capacity on blood obtained from a finger prick. It should prove useful where measurements are needed on small animals such as mice or rats or under conditions where venipuncture is difficult or undesirable, as in premature infants.

Summary. The Roughton-Scholander microgasometric procedure has been modified to provide a simple and accurate micromethod for the determination of the O_2 capacity of blood. Only 39.3 cmm are required and an analysis may be completed in 10 to 12 minutes. O_2 capacities obtained by the micromethod agree well with those measured by the Sendroy macromethod, the average difference being ± 0.16 vol %.

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Renal Excretion of Mannitol.*

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The evidence that the clearance of inulin in man is a measure of the rate of glomerular filtration, although necessarily indirect, seems fairly conclusive.[1] Other studies by Smith and his colleagues[2] have suggested that a number of hexitols (including mannitol) are excreted solely by glomerular filtration. Finally, Gilman has reported[3] that the clearance of thiosulfate is also a measure of the filtration rate.

Recently, a patient was observed in this laboratory whose inulin clearance was found to be 144 ml per minute, but whose mannitol clearance, 3 months later, was 94 ml per minute. Since this patient had no demonstrable kidney disorder and since there was no reason to expect any significant change in renal function during the elapsed period, it appeared likely that inulin and mannitol clearances are not always identical. Further observations on this point, therefore, were undertaken and form the basis of this report.

Some support for the belief that the mannitol clearance is less than that of inulin is obtained from published reports. The most extensive series of observations on the inulin clearance in normals is that of Smith and his colleagues[4] who found the average value of 131 ml per minute with a standard deviation of 21.5 ml per minute in 67 males, and an average of 117, with a standard deviation of 15.6, in 21 females. The findings of most other investigators are in line with those of Smith and are summarized in Table I. Also included are previously unpublished data from this laboratory. The average inulin clearance for the entire group of 133 males is 128 ml per minute. In contrast, the average mannitol clearance in 52 males is 116 ml per minute. The difference between the mean mannitol and inulin clearances among the male patients is statistically significant. There is little difference between the mannitol and inulin clearances among the female patients.

There are but few published data on direct comparisons between mannitol and inulin

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1 Smith, H. W., *Physiology of the Kidney*, Oxford University Press, New York, 1937.

2 Smith, W. W., Finkelstein, N., and Smith, H. W., *J. Biol. Chem.*, 1940, **135**, 231.

3 Newman, E. V., Gilman, A., and Philips, F. S., *Bull. Johns Hopkins Hosp.*, 1946, **79**, 229.

4 Smith, H. W., *Lectures on the Kidney*, University of Kansas, Extension Division, 1943.

TABLE I.
Summary of Inulin and Mannitol Clearances in Normal Human Subjects Reported from Various Laboratories. All values are corrected to a surface area of 1.73 sq. m.

	Male			Female		
Author	No. of subjects	Mean clearance ml/min.	Standard deviation	No. of subjects	Mean clearance ml/min.	Standard deviation
			Inulin.			
Smith[4]	67	131	21.5	21	117	15.6
This laboratory*	26	126	17.1	7	111	16.5
Friedman[6,7]	16	129	30.6	5	126	14.0
Bradley[12]	8	114	15.0	6	128	19.5
Foa[8]	7	117	30.8			
Miller[9]	5	115	16.4			
Talbott[10]	4	150	35.8			
Total group	133	128	23.5	39	119	16.4
Standard error of the mean: 2.04.						
			Mannitol.			
This laboratory*	23	111	17.8	2	108	
Bradley[12]	10	141	43.0	23	124	18.9
Newman†[11]	9	103	15.6			
Klopp[13]	6	117	23.8			
Lauson‡[14]	4	107	25.5	1	125	
Total group	52	116	28.6	26	123	24.5
Standard error of the mean: 3.97.						

