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Endogenous Creatinine Clearance and Serum Creatinine in the Clinical Assessment of Kidney Function

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THE clearance of inulin has long been the standard reference procedure for the measurement of glomerular filtration rate. It has not, however, achieved widespread clinical application for several reasons: inulin, being foreign to the body, requires a sustained infusion to achieve a constant plasma level; since measurement periods are short, maximum accuracy of urine collections requires catheterization of the patient, with attendant risk of urinary infection; and finally, the chemical estimation of inulin is tedious.

Although a number of substances have been advocated over the years as substitutes for inulin, none possesses the combined advantages of creatinine. Since creatinine is present naturally in body fluids, no infusions are required in the determinations of its clearance. Because its plasma level is fairly constant (see later) urine collection periods can be prolonged to achieve accuracy, thereby obviating catheterization of the bladder. Lastly, although the measurement of creatinine, especially in serum, requires more care than it is frequently given, it is nevertheless relatively simple.

Until recently, methods for measuring creatinine have been of two types. The first employs the non-specific Jaffe colour reaction, developed directly on deproteinized serum filtrate or diluted urine, and incorporates a small amount of non-creatinine chromogen into its results (total chromogen or T.C. procedure). The second, more laborious procedure measures only creatinine (True procedure) and involves adsorption of creatinine on Lloyd's reagent, its elution and subsequent colour development with the Jaffe reagents. With the advent of automated chemistry, a modification of the first technique has been devised utilizing the Technicon Auto-Analyzer (A.A. procedure) in which dialysis replaces serum deproteinization.

While numerous comparisons have been published in the past between the clearances of inulin and those of T.C. and True creatinine (see Discussion), there is still disagreement as to which creatinine clearance most closely approximates that of inulin. Furthermore, no detailed data have appeared comparing the A.A. creatinine clearance with that of inulin or the other creatinine procedures. With the passage of time we, in agreement with many others, have become convinced of the superiority of the 24-hour creatinine clearance over that determined in shorter time intervals, usually in the morning (see Discussion). Again there are practically no data or comparisons of 24-hour clearances utilizing the three creatinine procedures or, more important, comparing their results with inulin clearances performed in the same patient.

After a detailed study of the three procedures for measuring creatinine incorporating some changes in technique,[1] it was decided to explore the above questions. At the same time, since the measurement of serum creatinine is gaining new prominence in the assessment of kidney disease,[2] opportunity was taken to examine its normal range, sex difference and, in a limited way, its variation during the day.

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PLAN OF STUDY AND METHODS

Studies were carried out on 89 patients all of whom were admitted to our Metabolic-Renal Unit. The group consisted of 49 males and 40 females ranging in age from 14 to 58 years. They were admitted for investigation mainly of kidney disease, hypertension and kidney stones. None had heart failure, hyperglycemia, glycosuria or ketonuria. The 24-hour creatinine clearance was first estimated utilizing the urine passed from 8 a.m. on the first day to 8 a.m. on the second. Blood specimens were drawn in the fasting state at 8 a.m. on both mornings and the average of their serum creatinine concentrations was taken as the "P" value in the denominator of the clearance formula.

Two blood samples were analyzed because the estimation of creatinine in serum is more difficult than that in urine.[1] Creatinine in the sera and urines was measured by all three methods: T.C., True and A.A. During the 24-hour clearance the patients, who were not catheterized, took their usual meals, pursued ordinary hospital activities with avoidance of strenuous exertion and were not subjected to any vigorous diagnostic procedures. At 4 p.m. during each clearance a single sample of blood was drawn for creatinine estimation as a crude test of the constancy during the day of serum creatinine. This time was chosen as the latest which would permit convenient processing of the blood sample by a technician during an ordinary working day.

Immediately after the end of the 24-hour creatinine clearance, inulin clearances were performed in the conventional manner,[3] employing a priming dose and a sustaining infusion of inulin in saline administered by a constant infusion pump. Plasma levels of inulin between 20 and 30 mg. per 100 ml. were almost always attained. For these studies all patients were catheterized. Simultaneously with the inulin clearance and utilizing the same serum and urine samples, creatinine clearances were determined employing the three above-mentioned methods.

