TABLE I.
Recovery of Various Concentrations of Sodium Ferrocyanide Added to Serum and Urine.

Amt $Na_4Fe(CN)_6$ added to serum and urine mg%	No. of single determinations	Mean recovery mg%	Stand. Dev. from mean, mg%
5	14	4.96	± 0.10
10	15	9.96	± 0.28
15	15	14.88	± 0.38
20	13	20.03	± 0.38

stable within 1% transmission for at least one hour after the addition of the ferrous sulfate.

Reagents. 1. *Sodium ferrocyanide.* Recrystallized in 95% ethyl alcohol. Standard solutions of 5, 10, 15, 20 and 25 mg% are prepared from this salt. 2. *Acid mixture.* 35 ml concentrated nitric acid and 40 ml 60% perchloric acid are added to 425 ml distilled water. 3. *Trichloroacetic acid.* 20%. 4. *Ferrous sulfate solution.* $FeSO_4 \cdot 7H_2O$, fine crystal, c.p. is added to saturation in 1N H_2SO_4. One part of this is added to 5 parts of the duponal solution and 7-10 drops of concentrated hydrochloric acid are added per 100 ml of this mixture. If the ferrous sulfate-duponal solution becomes turbid, it should be filtered; the solution remains clear after this filtration. 5. *Duponal solution.* Approximately 25 mg of duponal per liter of distilled water.

Procedure. Two ml of ferrocyanide standard, serum or urine, in proper dilution, are pipetted into a 20 x 150 mm ignition tube and 6 ml of distilled water added and thoroughly mixed with a footed stirring rod. Eight ml of acid mixture are then added with gentle stirring and the mixture allowed to stand 10-30 minutes with occasional stirring. After the addition of 2 ml of 20% trichloroacetic and further stirring, the tube is stoppered and centrifuged at 2500 r.p.m. for 15 minutes. Two ml of water treated in the same way provide a reagent blank. One ml of the ferrous sulfate-duponal solution is added to a 5 ml aliquot of the supernatant solution and the intensity of the blue color is then determined with a Coleman, Jr. spectrophotometer at a wave length of 700 millimicra. The reagent blank is set at 100% transmission.

Results. Recoveries of added sodium ferrocyanide to serum and urine in amounts of 5, 10, 15 and 20 mg%, give mean recoveries of 4.96, 9.96, 14.88 and 20.03 mg% respectively with standard deviations ranging from ±0.10 to ±0.38.

Conclusion. The above described procedure is a satisfactory method for measuring the sodium ferrocyanide concentration of body fluids at concentrations of 5 mg% or more.

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Sickle Cell Anemia, Blood Viscosity and Sodium Tetrathionate.* (17863)

WILLIAM M. MCCORD AND VINCE MOSELEY
From the Department of Chemistry and the Department of Medicine, Medical College of the State of South Carolina, Charleston, S. C.

A decrease in blood viscosity, caused by the intravenous injection of sodium tetrathionate, has been reported by Theis and Freeland(1). This decrease apparently is accompanied by an increase in oxygen satura-

* Aided by a grant from G. D. Searle and Co.

1. Theis, F. F., and Freeland, M. R., *Arch. Surg.*, 1940, v40, 190.

TABLE I.

Patient	Viscosity Difference—Sec. (CO_2—O_2) Before	1-4 hr after	24 hr after	Hemoglobin—g % Before	1-4 hr after	24 hr after	Plasma protein—g % Before	1-4 hr after	24 hr after
				Whole Blood.					
W.P.	81.9	49.7							
T.G.	63.0	61.5							
M.	12.7	5.5	4.8	8.13	6.25	6.66	7.49	7.82	7.47
A.P.	13.0	8.7	10.6	7.77	7.10	8.09	7.26	6.20	6.74
I.G.	13.1	7.3		8.67	8.22		6.23	6.77	
S.	11.1	10.7	4.7	7.01	7.24	6.79	7.25	6.88	7.56
C.	15.3	17.0	11.8	9.56	8.98	9.96	7.46	7.74	6.86
M.G.	11.6	3.5		10.1	10.1		8.42	8.42	
J.	7.4	11.7		7.24	7.46		6.79	7.14	
A.	10.5	9.1	4.0	8.49	7.46	8.09	8.65	8.65	8.44
				Whole Blood Treated *in Vitro*.					
E.M.	3.8	4.1		6.21	6.21				
				Serum					
J.	0	0	0					8.31	7.75
				Serum Treated *in Vitro*.					
L.	0	0							

tion. The pathogenesis of sickle cell anemia, while yet obscure, seems to be associated, according to Pauling, *et al.*(2) with an abnormality in the structure of the hemoglobin molecule. This abnormality is reflected by changes in the shape of the erythrocyte, under reduced oxygen tension, which cause an increase in blood viscosity, thus setting up a vicious cycle, which may terminate in a state of crisis. The purpose of this investigation was to determine whether sodium tetrathionate would break the above mentioned vicious cycle by decreasing the viscosity of the blood of sickle cell anemia patients.

