

A FACTOR IN THE ETIOLOGY OF A SUB-BREATHING  
STAMMERER

METABOLISM AS INDICATED BY URINARY CREATINE AND  
CREATININE

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Human behavior is a function of many variables and one of the problems of psychology is to predict as nearly as possible these variables; just in so far as these variables can be predicted and controlled, so far can human behavior be predicted and controlled. Many of these factors, it is well known, form the coenesthetic background of human behavior, that mass of organic stimuli which under normal conditions does not get across the limen of consciousness, but which profoundly influences mental content and human behavior.

Now, as the mass of stimuli which go from organs is vitally connected with the physical health of these organs, and this in turn is closely connected with the chemical bodies present therein, it requires little imagination to appreciate the bond which must unite the science of psychology with that of chemistry—the title “constructive chemistry in the service of medicine” (1) might be changed to “constructive chemistry in the service of interpretation of human behavior.” It was with the purpose of investigating one of these coenesthetic factors and with the hope of finding its bearing on the speech of a particular stammerer, that this investigation was begun.

The medical history of the subject in infancy, reveals little other than a continuous struggle with faulty metabolism—as an eminent pediatricist said of him as a baby, “his body chemistry changes so rapidly that it is difficult to find a food that will agree with him for any length of time.” There is no history of convulsions or spasms; he commenced to talk at an early age, walked at thirteen months, normal dentition, normal birth.

As nearly as can be determined he began stammering, in childhood, shortly after he began to talk very well, certainly by the time he was two and a half years old.

From the beginning the most noteworthy factor in his speech performance was, that there would be periods in which he stammered almost incessantly followed by periods in which he stammered very little. On being taken to the country for the summer at the age of three, the new environment, unusual sights, etc., was accompanied by a great increase in stammering; by the time he had been there a month the stammering had nearly passed by; the entire days were spent in playing quietly in the open. These periods of fluctuation have continued as the subject grew older. At the age of six and one-half he was diagnosed by Prof. Edwin B. Twitmyer as a sub-breathing stammerer. At the time of the beginning of this investigation he was nine years one month of age and weighed 74 pounds. The diet has always been directed most intelligently and fresh air, exercise, etc., have been matters for daily consideration.

As time went on these fluctuations of stammering became closely connected with the body chemistry of the individual and particularly with his powers of elimination. Thus, it was noted, without exception that the times when he stammered very little came after an illness (such as grippe, bilious attack, etc.) in which purgatives (i.e., Sal Hepatic, cascara, castor oil or calomel) had been administered or, when there was no acute attack, but the general condition of excitability and peevishness (accompanied by much stammering) warranted this treatment. There was, then, the distinct indication that the difference in the elimination products ran parallel with the variation in speech performance, which fact might be of particular service in the treatment.

In the kind of elimination product which should be chosen for analysis it was very desirable to choose one that was the least affected by a slight change in diet and one which should serve as some sort of an index of metabolism. The saliva of stammerers has been ably investigated by Starr (2); the blood was hardly practicable and was strongly advised by against Prof.

Edwin B. Twitmyer of the University of Pennsylvania; there remained the urine and feces, and from these the urine was chosen. Now which particular component of the urine should be taken was again influenced by that which should show the least change with slight change of diet, and for that reason creatinine and creatine were chosen.

#### GENERAL DISCUSSION OF CREATININE AND CREATINE

The literature on these subjects is very voluminous and rather contradictory. Creatinine appears normally in all urine, creatine at times. By Folin (9) one of the most fundamental facts concerning their excretion is that the amount excreted in urine per day is independent of the amount of protein taken in food and is wonderfully constant for each individual, though different for different individuals. During times of more intensive metabolism such as fevers (10) "the creatinine output is increased . . . and though increased, still appears to indicate the amount of a certain type of normal endogenous metabolism while creatine possibly indicates the amount of abnormal endogenous protein."

By some authorities (11) the appearance of creatine in the urine is related to (a) carbohydrate deficiency, (b) excess of protein; but as the amount excreted is not influenced by doubling the calorific value intake, it is concluded that whatever the origin it is not in the exogenous protein of the diet—this view that excretion of creatine is not necessarily connected with carbohydrate deficiency finds support and proof by Underhill (12). An attempt has been made to relate the appearance of creatine in urine to acidosis but conclusion has been reached that acidosis per se is no factor (13). MacLeod (14) claims that creatine is in some way concerned in the formative metabolism of the cell, with its general growth and maintenance.

Since, in this investigation, little creatine was found, but for the most part creatinine, we are interested in creatine mainly because of its structural relationship to creatinine (which is the anhydride of creatine) and the possible conversion of the one into the other.