The customary formula with surface area correction was used:

$$C = \frac{UV}{P} \times \frac{1.73}{S.A.}$$

where C is the clearance of inulin or creatinine; U and P are the concentrations in urine and plasma (or serum), respectively, of inulin or creatinine expressed as mg. per ml., V is the urine flow rate in ml. per minute; 1.73 is the standard "normal" average surface area expressed in square meters, and S.A. is the actual surface area of the patient derived from a Dubois nomogram utilizing the patient's height and weight.

Inulin was determined by the method of Schreiner[4] and creatinine was estimated in three ways: as T.C.,[5, 6] as True[7] and by the AutoAnalyzer, incorporating minor modifications.[1]

Since earlier publications had indicated enhanced tubular secretion of creatinine in the presence of kidney disease, the patients were, for all comparisons, divided into two groups: those with "normal" inulin clearances equal to or greater than 90 ml. per minute (see Discussion) and those with subnormal values less than 90 ml. per minute. In addition, because recent investigation[8] has shown that tubular secretion of creatinine tends to be increased when there is marked proteinuria, the comparison of creatinine and inulin clearances was also made according to the patients' protein excretion. For this purpose the patients were subdivided into two groups: those excreting less than 2.5 g. of protein per 24 hours and those excreting 2.5 g. or more in 24 hours. The value of 2.5 g. per 24 hours was selected arbitrarily as this and greater amounts of proteinuria are found primarily in glomerular disorders (such as the nephrotic syndrome), while in predominantly tubular diseases (such as pyelonephritis and interstitial nephritis) this rate of protein excretion is seldom exceeded.

RESULTS

Table I shows the results of the inulin clearances in our patients. Fifty-four had clearances equal to or greater than 90 ml. per minute with a mean of 116, while 35 had clearances less than 90 ml. per minute with an average of 44.1.

TABLE I.—INULIN CLEARANCE RESULTS

Clearance of inulin (ml./min.)	*No. of cases*	*Mean (ml./min.)*	*Range (ml./min.)*
≥90	54	116	90.2-150
<90	35	44.1	3.0-89.3

Table II presents the comparisons between the clearances of inulin and creatinine, including the creatinine clearances performed simultaneously with those of inulin (morning) as well as those determined in the 24 hours preceding the inulin clearances. In both cases all three chemical estimations of creatinine were done. It can be seen that, at inulin clearances under 90 ml. per minute, all three morning and all three 24-hour creatinine clearances were significantly greater than those of inulin. The largest differ-

TABLE II.—COMPARISON OF CLEARANCES OF INULIN (C_{in}) AND CREATININE (C_{cr})

	C_{cr} (*morning*)*				C_{cr} (*24-hour*)†			
C_{in} *ml./min.)*	*No. of cases*		$d\blacktriangle$ *(ml./min.)*	*P*‡	*No. of cases*		$d\blacktriangle$ *(ml./min.)*	*P*
≥90	54	$C_{T.C.} > C_{in}$	6.1	<0.001	38	$C_{T.C.} > C_{in}$	2.8	N.S.
	54	$C_{True} > C_{in}$	18.1	<0.001	37	$C_{True} > C_{in}$	11.5	<0.001
	51	$C_{A.A.} > C_{in}$	2.7	N.S.	35	$C_{in} > C_{A.A.}$	1.0	N.S.
<90	35	$C_{T.C.} > C_{in}$	8.3	<0.001	32	$C_{T.C.} > C_{in}$	6.2	<0.001
	35	$C_{True} > C_{in}$	11.8	<0.001	32	$C_{True} > C_{in}$	8.3	<0.001
	35	$C_{A.A.} > C_{in}$	6.7	<0.001	32	$C_{A.A.} > C_{in}$	5.2	<0.005

