

Biometry Research Unit, Indian Statistical Institute, Calcutta

## Indices of Blood Biochemistry in Relation to Age, Height, and Weight

By B. C. DAS

Clinical tests of blood biochemistry are used to evaluate the *milieu interne* for the diagnosis of pathological and abnormal conditions. As used by *Claude Bernard* (1813-1878), *milieu interne* refers to the blood and lymph bathing the cells of the body. It is concerned with respiration, nutrition, excretion, maintenance of water content, temperature regulation, protection, regulation by hormones, and other functions of and related to the body cells and tissues. The clinical tests are designed to assess the effectiveness with which these functions are being carried out. Collection of data using these tests under conditions of normal physiological functioning provides the standards against which abnormal and pathological conditions can be identified and diagnosed.

It has been recognized that changes in the blood biochemistry may be associated with the growth and aging of the organism. Hematological and blood chemistry observations during growth have been reported for children from four to twelve years of age by *Macy and Kelly* (1957). *Geschickter* (1959) has pointed out that the regressive changes in the tissues that occur with aging must be associated with changes of the blood and blood supply. For several clinical indices of blood biochemistry, changes with age have been observed in European and North American populations, e.g., for inorganic phosphorus, hemoglobin, leukocytes and lymphocytes (*Goodale*, 1955). The nature of the changes with age has not been clearly indicated, however, neither expected values at different ages nor statistical measures of relationship having been reported.

Previous research from this laboratory has shown that serum globulin fractions and total serum globulin increase linearly with age and decrease as weight increases, that serum albumin is not related to age or weight, and that while serum lipoprotein increases with age, it is more highly correlated with weight (*Das and Bhattacharya*, 1961). The relationships of systolic and diastolic pressure, and of cholesterol ester and free cholesterol, to age and weight have also been reported (*Das*, 1959, 1960, 1961; *Das and Mukherjee*, 1963). These results were obtained for a population in which age and weight are not significantly correlated. The present research was carried out to explore the relationships of clinical indices of blood biochemistry to age, height, and weight in a normal population. It was also sought to determine whether the ranges accepted in Europe and North America for these indices are appropriate for diagnostic purposes in India.

### Methods

Three hundred fifty-eight male employees of the Indian Statistical Institute, Calcutta, India, served as subjects for this experiment. All subjects were residents of West Bengal, India, were engaged in office work, and were nonvegetarians whose main source of carbohydrate was rice. They were selected by drawing random samples, within age groups, of the employees of the Indian Statistical Institute. No cases of medically diagnosed pathological conditions, including hypertension, occurred in the sample.

The following routine was adopted for collection of the experimental data. Subjects came to the laboratory in the morning in a fasting condition (no meal since the previous evening), and venous blood was removed for the clinical tests. The venous blood was always taken between 9.30 a.m. and 10.30 a.m. and was immediately processed for the different estimations. A light meal was given to the subjects and two hours later they returned to the laboratory where they sat for half an hour before blood pressure was measured. Then, age and customary diet were recorded, pulse and blood pressure determinations made (*American Heart Association*, 1951) and height and weight recorded. Height was measured with an anthropometer (G.P.M., Switzerland) and weight was measured on a Salter Scale (Grosvenor, England) which had been standardized.

Measurements on 28 variables were obtained for each subject. The variables have been assigned identifying numbers for the present report: age in years, height in centimeters, and weight in kilograms being numbers 1, 2, and 3 respectively. Numbers 4 to 28 are clinical indices, and their numbers and names are given in columns (1) and (2) of table I. For each clinical index, table I gives the method of determination in column (3), the material analyzed in column (4), and the unit of measurement in column (5). The references followed were *Tarnoky* (1958) for variable 4; *Hauk et al.* (1954) for variables 5, 7, and 9; *Varley* (1962) for variables 6 and 11; *Gradwohl* (1956) for variable 8; and *Hepner* (1955) for variable 10 and variables 12 to 25 inclusive.