What is the significance of low excretion of creatinine? We find according to Mellanby (15) that diminished excretion is due to—(a) disturbance of general circulation, (b) depressed activity of the liver. Of these two factors he claims that (b) is of far greater importance. Feinblatt (16) fixes the origin and significance of creatinine in the following statement:

because of its uniform rate of elimination even on a low protein diet, and of its relation to creatine, it furnishes a valuable index to the level of tissue metabolism. Of all non-protein nitrogenous products it is the least influenced by dietary factors. It is believed to be derived exclusively from creatine contained in all tissue of which the substance is the anhydride.

Benedict and Osterberg (17) claim also to have definitely established the origin of urinary creatinine from creatine in the metabolism of organisms and claim that the process is very slow requiring days or even weeks for its completion. This view is supported by Shaffer (18) who says that muscles are tissues in which creatinine is formed and that urinary creatinine is formed in muscle from creatine in proportion to the normal endogenous reactions of life. He disagrees with Mellanby that it is formed in the liver.

Behre and Benedict (19) believe that no creatinine exists in the blood or that it exists in traces and that (a) it may be that the kidneys accumulate it or that (b) the kidney does it from some precursor in the blood.

Thus we find that there are many views as to the exact origin, nature and purpose of creatine and creatinine; it is not the purpose of this investigation to try to settle any one of these points. The fact that concerns us most is the undisputed connection of the excretion of creatinine with metabolism for as Mathews (20) says

The amount of creatinine excreted daily by human beings varies with the weight, muscular development, state of health, sex and age, and slightly with creatine and creatinine intake, but is almost or quite independent of protein intake of the body. These facts show that it has a very different significance from urea and that they stand in special relation to the fundamental metabolism of the body.

## NORMAL QUANTITIES OF CREATININE AND CREATINE

Since the quantity of creatinine excreted is so significant, it is necessary to determine what is considered the normal. The amount excreted in twenty-four hours is usually given in terms of "creatinine coefficient." This phrase is interpreted in two ways, one meaning the number of milligrams of creatinine per kilo body weight, the other meaning the number of milligrams of creatinine *nitrogen* per kilo body weight; it is used in the latter sense at all times in this paper and when references are given to other investigators, for the purposes of comparison, the results are given similarly.

Here again we find a great divergence of opinion and results. Much work has been done on the norms for adults but exhaustive studies of individual children over a long consecutive period seem to be lacking. For adults, Amberg and Morrill (21) give 6.71, 7.78, 9.94 and claim that it is dependent upon three factors (*a*) degree of muscular development, (*b*) muscle tonus, (*c*) proportion of adipose tissue. Schulz (22) gives 6.28 to 7.06 for adults; and points out that it is increased by muscular exercise of all kinds, for the time being, though not necessarily for the entire twenty-four-hour period. Shaffer (23) says that it is 8.1 upwards, at age of five or six it is one-half the adult, and accentuates it by the general statement that "children have a creatinine coefficient a great deal below this figure—8.1." Examples are given of isolated twenty-four-hour specimens in which the results are 4.1 to 6.3.

Krause (24) has reported some work on boys and girls but the results for the most part are in terms of milligrams of creatinine, i.e., the weight of the child is not reported. In those cases in which the weight is reported, the coefficient is 3.6 to 5.7. From this array of statistics from able investigators, it appeared that the subject under investigation gave the poorest speech performance when he approximated the "normal coefficient" and the best speech performance when the coefficient was above the norm.

However, Harding and Gaebler (25) have recently (1922) reported a study on children under high protein diet, and state

that children produce per kilo body weight as much creatine and creatinine as an adult man, and at nine to nine and one-half years the coefficient has been found to be as high as 8.89. This statement he reaffirms in his article in August, 1923 (26).

Now this study, though based on a limited number of children and carried out for a limited length of time, was carried out under rigorous conditions of diet. Other reports do not appear to have been made on more than one twenty-four-hour period with a given child and little is stated regarding the diet, other than the presence or absence of meat. These results agree far better with those of the present investigation; that is, we believe that the subject should give a creatinine coefficient of 8 or above, and experience has shown that the more nearly he approximates 8 and maintains it, the better is the speech performance. The diet given in these cases also corresponds to the diet given the subject, i.e., cereal, eggs, bread, butter, milk, potato, vegetables, sugar and oranges, with the exception of meat given five days out of the seven, which would increase urinary creatinine (27).

With regard to the effect of the action of purgative on creatinine excretion, Burns (28) has found results similar to ours, i.e., the administration of a purgative causes increase in urinary creatinine.

The questions which were foremost in the mind of the experimenter were:

1. What difference will be found in urinary creatine and creatinine after purgative?
2. What effect will be the breathing exercises and the speech training given to stammerers have upon this excretion?
3. What general relation will there be found to be between his speech performance and these results?
4. If creatine or creatinine be found low or high on any given day is this loss or gain "made up" the next day or on any successive day?