*Short-period creatinine clearances determined in the morning, simultaneously with the inulin clearances.
†Creatinine clearances determined during the 24-hour period immediately preceding the measurement of the inulin clearances.
$d\blacktriangle$ refers to mean difference in ml. per minute.
‡In this and the succeeding tables P values greater than 0.05 are considered not significant (N.S.).

ences occurred with the True creatinine clearances and the least with those of the A.A. procedure. With inulin clearances equal to or greater than 90 ml. per minute, the True creatinine clearances were again significantly greater than those of inulin and again showed the largest deviations from them. In this range of inulin clearance, however, no significant difference was found between the clearances of inulin and of A.A. creatinine whether performed simultaneously or over the preceding 24 hours. While the T.C. creatinine clearances performed simultaneously were greater than those of inulin, those determined in the preceding 24 hours were not significantly different. When the patients were divided into those excreting 2.5 g. per day or more of urine protein and those excreting less, all the above comparisons between inulin and creatinine clearances held true in both ranges of inulin clearance.

Table III presents the analysis of the ratio of creatinine clearance (True and A.A.) to inulin clearance, categorized according to the levels of urine protein excretion and inulin clearance. If, in scanning the table vertically, one compares the data when the inulin clearance equalled or was greater than 90 ml. per minute to those when it was less, one finds that the ratio C_{cr}/C_{in} of the latter was always greater than that of the former at both levels of protein excretion. Statistically in column (a) when $C_{in} < 90$ ml. per minute, the mean ratio of True creatinine clearance to inulin clearance, 1.34, was significantly greater than the ratio 1.13 when $C_{in} \geq 90$ ml. per minute ($P < 0.001$). Similarly when $C_{in} < 90$ ml. per minute, the mean ratio of A.A. creatinine clearance to inulin clearance, 1.25, was significantly greater than the ratio, 1.01, when $C_{in} \geq 90$ ml. per minute ($P < 0.001$). Also, in column (b), when $C_{in} < 90$ ml. per minute, the mean ratios of creatinine clearance to inulin clearance, 1.61 and 1.53 (True and A.A. respectively), exceeded the corresponding ratios, 1.36 and 1.16, when $C_{in} \geq 90$ ml. per minute.

If one examines Table III horizontally, one observes the relationship between the ratio C_{cr}/C_{in} and the urine protein excretion. Thus, when $C_{in} \geq 90$ ml. per minute, the ratio C_{cr}/C_{in} of those patients excreting less than 2.5 g. per 24 hours of urine protein was significantly lower than that of those patients excreting more protein. Similarly, when $C_{in} < 90$ ml. per minute, the ratio C_{cr}/C_{in} of those patients excreting less than 2.5 g. per 24 hours of urine protein was

TABLE III.—ANALYSIS OF RATIO, C_{cr}/C_{in}, ACCORDING TO URINE PROTEIN EXCRETION AND LEVEL OF INULIN CLEARANCE

		(*a*) *Protein <2.5 g./24 hrs.*			(*b*) *Protein ≥2.5 g./24 hrs.*			
		$\frac{C_{cr}}{C_{in}}$			$\frac{C_{cr}}{C_{in}}$			
C_{in} *(ml./min.)*	C_{cr}	*No.*	*Mean*	*S.D.*	*No.*	*Mean*	*Range**	*P* (*a*) *vs.* (*b*)
≥90	True	46	1.13	0.103	7	1.36	0.98-1.62	<0.001
	A.A.	44	1.01	0.101	6	1.16	0.86-1.41	<0.005
<90	True	28	1.34	0.252	7	1.61	1.29-2.10	<0.025
	A.A.	28	1.25	0.276	7	1.53	1.15-2.12	<0.05

*Because of insufficient numbers, S.D. was not calculated in this group.

TABLE IV.—SERUM CREATININE CONCENTRATION* IN PATIENTS WITH GLOMERULAR FILTRATION RATES EQUAL TO OR GREATER THAN 90 ML. PER MINUTE.