Standard error of the difference of the means for males: 4.46, t = 2.69. Probability of such a difference occurring by chance, less than 0.007.
* Includes published[5] and unpublished data.
† Estimated from Fig. 4 of the paper by Newman *et al.*[11]
‡ Data derived from patients 17-76 days after recovery from shock.

clearances. The average ratio of mannitol to inulin clearance in 8 observations on 6 patients reported in the original paper on mannitol[2] was 0.99 with a range of 0.96 to 1.03. One of these patients had hypertension, 2 were normal ante-partum females, while the remainder had preeclampsia. Eight comparisons were reported[5] among 7 patients with various degrees of renal functional impairment due to glomerulonephritis. The mean ratio was 0.96, with a range of 0.92 to 1.03. Lauson and his colleagues reported[14] a mean mannitol to inulin clearance ratio of 0.90 among 5 patients suffering from shock. The authors state, however, that they were experiencing minor technical difficulties with the inulin method and that ". . . it appears likely that the average difference of 10% represents a systematic technical error rather than a true difference in clearance." It is to be noted that the majority of the reported comparisons between mannitol and inulin clearances were performed in patients who either did have or probably had some renal abnormality. Three comparisons between the mannitol and creatinine clearances were performed in 2 dogs and are reported in the original paper on mannitol.[2] In one dog, the ratio was 0.98, but both experiments in the other animal yielded a ratio of 0.92.

5 Earle, D. P., Taggart, J. V., and Shannon, J. A., *J. Clin. Invest.*, 1944, **23**, 119.

6 Friedman, M., Selzer, A., and Rosenblum, H., *J. Clin. Invest.*, 1941, **20**, 107.

7 Friedman, M., Selzer, A., Sugarman, J., and Sokolow, M., *Am. J. Med. Sc.*, 1942, **204**, 22.

8 Foa, P. P., Woods, W. W., Peet, M. M., and Foa, N. L., *Arch. Int. Med.*, 1942, **69**, 822.

9 Miller, B. F., Alving, A. S., and Rubin, J., *J. Clin. Invest.*, 1940, **19**, 89.

10 Talbott, J. H., Pecora, L. J., Melville, R. S., and Consolazio, W. V., *J. Clin. Invest.*, 1942, **21**, 107.

11 Newman, E. V., Bordley, J., and Winternitz, J., *Bull. Johns Hopkins Hosp.*, 1944, **75**, 253.

12 Bradley, S. E., personal communication.

13 Klopp, C., Young, N. F., and Taylor, H. C., *J. Clin. Invest.*, 1945, **24**, 117, 189.

14 Lauson, H. D., Bradley, S. E., and Cournand, A., *J. Clin. Invest.*, 1944, **23**, 381.

Methods. All inulin and mannitol† clearances reported in this paper for man were performed in patients with no demonstrable renal abnormalities, and represent the average of at least 3 consecutive periods. Urine was collected by a many-eyed catheter, with bladder rinses with sterile water, followed by an injection of air. Creatinine clearance was used as the standard of comparison in similar studies in the dog.

Chemical. Inulin was measured by Harrison's modification[15] of the Alving method.[16] The urines as well as the plasmas were yeasted so that any possible adsorption of inulin on yeast would not constitute an error in the determination of the inulin clearance. Mannitol was measured in some instances by a modification[5] of Smith's technique[17] and, in all experiments, by the chromotropic acid method of Corcoran and Page.[18] Creatinine was estimated by a modification of the Folin procedure.[19]

Since much of this study was based on simultaneous mannitol and inulin clearances, it was important to investigate any possible interference of one substance with the chemical determination of the other. In these experiments, the concentration of substance run through its specific analysis was in the range utilized in the clearance studies, while the possible interfering substance was added both at the concentration used in the clearances and in excess. Each study was done in duplicate. The results of the examination of the inulin procedure will be presented in detail.

One mg percent and 10 mg % solutions of mannitol reacted with diphenylamine reagent as in the inulin procedure gave readings identical to water treated in the same manner. A 1 mg % standard inulin solution gave the same result when run through the reaction above or in the presence of 1 to 10 mg % mannitol. Five, 10, and 15 mg % inulin standards, yeasted and then run through the diphenylamine reaction, resulted in a mean recovery of 99.5% compared to aqueous non-yeasted solutions, while the mean recovery for a similar series of inulin standards was 99.6% in the presence of 150 mg % mannitol. The mean recovery of 10, 15, and 25 mg % inulin solutions when added to normal plasma was 98.8% and when added to plasma containing 100 mg % mannitol, was 97.6%. The addition of 150 mg % mannitol to normal plasma had no effect on the apparent inulin blank, nor did 150 mg % mannitol solution change the apparent concentration of inulin in the plasma of a patient receiving this material. Finally, the presence or absence of mannitol in urine containing inulin made no difference in the result obtained with the inulin reaction. The urines in these studies were yeasted.