To this end a preliminary study of creatinine, creatine, acidity, total acidity, specific gravity was made on about 500 samples of urine; these tests extended over three months. In this

study it was thought that the yield of a given product at a given time might be significant and for this reason analyses were made on each separate excretion and results noted. During this period it was often difficult to obtain every specimen of a given day, as the subject would sometimes forget to use the receptacle, or would lose a part of it, etc. For this reason table 1 does not contain successive daily totals although analyses were made every day excepting the two weeks stretch from August 13 to 27. This preliminary study served as an indication as to whether continued investigation might be fruitful and whether the excretions were ever as uniform as they are generally supposed to be (3). The literature abounds with statements that the excretion of creatinine and creatine is wonderfully constant for each individual, though it varies with different individuals. That this was not indicated in this subject can be readily seen in the preliminary study where the value runs from 4.9 to 10.3 per day.

#### SPEECH TRAINING

The exercises for the correction of stammering were given twice daily—at rising 7:00 a.m. and on retiring 8:00 p.m. Great effort was made to secure the coöperation of the subject and the device of timing the number of seconds that each exercise was held was found to be an excellent expedient; then, to mark the improvement as time progressed, stimulated the interest of the subject so that he soon began to look upon the proper performance of his exercises in the nature of a game. Also the matter was put upon a salary basis by the parents, for the subject, which helped give spirit to the work.

#### *Procedure*

The exercises which varied only slightly from the regular procedure of the clinic, and then only in number, not in kind were as follows:

Hands on hips, heels together, head well up before an open window:  
I. Ten long, deep breaths.

TABLE 1  
*Preliminary study of creatinine, creatine, etc., in the urine of a sub-breathing stammerer*

DATE	CREATININE	CREATINE	TOTAL	CREATININE COEFFICIENT	TITRATABLE ACIDITY	SPECIFIC GRAVITY	SOLIDS	VOLUME	REMARKS	SPEECH
	<i>mgm.</i>	<i>mgm.</i>						<i>cc.</i>		
July 3	933	0	933	10.33	305.3	1017	47.615	1075	Had been talking very poorly; cascara two days before; doing better	C
July 5	822	0	822	9.10	431.3	1018	61.542	1315	Cascara again; talking well considering July 4	C+
July 9	486	0	486	5.35	267.1	1017	47.393	1027	Talking has improved steadily has had a thorough purging	B
July 10	551	0	551	6.06	296.2	1015	39.585	1089	Not talking so well today, some company	B-
July 12	415	50	465	4.57+0.605	285.3	1005	17.108	1316	Talking worse	C+
July 13	525	0	525	5.78	280.3	1019	41.516	584		C
July 14	496	0	496	5.46	316.0	1012	32.021	1026		D
July 16	610	0	610	6.71	240.0	1010	31.696	1132		D
July 17	600	0	600	6.60	252.2	1014	40.986	1126		D+
July 19	596	0	596	6.56	474.9	1010	23.036	886		D+
July 20	657	0	657	7.23	711.5	1014	39.239	1078		D
July 24	595	48	643	7.07+0.53	574.6	1015	46.653	1197	Formal training in exercises begun	D
July 25	622	15	637	6.84+0.165	436.6	1017	47.294	1070	Coöperation is good; seems interested in the idea	D
July 26	570	19	589	6.27+0.21	283.9	1020	42.432	816		D
July 27	590	19	609	6.49+0.21	380.9	1012	33.259	1066		D
July 30	660	0	660	7.26	268.2	1015	37.362	958	A very slight improvement in talking is noticed	D+
July 31	674	67	741	7.41+0.737	435.9	1012	47.892	1535		C-
August 2	536	84	620	5.90+0.92	297.6	1013	31.434	930		C
August 3	613	47	660	6.74+0.517	313.8	1014	38.584	1060		C
August 6	615	0	615	6.76	571.1	1012	36.816	1180		C
August 7	624	73	697	6.86+0.80	378.2	1013	32.279	955	Preparing to go away; very excited	C-

August 9	430	0	430	4.73	159.1	1015	21.840	560	Went to shore; very much excited	D
August 13	610	0	610	6.84	246.4	1018	41.184	880	Ocean bathing every day; new surroundings, etc.	E+
August 27	613	0	613	6.97	239.2	1020	47.840	920	Has had one bilious attack; another is following	B
August 29	378	0	378	4.30	137.2	1018	29.714	635	Laxative given; had to be given again; getting a cold	C+
August 30	602	146	748	6.84+1.66	259.9	1017	40.443	915	More laxative necessary. (Sal Hepatica)	B
August 31	665	72	737	7.45+0.82	259.7	1017	33.371	755	Not getting better more laxative	B+
September 3	651	0	651	7.40	323.4	1015	36.660	940	Doctor gave calomel and bismuth on September 1; braces on teeth removed	A
September 5	664	0	664	7.55	231.8	1014	33.852	930	School began; doing very well	A
September 6	665	16	681	7.57+0.18	247.8	1014	32.214	885	Out of doors less but talking very well	A
September 7	594	0	594	6.74	357.1	1013	38.194	1130		A
September 10	595	0	595	6.75	197.1	1015	42.705	1095	Seems a little less calm; effects of school?	A-
September 11	606	0	606	6.79	322.6	1017	48.178	1090	Extra deep breaths and longer exercises begun	A-
September 12	770	0	770	8.75	254.2	1017	45.305	1025		B
September 13	837	0	837	9.51	267.4	1019	47.177	955		B
September 17	704	0	704	8.20	219.2	1016	47.224	1015		B+
September 18	751	53	804	8.54+0.603	243.8	1015	44.850	1150		A
September 19	711	69	780	7.98+0.774	377.5	1017	40.885	925		B
September 21	717	0	717	8.04	346.1	1015	46.995	1205		B+
September 24	743	30	773	8.33+0.34	255.2	1014	42.224	1160	Had been to Zoological Gardens; talked poorly then; better later	B
September 26	722	0	722	8.10	254.6	1018	40.248	860		B+