	No. of cases	*Mean creatinine concentration (mg./100 ml.)*	*S.D.*	*Actual range (mg./100 ml.)*	*Mean surface area M^2*	*Mean creatinine concentration ÷ mean surface area*
Males	34	1.04	0.145	0.67 - 1.36	1.83	0.57
Females	26	0.83	0.125	0.57 - 1.17	1.56	0.53

*These values are by the "total chromogen" procedure from bloods drawn in the fasting state.

significantly lower than that of those patients excreting more. These relationships applied to both the True and A.A. creatinine clearances. It is interesting that, as indicated in column (b) of Table III, the highest ratios of C_{cr}/C_{in} are to be found in those patients with inulin clearances of less than 90 ml. per minute and who excrete more than 2.5 g. per 24 hours of urine protein.

Table IV presents the fasting concentration of serum creatinine in males and females with glomerular filtration rates equal to or greater than 90 ml. per minute. The number of patients has been augmented by some, not in the series, who had only 24-hour creatinine clearances done, the results of which fell within this range. The mean serum creatinine of the males was 1.04 mg. per 100 ml. with an observed range of 0.67 to 1.36 mg. per 100 ml., while the mean for the females was 0.83 mg. per 100 ml. with a range of 0.57 to 1.17 mg. per 100 ml. The difference between the mean concentrations of males and females was highly significant ($P < 0.001$). Of note are the findings that the mean height, weight and surface area of the two groups were significantly different, the P value for the difference between the means of each parameter being < 0.001. In Table IV are shown the mean surface areas for the males and females and the ratios obtained when the mean serum creatinine concentration of each group was factored by its mean surface area. It can be seen that, in this way, the sex difference between the creatinine concentrations is practically eliminated.

Table V shows the comparison between the mean fasting (8 a.m.) creatinine concentrations and those of the bloods drawn at 4 p.m. It can be seen that when the inulin clearance equalled or was greater than 90 ml. per minute or when the serum creatinine level equalled or was less than 1.4 mg. per 100 ml., the mean morning serum creatinine value was significantly less than that at 4 p.m. In contrast, when inulin clearance was less than 90 ml. per minute or when the serum creatinine concentration was greater than 1.4 mg. per 100 ml., there was no significant difference between the morning and afternoon values.

TABLE VI.—SERUM NON-CREATININE CHROMOGEN* (N-CC) AT RISING LEVELS OF SERUM CREATININE

Creatinine concentration† (mg./100 ml.)				
Range	*Mean*	*N*	*Mean N-CC (mg./100 ml.)*	*Mean N-CC ÷ mean creatinine concentration %*
≤1.4	0.958	351	0.088	9.2
1.41 - 4.99	2.81	89	0.117	4.2
>5.0	9.58	74	0.309	3.2

*Non-creatinine chromogen was derived from the difference in creatinine concentration values yielded by the "total chromogen" and "true" procedures.
†As measured by the "total chromogen" procedure.

Table VI shows the relationship between the level of serum creatinine and that of the non-creatinine chromogen as measured in the T.C. procedure. Much of the raw data is derived from a companion study.[1] It can be seen that as the serum creatinine concentration rose, so did the absolute value of non-creatinine chromogen. In contrast, however, the percentage of total chromogen made up of non-creatinine chromogen decreased successively as the serum creatinine increased.

TABLE V.—COMPARISON OF MORNING (A.M.) AND AFTERNOON (P.M.) CONCENTRATIONS OF SERUM CREATININE*

C_{in} (ml. per min.)	*No.*		*d† (mg./100 ml.)*	*P*	*Serum creatinine▲ (mg./100 ml.)*	*No.*		*d (mg./100 ml.)*	*P*
≥90	38	A.M.<P.M.	0.087	<0.001	≤1.4	49	A.M.<P.M.	0.092	<0.001
<90	34	A.M.<P.M.	0.035	N.S.	>1.4	23	A.M.=P.M.	0.000	N.S.

*A.M. refers to the average of the two serum concentrations of bloods drawn in the fasting state at 8 a.m. as part of the 24-hour creatinine clearance.
P.M. refers to the creatinine concentration of the blood drawn at 4 p.m. The comparison is made in two different ways: according to whether the inulin clearance was greater or less than 90 ml. per minute and according to whether the fasting A.M. serum creatinine concentration (T.C.) was greater or less than 1.4 mg. per 100 ml.
†d refers to the mean difference in mg. per 100 ml.
▲refers to the A.M. value.