Similarly, the presence of inulin did not alter the analysis of mannitol by the chromotropic acid method, either in aqueous solutions, in plasma, or in urine, the recovery of mannitol in all these circumstances ranging from 99 to 101% whether or not inulin was present. Inulin also did not alter the apparent plasma blank for mannitol. Inulin did not interfere with the analysis of mannitol by the titration method, either in aqueous solutions or after yeasting.

Results. Simultaneous inulin and mannitol clearances were performed as 9 separate experiments in 8 normal subjects. In each instance, the mannitol clearance was less than that of inulin, the mannitol to inulin clearance ratio ranging from 0.79 to 0.96. The mean inulin clearance for the group was 125 ml per minute, the mean mannitol clearance was 109, yielding a mean clearance ratio of 0.87. These data are summarized in Table II. Mannitol was measured by Smith's titration method as well as by the chromotropic acid method

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† The mannitol used in these studies was obtained through the courtesy of Dr. Karl Beyer of Sharp & Dohme, Inc.

15 Harrison, H. E., PROC. SOC. EXP. BIOL. AND MED., 1942, **49**, 111.

16 Alving, A. S., Rubin, J., and Miller, B. F., *J. Biol. Chem.*, 1939, **127**, 609.

17 Goldring, W., and Chasis, H., *Hypertension and Hypertensive Disease*, Commonwealth Press, 1944.

18 Corcoran, A. C., and Page, I. H., *Fed. Proc.*, 1946, **5**, 130.

19 Folin, O., and Wu, H., *J. Biol. Chem.*, 1919, **38**, 81.

TABLE II.
Comparison of Inulin and Mannitol Clearances in Males.
All values represent the average of 3 or more consecutive periods and are corrected to a surface area of 1.73 sq. m.

Patient	Age	Date	Inulin clearance ml/min	Mannitol clearance ml/min	Clearance ratio mannitol/ inulin	Control inulin clearance ml/min	Decrease after mannitol %	Control mannitol clearance ml/min	Decrease after inulin %
Las	46	1/2	142*						
		2/26	141*						
		4/11	139	118	0.85	158	12.0		
Sol	42	4/9	105	101	0.96	124	14.8		
McC	31	4/18	155	140	0.90	134	11.5		
War	34	3/26	138	109	0.79				
		4/15	116	110	0.95	122	6.1		
Alv.	46	4/23	126	112	0.89			122	7.9
Vad	39	4/28	156	123	0.79			134	8.4
		5/15	144*						
Mor	40	3/24	116*						
		4/7	96.5	84.8	0.87				
Sch	48	3/19	96.0	94.0	0.88				
Avg			125	109	0.87	135	11.1		

* Not included in average.

in 2 of the experiments. The results were identical for both methods. In 4 of the studies shown in Table II, control inulin clearances were measured prior to the infusion of mannitol. The inulin clearance was lower after mannitol in each instance, the average depression being 11.1%. The reverse experiment, wherein control mannitol clearances were obtained before the administration of inulin, resulted in smaller and transient depressions in mannitol clearance. The average decreases in mannitol clearance were 7 and 8%, but in each instance the mannitol clearance had almost returned to control values within one hour after the mannitol injection. In several patients (Las, Mor, and Vad, Table II), inulin clearances alone were performed on another date. In each instance, the inulin clearance done separately more closely approximated the inulin than the mannitol value when the two were done simultaneously.

The creatinine clearance is generally believed to be a measure of glomerular filtration rate in the dog. For this reason, mannitol and inulin clearances were compared separately to that of creatinine in 3 unanesthetized female dogs. These data are summarized in Table III. The mean inulin to creatinine clearance ratio was 1.01, while the mannitol to creatinine clearance ratio in the same 3 dogs was 0.88.