No determinations were made from September 26 to October 16. The subject performed his exercises, but with little co-operation; was not interested; talking grew worse until on October 5, twenty-five counts of stammering were made in five minutes; this continued until October 18.

TABLE 2  
Detailed study of creatinine, creatine, etc., in the urine of a sub-breathing stammerer

DATE	CREATININE	CREATINE	TOTAL	CREATININE COEFFICIENT	TITRATABLE ACIDITY	SPECIFIC GRAVITY	SOLIDS	VOLUME	REMARKS	SPEECH
	<i>mgm.</i>	<i>mgm.</i>						<i>cc.</i>		
October 12	510	0	510	5.647	194.1	1016	32.032	770	Talking very poorly; 1 cascara (5 grains) no effect; meat in diet	D
October 13	535	0	535	5.924	191.0	1015	29.200	750	Meat in diet; no better	D
October 14	530	0	530	5.869	224.0	1015	31.200	800	No apparent change	D
October 15	545	0	545	6.035	175.7	1016	29.664	720	Special effort begun; 20 deep breaths per day; coöperation secured	D
October 16	660	0	660	7.309	311.5	1016	36.460	885	Restful day, deep breathing; meat in diet; better	C-
October 17	680	0	680	7.531	328.6	1018	36.300	775	Doing better; meat in diet; contracting a cold	C
October 18	671	75	746	7.431+0.830	300.2	1019	43.610	883	Extra deep breaths; doing better; meat in diet	B
October 19	679	23	702	7.52+0.254	300.1	1018	42.585	910	No meat in diet, egg; kept fairly quiet; talking better	B+
October 20	755	0	755	8.306	292.6	1015	46.137	1183	No meat, egg; talking better	B+
October 21	750	0	750	8.306	497.2	1017	49.062	1110	Meat, grade in talking slightly better	B+
October 22	690	71	761	7.64+0.786	392.5	1018	42.926	917	Went to town, but tried to curb his excitement; meat in diet	B+
October 23	698	0	698	7.730	180.6	1021	41.366	740	Played out of doors all day; meat in diet	B+
October 24	600	43	643	6.64+0.476	578.1	1016	48.410	1175	Rainy day; not out much; no meat in diet; coöperation lacking	C

October 25	677	0	677	7.497	446.5	1015	45.820	1175	Meat in diet; slightly better even though company is present	C+
October 26	673	0	673	7.453	371.2	1016	47.792	1160	Meat in diet. Vacation begun until Monday, November 5	C+
October 27	694	0	694	7.686	467.4	1014	49.530	1270	Meat in diet; coöperation is better	B
October 28	691	0	691	7.652	435.7	1025	44.525	685	Meat in diet; company in house; very tired at night; trying hard	B
October 29	750	0	750	8.306	388.1	1020	51.480	990	More nervous excitement; more to be thrown off?	B+
October 30	620	0	620	6.866	413.3	1015	47.970	1230	Meat in diet; talking worse; effect of company?	C
October 31	670	0	670	7.421	345.1	1017	45.160	965	No meat, much excitement; Hallowe'en coming	B
November 1	734	0	734	8.129	454.6	1021	52.416	960	Hallowe'en; meat in diet	B+
November 2	775	0	775	8.583	368.7	1024	55.848	895	Still talking fairly well	B+
November 3	753	0	753	8.339	533.4	1020	58.760	1130		B+
November 4	684	0	684	7.577	468.0	1017	44.400	1000	Sunday; company; not talking quite so well; meat in diet	B
November 5	744	0	744	8.240	349.1	1013	33.272	1265	First day back in school	B+
November 6	754	0	754	8.450	440.8	1017	45.521	1030	Meat in diet; trying hard	B+
November 7	728	0	728	7.962	345.5	1020	49.880	940		B+
November 8	690	0	690	7.642	316.8	1025	57.203	880	Went to dentist; braces removed; not talking quite so well	B
November 9	660	0	660	7.310	357.0	1017	51.932	1175	Speech poorer; 3 grains cascara given	C+
November 10	703	0	703	7.780	488.0	1020		1020	Meat in diet	C
November 11	658	0	658	7.286	157.0	1014	47.32	1300		C
November 12	673	103	776	7.453+1.140	416.4	1014	55.14	1515	Speech improving; doing exercises well	C+
November 13	725	149	874	8.020+1.650	383.3	1019		995		B