DISCUSSION

In 1935 Shannon[9] showed that exogenously infused creatinine was secreted by human renal tubules. The use of endogenous creatinine clearances was apparently first suggested by Popper and Mandel[10] in 1937 and was made practicable with the development by Miller and Winkler[11] of a chemical procedure capable of measuring accurately the low levels of creatinine present in normal serum. Since then numerous comparisons have been made between the clearance of endogenous creatinine and that of inulin, with confusing and conflicting results. Thus, in normals and using a T.C. creatinine procedure, mean ratio values for C_{cr}/C_{in} of close to 1.0 have been found[6, 12] while others[13, 14] have reported values distinctly lower and still others[15] somewhat higher. In other series of normal subjects in whom True creatinine estimations were employed,[11, 14, 16] mean C_{cr}/C_{in} ratios close to 1.0 were reported. It is difficult to understand how both T.C. and True creatinine clearances can be equivalent to inulin clearances when serum T.C. creatinine averages 9% greater, within the normal range (Table VI), than serum True creatinine while their values in urine are practically identical.[1]

In patients with kidney disease the reports are similarly variable. Thus, employing a T.C. creatinine procedure, some workers[12, 14, 17] reported C_{cr}/C_{in} to be greater than 1.0 while Brod and Sirota[6] reported a mean ratio of about 1.0.

Among the reasons for discrepancies in the published data are: the fact that the ratio C_{cr}/C_{in} differs in health from that in disease, that certain diseases or conditions affect C_{cr}/C_{in} more than others and that different techniques have been employed in the comparisons. The last factor is especially significant inasmuch as some papers have used the T.C. procedure while others have utilized the True creatinine method. Furthermore, even when T.C. creatinine has been measured, different methods have been used which incorporated subsequently proved technical errors.

Two major sources of error in the T.C. creatinine estimation are to be found, first, in the precipitation of serum protein where too concentrated reagents lead to loss of creatinine by adsorption on to the protein precipitate and, second, in failure to recognize that, in serum, colour development after addition of the reagents increases progressively beyond the time when urine and aqueous creatinine solutions have reached a plateau. Therefore, in serum, the exact time at which the absorbance of the coloured solution is measured is critical.[1] Only after these and other technical problems were analyzed[1] was it considered worth while to reexamine the relationship of creatinine clearance to that of inulin.

At this point it is worth commenting on the criteria used for our division of inulin clearances into normal and subnormal. From a review of the literature it is clear that the published normal ranges of inulin clearances are both broad and variable. Indeed it is disappointing how frequently one searches textbooks and journal articles dealing with normal kidney function without finding any mention of the normal *range* of glomerular filtration rate. Perhaps the broadest range of inulin clearance is given by Smith[18] who offers 72 to 176 ml. per minute in males and 81 to 137 in females. Goldring and Chasis[19] reported ranges of 87 to 175 and 85 to 149 ml. per minute in males and females respectively. In contrast, more elevated lower limits are given by Hogeman,[20] who reported a range of 96 to 148 ml. per minute in a group of both sexes, and Reubi[21] who, employing thiosulfate clearance, the value of which he equates with that of inulin, published a range of 93 to 159 ml. per minute in a large but similarly mixed group. All these values are corrected for surface area and represent mean values ±2 S.D. We have compromised and selected, arbitrarily, 90 ml. per minute as the lower limit of normal glomerular filtration rate. It is recognized that there may well be some overlap between the normal and subnormal ranges at this level.