It has been shown(3) that by using an Ostwald type viscosimeter the relative viscosity of blood samples, after exposure to oxygen and carbon dioxide, can be expressed in seconds and that the difference, expressed in seconds, should be a fairly accurate measure of the viscousness of the blood of a patient with sickle cell anemia in crises. Simple measurements of absolute viscosity were deemed unsatisfactory in that changes in protein concentration or water balance, hemolysis or peptization of serum proteins would make the evaluation of results difficult. The use of the "difference value" simply uses the oxygenated blood as a control and thus eliminates, to a great extent, factors other than those pertaining to the erythrocyte itself.

Twelve patients, with definitely proved sickle cell anemia in crisis, or seemingly approaching crisis, were given sodium tetrathionate† by vein. Relative viscosity under carbon dioxide and oxygen, hemoglobin, and serum protein determinations were made, before and after the drug, upon a number of successive days. By reference to Table I, it is apparent that sodium tetrathionate does reduce the viscosity of blood of patients with sickle cell anemia. This reduction in viscosity is not due to hemolysis or destruction of cells, nor to a peptization of the serum proteins as can be seen from the data presented, in that no significant change occurred in hemoglobin or protein concentration.

No theory as to the mode of action of the tetrathionate can yet be formulated. It has no effect upon whole blood or serum *in vitro*. It has no action upon serum when injected intravenously. Seemingly its action is asso-

2. Pauling, L., Itano, H. A., Senger, S. J., and Wells, I. C., *Science*, 1949, v110, 543.

3. McCord, W. M., Kelly, W. H., Switzer, P. K., and Culp, F. B., PROC. SOC. EXP. BIOL. AND MED., 1948, v69, 19.

† Obtained from G. D. Searle and Company as "Tetrathione."

ciated with the metabolic processes of the blood and may modify or affect the structure of the hemoglobin molecule. Pauling, *et al.* (2) used tetrathionate in a buffer system to prevent formation of methemoglobin during electrophoretic analytical procedures and indicates that tetrathionate did not affect the structure of "sickle cell hemoglobin", during these measurements. We propose to attempt to detect changes in the structure of the "sickle cell hemoglobin" in patients after treatment with tetrathionate. Clinical observation in regard to the use of this drug will be reported at a later date.

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Confirmation of Presence of a Gastric Secretory Depressant in Gastric Juice of Humans. (17864)

CHARLES M. BLACKBURN, CHARLES F. CODE, DONALD P. CHANCE, AND EARL E. GAMBILL

From the Divisions of Medicine, Physiology and Surgery, Mayo Clinic, Rochester, Minn.

In a series of papers Brunschwig and his co-workers(1-5) have reported finding a gastric secretory depressant in gastric juice of human beings. The gastric juice tested was secreted in response to subcutaneous injections of histamine after an overnight fast. Samples of the juice or alcoholic precipitates of the juice were injected into dogs with subtotal, wedge, or Heidenhain types of gastric pouches while these were secreting in response to frequently repeated feedings of small amounts of meat. Although inhibition of secretion sometimes occurred during tests of acid gastric juice, it was observed more frequently after the injection of achlorhydric gastric juice and especially when the achlorhydria was associated with pernicious anemia or cancer of the stomach. Using a quantitative method for the determination of inhibition of gastric secretory activity(6), we have confirmed the finding of Brunschwig and co-workers, that intravenous injections of alcoholic precipitates of achlorhydric gastric juice from human beings produce pronounced inhibition of gastric secretion in dogs.

Methods and procedure. Gastric juice was collected from human beings after an overnight fast by means of a Sawyer tube and intermittent suction. Histamine in a dose of 0.01 mg per kilo of body weight was used as the secretory stimulant. Achlorhydric gastric juice was collected from 2 groups of individuals: (1) 5 patients with pernicious anemia and (2) 8 persons who had no abnormality of the hemopoietic system and on whom roentgenographic studies of the stomach and duodenum were negative. Gastric juice containing acid was obtained from 9 individuals on whom tests of the stomach and upper part of the intestine revealed normal function. When the volume of juice obtained was small, samples from individuals in the same group were pooled. Alcoholic precipitates of the juice were prepared according to the method of Brunschwig and co-workers(3) by adding ethyl alcohol to the gastric juice until the concentration was 80%. After standing overnight the clear supernatant fluid was siphoned off, and the precipitate in the residue was collected by centrifugation. The

1. Brunschwig, A., van Prohaska, J., Clarke, T. H., and Kandel, E. V., *J. Clin. Invest.*, 1939, v18, 415.

2. Brunschwig, A., Clarke, T. H., van Prohaska, J., and Schmitz, R. L., *Surg., Gynec., and Obst.*, 1940, v70, 25.

3. Brunschwig, A., Schmitz, R. L., and Rasmussen, R., *J. Nat. Canc. Inst.*, 1941, v1, 481.

4. Brunschwig, A., Clarke, T. H., van Prohaska, J., and Schmitz, R., *Ann. Surg.*, 1941, v113, 41.

5. Brunschwig, A., Rasmussen, R. A., Camp, E. J., and Moe, R., *Surgery*, 1942, v12, 887.

6. Code, C. F., Livermore, G. R., Jr., Ratke, H. V., and Blackburn, C. M., (Abstr.), *Am. J. Physiol.*, 1948, v155, 430.