TABLE 2—Continued

DATE	CREATININE	CREATININE	TOTAL	CREATININE COEFFICIENT	TITRATABLE ACIDITY	SPECIFIC GRAVITY	SOLIDS	VOLUME	REMARKS	SPEECH
	<i>mgm.</i>	<i>mgm.</i>						<i>cc.</i>		
November 14	684	0	684	7.575	390.2	1021	38.63	715		B+
November 15	730	0	730	8.080	400.7	1020	48.62	935		B+
November 16	612	0	612	6.780	342.4	1018	43.29	925		B+
November 17	604	0	604	6.690	443.2	1014	45.13	1240	Much worse talking; went to children's party on November 16	C C
November 18	581	0	581	6.430	418.9	1017	56.27	1245	Interest lagging in exercises; developing a cold; slight laxative	D
November 19	621	0	621	6.880	417.9	1013	45.86	1360	Laxative had little effect; still has a cold	D
November 20	639	0	649	7.080	378.5	1020	40.196	773	Took exercises very well, should show improvement	D+
November 21	674	0	674	7.460	351.5	1018		910		C
November 22	764	0	764	8.460	437.5	1020		1185	Improving	C+
November 23	700	0	700	7.720	428.9	1020		1150	Doing still better	B

II. "My first exercise is to take a deep breath and exhale it in three puffs"—which was said by subject and performed.

III. "My second exercise is to take a deep breath and say 'ah'"—mouth well opened, position of tongue noted, etc.

IV. "My third exercise is to take a deep breath and say "ē"—mouth extended as in a broad smile, teeth together.

V. "My fourth exercise is to take a deep breath and say "ō"—lips projected fully.

VI. "My fifth exercise is to take a deep breath and say 'ē—āh.'"—

VII. "My sixth exercise is to take a deep breath and say 'ē—āh—ō.'"—

Each exercise was repeated four times, the sentence being repeated as well as the vowel sound. It seemed best as time went on to increase the number of deep breaths, so before each time he repeated an exercise he took three "preparatory" breaths; this enabled him to hold the given syllable a much greater length of time. These exercises were followed by some recitation or slow speaking of difficult sentences, in which great attention was paid to careful enunciation. In the mornings only the ee, ah, oo, exercises were taken. The time given to exercises in the morning was ten to fifteen minutes; in the evening twenty-five to thirty minutes.

#### *The speech scale*

It was realized in the beginning that one of the most important features of this work was to reach some method by means of which one could state what the degree of stammering was on a given day. For this reason the work of outlining some sort of speech scale was begun seven months before the analysis of the urine.

The first idea was to make out a list of sounds upon which he most frequently stammered, make up some literature containing these sounds and get his performance of this task. This yielded no results.

The second plan was to have him read from a simple primer and count the number of times he stammered in a given length of time. The result of this was that he seldom stammered

when reading easy material; the material was made gradually more difficult but no stammering resulted.

The third method was to have him tell a story in his own words, thus more nearly duplicating the conditions of ordinary conversation. This seemed to bring out the stammering very well and was used in remainder of the work. The procedure was simple, the subject was asked to tell a story and the number of stammers made during five minutes was recorded. It was tried for periods of time ranging from five to fifteen minutes but the five-minute interval was believed to be the one which yielded the best results.

For a time careful tabulation was made of the stories chosen to see whether their emotional element would throw any light on the subject but since the stories were found to be those of the usual nine-year repertoire the subject matter was not considered indicative.

There is no doubt in the mind of the experimenter that as time progressed, the story telling of itself became a sort of training, but compared closely with the general daily performance, and with each other, over a limited period of time, it gives an excellent measure of speech performance in this subject. This was quintiled as shown in the table and is believed to be a real measure of speech. This is not given by the investigator as a rigid method of measuring stammering in all subjects or in one subject but is given as a method, which, when quintiled represents a true curve of speech performance.

It would be pleasing if it could be said, that here we have as accurate a measure of the stammering of the subject as we have of the measure of creatine, creatinine, etc.; but we are dealing here with a human equation which is variable in its reaction from hour to hour and even minute to minute. As James says (4): "The desire on the part of men educated in laboratories not to have their physical reasonings mixed up with such incommensurable factors as feelings is very strong"—and the fact that feeling is one of the factors in stammering is one of the reasons that it is so difficult to measure.

In general then the story was told, the count taken, compared with the estimated performance during the day and the grade assigned.

The scale was as follows:  $A = 0-5$ ;  $B = 5-15$ ;  $C = 15-25$ ;  $D = 25-35$ ;  $E = > 35$ .