Table II shows that in both the normal and subnormal range of inulin clearance, True creatinine clearance is considerably greater than that of inulin, confirming the renal tubular secretion of creatinine. Since, in both the morning and 24-hour clearances, in the normal as well as in the subnormal ranges of inulin clearance, True creatinine clearance showed the greatest divergence from inulin clearance of all three creatinine procedures, there appears to be nothing to recommend its use in routine clinical studies. An exception is the presence of high blood levels of glucose or ketones which interfere in the Jaffe colour reaction employed in both the T.C. and A.A. methods.[1] In these situations the True procedure is the most reliable. Table II also shows that, in the subnormal range of inulin clearance, all three creatinine clearances exceed that of inulin but that the least divergence occurs with the A.A. method. Presumably the T.C. and A.A. creatinine clearances exceed inulin clearance to a lesser extent than does True creatinine clearance because they include, in their serum measurement, noncreatinine chromogen which, by increasing the

"P" value in the clearance formula, lowers its value. In the normal range of glomerular filtration rate, the A.A. creatinine clearance again shows the least divergence from that of inulin and is, in fact, not significantly different from it. Where many measurements of creatinine clearance have to be made, the A.A. creatinine procedure is the one of choice. If, however, only a few such analyses are done at one time, then automation loses its advantages and the manual T.C. procedure may be substituted, at least in 24-hour creatinine clearances.

In all clearance procedures the major source of error is the measurement of V, the rate of urine flow. This is not only because of errors in the exact timing of urinary bladder emptying but also owing to incomplete emptying of the bladder by the patient. The latter error, which occurs fairly frequently, will obviously cause greater inaccuracy in the smaller urine volume of a shorter clearance period than in the larger volume measured over an entire 24-hour period. Admittedly such collections are feasible only where excellent co-operation on the part of the patient and ward staff can be assured. Also favouring the use of 24-hour creatinine clearances is the observation in Table II that these differ from inulin clearances to a lesser extent than the shorter morning clearances, both in the normal and disease ranges of glomerular filtration rate. Presumably this is because the 24-hour clearance incorporates the diurnal reductions found in glomerular filtration rate[22] and thus reduces the increment of this clearance over that of inulin when both are done only in the morning (Table II).

Berlyne *et al.*,[8] employing the A.A. technique, reported a mean value for C_{cr}/C_{in} of 1.85 in six patients with marked proteinuria and suggested that, in this circumstance, creatinine clearance may not accurately measure glomerular filtration rate. Although five of their six patients had inulin clearances of less than 90 ml. per minute, the authors do not clearly distinguish the factor of reduced kidney function, in increasing C_{cr}/C_{in}, from that of massive proteinuria. Table III shows that reduced glomerular filtration rate and marked proteinuria are independently associated with an increase in C_{cr}/C_{in} and therefore with increased tubular secretion of creatinine. When the 46 patients with inulin clearances $\geq$ 90 ml. per minute and excreting less than 2.5 g. per 24 hours of urine protein were further subdivided into 33 excreting "no" protein ($<$ 0.1 g. per 24 hours) and 13 excreting between 0.1 g. and 2.5 g. per 24 hours, the ratios of C_{cr}/C_{in} in the two groups were found to be identical, 1.13. Yet this ratio is significantly lower than that of patients with the same range of inulin clearance and excreting 2.5 g. per 24 hours or more of urine protein. This suggests that the excretion of some threshold amount of urine protein, perhaps about 2.5 g. per 24 hours, may be necessary before increased tubular secretion of creatinine is manifested. Column (b) of Table III also demonstrates that the greatest increase in C_{cr}/C_{in} occurs when reduced filtration rate and massive proteinuria co-exist. The reasons for this increase are uncertain, although teleologically, in the case of reduced filtration rate, an increase in tubular secretion of creatinine would certainly help rid the body of a non-metabolized waste product. The increased tubular secretion of creatinine associated with marked proteinuria is in line with another tubular disturbance that has been observed in the nephrotic syndrome, namely renal glycosuria.

Thus reduced glomerular filtration rate and marked proteinuria may be grouped with congestive heart failure[6, 17] and normal infancy[6] as conditions in which endogenous creatinine clearance may not accurately reflect inulin clearance. Despite these limitations (which must, however, be taken into account) we feel that the clearance of endogenous creatinine remains the most useful of the simple clinical substitutes for inulin clearance.