Although metabolic alteration of a compound does not preclude its use as a measure of glomerular filtration rate by urine collection techniques, the application of the constant infusion technique[20] requires that there be no metabolic alteration of the substance. If the renal clearance of a substance, as measured by urine collections, is less than the clearance as measured by the infusion technique, it may be concluded that some of the substance is metabolized. The ratios of the clearances of mannitol as measured by the two techniques ranged from 0.75 to 1.00, with an average of 0.88. The mean urine to infusion pump clearance for inulin on 31 subjects yielded a ratio of 1.02.[21] In 3 normal subjects, 80, 85, and 90% of administered mannitol was recovered from the urine collected for 24 hours after the dose. Smith and his co-workers[2] likewise found 81 and 89% recovery of mannitol during the 10½-hour period after the dose, but in similar experiments with inulin, recovered 95 and 97% in the same 2 patients.

Discussion. The mannitol clearance has been found to be consistently lower than the simultaneously measured inulin clearance in 9 patients with no demonstrable renal dis-

20 Earle, D. P., and Berliner, R. W., PROC. SOC. EXP. BIOL. AND MED., 1946, **62**, 262.

21 Unpublished data.

TABLE III.
Comparison of Inulin and Mannitol to Creatinine Clearances in Dogs.
All values represent the average of 3 or more consecutive periods.

	Clearances, ml/min				Ratios	
Dog No.	Creatinine	Inulin	Mannitol		Inulin/creatinine	Mannitol/creatinine
1	52.0	53.0			1.02	
	66.2		59.5			0.90
2	51.5	52.2			1.01	
	39.7		33.9			0.85
3	69.3	69.2			1.00	
	62.4		55.8			0.89
				Avg	1.01	0.88

orders. In addition, the mannitol clearance is less than the simultaneous creatinine clearance in the dog, while the inulin clearance has been consistently equal to the creatinine clearance. Recently, Smith and his co-workers[22] have found that mannitol clearances are less than creatinine in the dog. Corcoran[23] has found an average mannitol/inulin clearance ratio of 0.902 with a standard error of 0.017 for 42 observations in dogs and human beings. The mannitol/creatinine clearance ratio in the dogs averaged 0.872 with a standard error of 0.013, and the average mannitol/thiosulfate clearance ratio in dogs was 0.89. Hoobler[24] observed an average mannitol/thiosulfate clearance ratio of 0.89 with a standard deviation of 0.07 in 52 observations in 14 patients.

It also appears that the administration of mannitol by intravenous infusion results in a small but significant decrease in the inulin clearance and presumably in the glomerular filtration rate. This action is not necessarily due to the mannitol itself, but may be the result of other substances contained in the mannitol preparation and is presumably due to alterations in the glomerular hemodynamics.

The evidence summarized by Smith[1] for the belief that inulin is a measure of glomerular filtration rate in man is very strong. This belief is strengthened by the identity between inulin and creatinine clearances in the dog, it being generally believed that creatinine clearance is a measure of filtration rate in this species. For these reasons, it seems probable that the discrepancies between mannitol and inulin clearances are due to reabsorption of a small proportion of filtered mannitol by the renal tubules.

It appears likely, therefore, that mannitol cannot be used as a precise measure of glomerular filtration rate in man. Since some mannitol undergoes metabolic alteration, it certainly cannot be utilized for the measurement of filtration rate by the infusion pump technique. It should be noted that mannitol is an excellent osmotic diuretic, and that although thiosulfate appears to be a measure of filtration rate, it in itself is an electrolyte and in its excretion must carry with it its equivalent cation. For these reasons, neither mannitol nor thiosulfate is ideal for the measurement of filtration rate during the course of experiments directed toward the study of water or electrolyte excretion. Inulin, therefore, remains the substance of choice for the measurement of glomerular filtration rate in man under the widest variety of circumstances.

Conclusions. 1. The renal clearance of mannitol is frequently less than the simultaneous clearance of inulin in man. 2. The intravenous injection of mannitol depresses the inulin clearance in man. 3. The clearance of inulin is equal to that of creatinine in the dog, but the mannitol clearance is usually less than that of creatinine. 4. Mannitol undergoes metabolic alteration in man, and therefore, cannot be utilized in the infusion pump technique for measuring the rate of glomerular filtration. 5. Inulin appears to be the most generally useful substance for the measurement of glomerular filtration in man.

22 Brode, J., *et al.*, to be published.

23 Corcoran, A. C., personal communication.

24 Hoobler, S. W., personal communication.