#### CHEMICAL PROCEDURES

##### *Procedure for determination of creatinine in urine*

*Principle of method.* (Folin procedure.) When picric acid in alkaline solution is treated with creatinine, it is reduced to picramic acid; in the alkaline solution the color is reddish brown. Within certain limits the amount of creatinine is proportional to the depth of the color. The quantity of creatinine is determined by the comparison of the intensity of the color of a solution thus treated, containing an unknown amount of creatinine, with the intensity of color obtained by so treating a solution containing a known amount of creatinine. The comparisons were carried out in a Duboscq colorimeter.

*Solutions used.* Standard creatinine solution containing 0.1 gram pure creatinine dissolved in tenth normal hydrochloric acid solution made up to 100 cc. with tenth normal hydrochloric acid. It therefore contained 1 mgm. of creatinine per cubic centimeter of solution.

*Method.* The urine was preserved by the addition of a few drops of toluene immediately after each excretion. One cubic centimeter of urine was measured by means of an Ostwald pipette into 100 cc. volumetric flask. To another 100 cc. flask was added 1 cc. of the standard creatinine solution. To each flask was added 20 cc. of a saturated solution of picric acid, previously tested (6), then 1.5 cc. of a 10 per cent solution of sodium hydroxide from a burette. These were mixed and allowed to stand for eight minutes, then diluted to the mark with distilled water, and shaken thoroughly. Samples of these solutions were immediately transferred to the cups of the colorimeter; the standard solution was set at twenty mm. and the unknown measured against it, i.e., the depth of the prism in the unknown was regulated, until on looking through the eye piece of the

colorimeter the entire field was the same color. If the reading of the unknown deviated from that of the known by more than five mm. the determination was repeated, using a suitable amount of urine.

All determinations were carried out in triplicate and usually quadruplicate. No more determinations were carried out with one standard than could be conveniently determined within a fifteen minute interval as it was found through experimentation that the standard and the unknown might fade unequally after that time. The known solution, set at a given mark was daily compared against itself to insure accuracy. Fresh standard solutions of creatinine were made up weekly; these solutions are however, remarkably stable, as those standing for a month have been checked up against freshly prepared solutions and still found to check exactly in the colorimeter.

#### *Determination of creatine*

The procedure followed here is the one recommended by Benedict (7). It depends on the quantitative conversion of creatine to creatinine by evaporation to dryness with hydrochloric acid and subsequent treatment of creatinine so formed (and also that originally present) as previously outlined.

*Procedure.* A suitable amount of urine (1 or 2 cc.) was measured out by means of an Ostwald pipette; 10 to 20 cc. of  $\frac{N}{10}$  HCl were added; a pinch of lead to prevent pigmentation was added and the mixture evaporated on the steam bath. After evaporation, the residue was taken up with the minimum amount of hot water and washed, quantitatively, through a funnel containing glass wool; this filtrate was received into a 100 cc. graduated flask.

This solution which now contains the creatine converted into creatinine is treated in the same manner as described under creatinine, with the exception that a 10 per cent solution of sodium hydroxide containing 5 per cent Rochelle salt was used in the place of a 10 per cent solution of sodium hydroxide. The purpose of the Rochelle salt is to prevent a shadow on the colorimeter, due to traces of dissolved lead which might otherwise be present.

The value obtained here represents creatine plus creatinine (in terms of creatinine); from which the value of the original creatinine was subtracted, which left the creatine (in terms of creatinine). These determinations were carried out in triplicate.

*Determination of titratable acidity*

Twenty-five cubic centimeter of urine were taken, 5 grams powdered potassium oxalate added, and titrated with  $\frac{N}{10}$  NaOH using phenolphthalein as an indicator. This figure calculated to the total volume of the urine per day gave the acidity of the urine.

*Total acidity.* (Formal titration.) After the acidity determination 10 cc. of a neutral solution of formaldehyde were added and the solution titrated with  $\frac{N}{10}$  NaOH.

*Specific gravity*

Specific gravity was taken with a hydrometer and calculations made for total solids according to Hawk (8).

*Preliminary study*

Weight of subject under investigation 72 to 74 pounds.

Height 4 feet  $9\frac{1}{2}$  inches.

The first day on which every specimen of a day was obtained was July 3; just previous to that date the subject had been talking poorly—Grade D. Cascara (5 grains) had been given on July 1, talking improved to C; his color was still bad and more cascara was given on July 4; creatinine coefficient became 10.3 on July 3; 9.10 on July 5; by July 7 his speech performance was of B grade, which continued until July 10.

With some slight fluctuations this fact is to be noted throughout the investigation; i.e., after a laxative, creatinine coefficient becomes higher, speech performance improves, creatinine coefficient may stay higher for a few days then go down followed by a decrease in speech performance. When the coefficient has been high, 8 to 9, it takes several days of a lower coefficient (6.5 to 7) before the speech performance goes down to a grade

of D; consistent low coefficient 5.5 to 6 accompanies D performance. Eleven total daily specimens were obtained before the speech training was begun and exclusive of July 3 and 5, which are patently the results of purgative, the average creatinine coefficient was 6.10.