Recently[2] the superiority of serum creatinine over blood urea nitrogen (BUN) as a means of following the progress of kidney disease has been emphasized. Thus BUN is much more susceptible than serum creatinine to variations in urine flow rate and to alterations in protein metabolism which might result from changes in dietary protein, from changes in tissue protein breakdown (as occurs with steroid hormone administration or in starvation) or from the presence of blood in the gut. On the other hand serum creatinine is relatively uninfluenced by alterations in protein metabolism and urine flow rate. It may, however, be lowered independently of changes in kidney function by conditions which reduce muscle bulk and therefore creatinine production, such as muscular atrophy or protein malnutrition.

Using a T.C. creatinine procedure, a number of normal ranges for serum creatinine have been reported and, again, differences in technique account for some of the discrepancies. Many of these reports demonstrate a difference between the sexes even though this is not always stressed. Although we did not study normal subjects, our results in patients with inulin clearances equal to or greater than 90 ml. per minute agree with data previously published on normal subjects. Thus our upper limits of normal serum

T.C. creatinine of 1.36 mg. per 100 ml. for males and of 1.17 mg. per 100 ml. for females, are similar to those of Doolan, Alpen and Theil,[23] who reported 1.39 and 1.1 mg. per 100 ml. respectively, and to those of Roberts,[24] who in an excellent survey reported 1.4 and 1.2 mg. per 100 ml. for males and females respectively by the A.A. creatinine procedure. We have recently shown in a large number of comparisons that there is no difference between serum T.C. and A.A. creatinine values below the level of 1.4 mg. per 100 ml.[1] We feel that the normal upper limit and sex difference in serum creatinine should be emphasized because erroneous normal values as high as 1.8 mg. per 100 ml. have been recently reported,[2] and because one still sees, attributed to *serum*, the normal range of the older *whole blood* method, 1 to 2 mg. per 100 ml.

As has been previously reported,[23] and confirmed in Table IV, the sex difference in serum creatinine is markedly reduced if the creatinine concentration is "corrected" by some measure of muscle mass such as body weight or surface area. Presumably if one factored the creatinine concentration by lean body mass, the sex difference in serum creatinine would disappear.

Doolan, Alpen and Theil have reported, without specifying the exact times, that in a group of normal subjects the serum creatinine concentrations were higher in the afternoon than in the morning. Our data (Table V) confirm this for subjects with serum creatinine concentrations and inulin clearances within the normal range: the serum creatinine concentration at 4 p.m. is significantly higher than that of blood drawn in the fasting state at 8 a.m. This may be due to the consumption by our "well" patients of diets relatively high in protein which contain cooked meat, hence pre-formed creatinine, and which may cause a post-prandial rise in serum creatinine.[25] It is not due to any fall in glomerular filtration rate, since this tends to be highest in the early afternoon.[22] Thus, in the performance of 24-hour creatinine clearances as we advocate, bloods for creatinine should be drawn in the fasting state in the early morning, since it is only under these conditions that we have established statistical identity between the clearance results using the A.A. or T.C. creatinine procedures and those of inulin.

In the disease range, however, where inulin clearance is reduced and serum creatinine elevated, Table V shows that there is no difference between the afternoon and morning serum creatinine concentrations. This may be because patients with decompensated kidney disease are frequently given low-protein diets containing less pre-formed creatinine and therefore have less tendency towards post-prandial elevation in serum concentration.[25] Another possibility, for which we have seen no evidence, is that, in the disease state, the creatinine clearance might be higher around 4 p.m. than it is in normal subjects and would thus tend to keep the serum creatinine concentration at that time from rising.

The data in Table VI confirm those of a number of earlier studies which showed that although the absolute value of non-creatinine chromogen increases as the serum creatinine concentration rises in disease, its value as a percentage of the total serum creatinine decreases. Thus changes in non-creatinine chromogen do not detract from the reliability of changes in serum creatinine as a means of following the progress of patients with advanced renal disease.

