† $P < 0.01$ that change in pressure equals that of controls (*5*).
‡ B, 200 mg/kg by gavage (four rats per group).
§ $P < 0.05$ that change in pressure equals that of controls (*5*).