Training for stammering was begun on July 23. The length of time he held the ee, ah, oo, at that time range from 15 to 20 seconds. The average of the first 10 total daily determinations during speech training (extending over a period of two weeks) with no purgative has increased the creatinine coefficient to 7.16. These days, like the preceding ones, were during vacation and spent out of doors. A slight improvement in speech is noted. The apparent effect of the exercises is to increase the creatinine coefficient.

On August 10 he went to the shore and preliminary excitement (which had been continuing for two or three days) throws the creatinine coefficient down to 4.73; speech grade D. No samples could be obtained until August 13 when the subject was talking very poorly, D; coefficient was 6.84; he was in new environment, among strangers, both of which things strongly excited him.

For two weeks then, no samples could be obtained; the subject had become more accustomed to the surroundings, but the unusual hotel fare, candy, etc., had brought about a bilious attack for which a purgative had been given two days before; the coefficient was then 6.97 but had no doubt been preceded by a much higher coefficient, and speech performance was B. He still appeared bilious and was contracting a cold and on August 29 more laxative was given; coefficient had fallen to 4.3; coefficient rose to 8.6 and speech improved.

On returning home, September 1, diarrhea developed and calomel was given, and on September 3, the constant was still high 8.6; speech performance reached a grade of A. This excellent performance continued until September 10, although school began on September 3 and subject was out of doors less. The coefficient became less and by September 10 was followed by decrease in speech performance. On September 11 longer

speech training and extra deep breaths were begun and creatinine coefficient increased; speech performance kept a high level B-A. *This period is by far the best period of speech performance that he has given for a long time. It was preceded by a very thorough cleansing by purgatives and by intensive speech training. It was thought at that time that the stammering was practically cured. A noteworthy fact is, that, during this period, so far as could be determined, there was no conscious effort to talk well.* From September 26 to October 15 it was impossible to make determinations, the exercises were continued but with less coöperation from the subject, he was not interested, impatient and peevish. The talking grew rapidly worse until on October 5 there were 25 stammers in the five minute story period—this grade of performance continued with little fluctuation until October 18.

#### *Detailed study*

On October 12, the detailed study which consisted of making analyses every day for forty-two days was begun, the creatinine coefficient was then 5.6; speech performance grade D. On October 15 far better coöperation was secured, 20 extra deep breaths were taken during the day and coefficient rose to 7 to 8.3 in three days and speech performance rose from D to B; the constant then fell somewhat, to 7.7. Here, there had been some excitement, he had gone to town, there were visitors in the home, and though the coefficient was fairly high (7.5) the speech performance was not so good.

The most important things to determine in this detailed study were (a) whether if creatinine be low on one day it is made up on the next—it was not; (b) whether a performance of a continued degree of excellence accompanied a fairly constant creatinine coefficient, under fairly constant conditions, i.e., lack of excitement. It was found that it did; (c) whether excitement was accompanied by decrease in creatinine and decrease in speech performance—in general excitement produced a decrease, not always apparent immediately in the creatinine because during periods of excitement the metabolism would be more intense, waste products greater, and creatinine coefficient should be higher to throw off the waste material.

## SUMMARY OF RESULTS

Chest measurements: Expansion before, 29 inches; after, 29 $\frac{3}{4}$  inches.

Length of time holding syllables: Before, 15 to 20 seconds; after, 38 to 45 seconds.

1. The urinary creatinine coefficient of the sub-breathing stammerer is lower than in the normal breather, on similar diet.

2. Increase in stammering is preceded by decrease of creatine and creatinine in urine and vice versa.

3. Increase in stammering follows any disturbance in metabolism caused by (a) overeating, (b) injudicious eating, particularly of sugars, (c) nervous situations, (d) fatigue.

4. Deep breathing exercises and speech training, when taken for thirty to forty minutes per day, are followed by increase in creatinine and better speech performance.

5. A weak purgative has little effect either on creatinine excreted or on speech performance—a stronger purgative produces high creatinine followed by better speech performance.

6. The specific gravity, total solids, "titratable acidity," and "total acidity" have little significance in this investigation.

7. The more nearly the creatinine coefficient approaches the normal, and maintains it, the better the speech performance.

8. Lack of zest and interest in performance of exercises is followed by lower creatinine coefficient and decrease in grade of speech performance.

9. Times of excitement are accompanied by less creatinine and more stammering; or if accompanied by the same creatinine coefficient, the stammering is worse as more waste products are being formed and should be thrown off.

## CONCLUSIONS

1. The subject should have a constant creatinine coefficient of about 8 to 8.5 under conditions of lack of excitement, under exciting conditions the coefficient should rise.

2. The creatinine coefficient may be increased in two ways (a) much deep breathing (29) and speech exercises (b) by means of a purging.