Where large numbers of serum creatinine concentrations have to be measured, as in a routine hospital laboratory, we feel that the Auto-Analyzer procedure is the one of choice. This is not only because of its speed, reproducibility and convenience but also because it obviates the potential errors in the total chromogen procedure[1] which are accentuated by the pressures of large work loads.

Summary In 89 patients inulin and creatinine clearances were compared, the latter being measured during two time intervals—simultaneously with inulin in short, morning collection periods and also during the preceding 24 hours. Creatinine was measured in both comparisons by three chemical techniques: as total chromogen (T.C.), as creatinine only (True), and by the Auto-Analyzer (A.A.). In 54 patients with normal inulin clearances no significant difference was found between the morning A.A. creatinine and inulin clearances. Both the T.C. and the True morning creatinine clearances were significantly greater. No difference was found between the 24-hour clearances of T.C. and A.A. creatinine and inulin clearances while the 24-hour True creatinine clearance was significantly greater. In 35 patients with subnormal inulin clearances all three creatinine clearances were significantly greater than those of inulin, the A.A. procedure showing the least and the True procedure the greatest differences. Thus, in health and disease, A.A. creatinine clearance approaches inulin clearance most closely while True creatinine clearance shows the greatest divergence. Reduced glomerular filtration and marked proteinuria (> 2.5 g. per 24 hours) were shown to be associated, independently, with an increase in tubular secretion of creatinine.

In subjects with normal inulin clearances, the upper limit of serum creatinine (T.C. or A.A.) in 34 males was 1.36 mg. per 100 ml., while it was 1.17 mg. per 100 ml. in 26 females. The sex difference of 0.2 mg. per 100 ml. is highly significant.

In these subjects the 4 p.m. serum creatinine levels were significantly higher than the 8 a.m. levels while in patients with reduced renal function no difference was found. The AutoAnalyzer procedure for serum creatinine is the method of choice when large numbers of sera must be analyzed.

Résumé Nous avons comparé chez 89 malades, la clearance de l'inuline et de la créatinine, cette dernière étant mesurée durant deux périodes séparées (en même temps que la clearance d'inuline), au moment des périodes de cueillette matinale et aussi pendant les 24 heures précédentes. Le coefficient d'épuration plasmatique de la créatinine a été mesuré par trois techniques chimiques différentes: chromogène total (C.T.), créatinine seule (vraie) et par auto-analyseur (A.A.). Chez 54 malades dont la clearance d'inuline était normale, on n'a guère trouvé de différence entre la clearance de créatinine du matin et celle d'inuline. Les deux clearances (C.T. et clearance vraie du matin) étaient toutes deux nettement plus fortes. On n'a pas constaté de différence entre les chiffres de créatinine des 24 heures de C.T. et par A.A. et la clearance d'inuline. Par contre, la clearance vraie de créatinine des 24 heures était nettement plus élevée. Chez 35 malades dont la clearance d'inuline était subnormale, les clearances de créatinine par les trois méthodes étaient plus fortes que la clearance d'inuline, les différences étant minima par la méthode A.A. et maxima avec la méthode vraie. De sorte que, à l'état normal et à l'état pathologique, la clearance de créatinine A.A. est très voisine de la clearance d'inuline, la plus forte divergence étant trouvée par la méthode vraie. On a démontré que la réduction de la filtration glomérulaire et une protéinurie prononcée (> 2.5 g par 24 heures) coïncidaient, indépendamment, avec une augmentation de la sécrétion tubulaire de créatinine.

Chez les sujets dont la clearance d'inuline était normale, la limite supérieure de la créatinine sérique (C.T. ou A.A.) chez 34 hommes était de 1.36 mg par 100 ml, alors qu'elle était de 1.17 mg par 100 ml chez 26 femmes. La différence de 0.2 mg par 100 ml suivant le sexe est très démonstrative. Chez ces sujets normaux, les valeurs de créatinine sérique étaient notablement plus fortes à 16 heures qu'à 8 heures, aucune différence n'ayant été constatée chez les malades dont la fonction rénale était diminuée. La méthode par auto-analyseur (A.A.) est la méthode idéale pour l'analyse d'un grand nombre d'échantillons.

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