3. The metabolism of the body as indicated by the creatinine coefficient is a very strong etiological factor in this sub-breathing stammerer.

4. It is believed that in sub-breathing stammerers, if the urinary creatinine coefficient is low, it may be concluded that an important factor in the etiology of the stammering is faulty metabolism and this must be corrected to effect a cure.

## REFERENCES

- (1) ABEL, J. J., ALSBERG, C. L., BACON, R. F., ELDRÉD, F. R., HUN, R., JOHNSON, T. B., STIEGLITZ, J., TAYLOR, F. O., AND HERTY, C. H.: A report on the future independence and progress of American medicine in the age of chemistry to the Chemistry Foundation, Inc.
- (2) STARR, H. E.: Hydrogen Ion concentration in saliva, etc., as an Index of fatigue and of emotional excitation, and applied to a study of the metabolic etiology of stammering. *Amer. Jour. Psych.*, July, 1922, xxxiii, 394-418.
- (3) SEDGWICK: Constancy of creatinine, etc. *Jour. Amer. Med. Assoc.*, 1910, iv, 1178.
- (4) JAMES, WM: Principles of Psychology. Henry Holt Co., 1890, i, p. 135.
- (5) HAWK: Physiological Chemistry, Folin Procedure. Blakiston and Sons, 6th edition, p. 530.
- (6) FOLIN, O., AND DORSY, E. A.: Impure picric acid as a source of error in creatine and creatinine determinations. *Jour. Biol. Chem.*, 1916-1917 xxxviii, 349.
- (7) BENEDICT, S. R.: *Jour. Biol. Chem.*, 1914, xviii, 191.
- (8) HAWK: *Physiol. Chemistry*, Malfatti, 6th edition, p. 525.
- (9) FOLIN: *Feschrift*. Olaf Hammarsten, iii, Upsula, 1906.
- (10) MYERS AND VOLIRO: Influence of fevers on elimination of creatinine. *Jour. Biol. Chem.*, 1913-1914, 489.
- (11) ROSE, W. C., DIMMIT, J. S., AND BARTLETT, H. L.: *Jour. Biol. Chem.*, 1918, 304, 601-614.
- (12) UNDERHILL, FRANK P.: *Proc. Soc. Exp. Biol. and Med.*, 1916, xiii, 113.
- (13) GAMBLE, J. L., AND GOLDSCHMIDT, S.: A study of creatinuria in infants. *Jour. Biol. Chem.*, November, 1919, xv, no. I.
- (14) MACLEOD, J. J.: *Physiology and Bio-Chemistry in Medicine*. Henry Kimpton & Co., 3rd edition, 1921.
- (15) MELLANBY, EDW.: Creatine and creatinine. *Jour. Physiol.*, February 28, 1908, xxxvi, no. 6.
- (16) FEINBLATT, HENRY: Creatininemia. *Amer. Jour. Med. Sci.*, August, 1923, lxvi, no. 617, no. 2.
- (17) BENEDICT, S. R., AND OSTERBERG, E.: Studies in creatine and creatinine metabolism. *Jour. Biol. Chem.*, May, 1923, lvi, 229-256.
- (18) SHAFFER, P. A.: Origin of creatinine. *Jour. Biol. Chem.*, xviii, 525.
- (19) BEHRE, J. A., AND BENEDICT, S. K.: Studies in creatine and creatinine metabolism. *Jour. Biol. Chem.*, May, 1922, v, 52.

- (20) MATTHEWS: *Physiological Chemistry*, 3rd edition, William Wood & Co., p. 706.
- (21) AMBERG AND MORRILL: *Jour. Biol. Chem.*, 1907, iii, 311.
- (22) SCHULZ, URLHELM: *Der Verlauf der Kreatinin Ausscheidung im Harn des Menschen. Archive für die Gesamte Physiologie*, 1921, clxxxvi, 126-172.
- (23) SHAFFER, P.: *Quart. Jour. Exp. Phys.*, vii, no. 1, 1906.
- (24) KRAUSE, R. A.: Age and metabolism and significance of excretion of creatine. *Jour. Exp. Phys.*, vii, no. 1.
- (25) HARDING, V. J., AND GAEBLER, O. H.: On the constancy of the creatine and creatinine excretion in children on a high protein diet. *Jour. Biol. Chem.*, 1922, liv, 579.
- (26) HARDING, V. J., AND GAEBLER, O. H.: Influence of the positive nitrogen balance upon creatinuria during growth. *Jour. Biol. Chem.*, August, 1923.
- (27) BURNS, D., AND ORR, J. B.: Influence of flesh feeding on urinary creatinine. *Jour. Biol. Chem.*, October, 1916, x, no. 3.
- (28) BURNS, D.: A note on the effect of purgation on creatinine content of urine. *Jour. Biol. Chemistry*, April, 1920, xiv, no. 2.
- (29) HOWELL: *Physiological Chemistry*.  
CANNON: *Basal Metabolism. Laboratory Manual of Physiology*.