### Results

For the entire sample of 358 subjects, age ranged from 17 to 83 years, with a mean of 32.69 years and a standard deviation of 13.14

Table I  
Laboratory Methods and Average Values for Clinical Indices of Blood Biochemistry

Number (1)	Variable Name (2)	Method (3)	Material analyzed (4)	Unit of measurement (5)	Mean (6)	Standard deviation (7)	Index Normal range (8)	Assessed range for Europe and North America (9)
4	Glucose	Astor & King	blood	mg/100 ml	76.19	21.56	41-112	65-95
5	Chlorides	Whitehorn	blood	mg/100 ml	511.48	72.14	392-631	570-620
6	Urea	Urease Neutralization	blood	mg/100 ml	23.86	5.96	14-34	15-40
7	Creatinine	Folin	blood	mg/100 ml	1.69	0.37	1.1-2.3	1-2
8	Calcium	Coulson & Hernandez	serum	mg/100 ml	9.50	1.43	7-12	9-11
9	Uric acid	Brown	blood	mg/100 ml	3.75	0.91	2.2-5.3	2.2-5.5
10	Inorganic phosphorus	Comori	serum	mg/100 ml	3.48	0.68	2.4-4.6	2.5-4.0
11	Thyroxin-uribidity	Maclean	serum	Unit	2.34	1.41	0-5	0-5
12	Bilirubin	Malloy & Evelyn	serum	mg/100 ml	0.69	0.34	0.1-1.3	0.2-0.8
13	Total cholesterol	Bloor & Knudson	serum	mg/100 ml	180.82	40.26	114-247	150-250
14	Cholesterol ester	Bloor & Knudson	serum	mg/100 ml	115.55	26.40	72-159	105-189
15	Free cholesterol	(calculation)	serum	mg/100 ml	65.58	32.56	12-119	45-61
16	Hemoglobin	Sahl-Hellige	blood	mg/100 ml	14.74	1.43	12-17	14-18
17	Mean corpuscular hemoglobin	(nomogram)	blood	micrograms	30.00	2.78	25-35	27-32
18	Mean corpuscular hemoglobin concentration	(nomogram)	blood	percentage	30.48	2.65	26-35	32-36
19	Erythrocytes	erythrocyte count	blood	million/c mm	4.88	0.57	3.9-5.8	4.5-6.0
20	Leukocytes	leukocyte count	blood	number/c mm	6 751.06	1 882.80	3 600-9 900	5 000-10 000
21	Neutrophils	differential count	blood	percentage	56.78	9.03	42-72	50-70
22	Lymphocytes	differential count	blood	percentage	33.76	8.56	20-48	20-40
23	Eosinophils	differential count	blood	percentage	8.88	6.99	0-20	1-3
24	Sedimentation rate	Wintrobe & Landsberg	blood	mm/hour	13.85	11.76	0-33	0-9
25	Volume of packed erythrocytes	Wintrobe	blood	percentage	47.80	5.05	39-56	40-54
26	Pulse rate (right)	standard procedure	-	pulsations/min	76.99	10.88	59-95	47-75
27	Systolic pressure (right)	auscultatory	-	mm Hg	118.01	15.18	93-143	96-155
28	Diastolic pressure (right)	auscultatory (5th phase)	-	mm Hg	74.81	10.58	57-92	57-101

years. Height ranged from 134.60 cm to 184.20 cm; the mean height was 166.03 cm, with a standard deviation of 6.94 cm. Weight ranged from 36.29 kg to 97.07 kg; the mean weight was 55.57 kg, with a standard deviation of 10.06 kg. Means and standard deviations for the clinical indices are given in columns (6) and (7) of table I. To define the limits of the normal values of the clinical indices, customarily taken as 90% of the individuals, normal ranges have been computed (*Bancroft*, 1957). The product of the normal deviate 1.65 and the standard deviation has been subtracted from the mean to obtain the lower limit of the normal range, and has been added to the mean to obtain the upper limit of the normal range. The normal ranges for these data are given in column (8) of table I; accepted ranges for Europe and North America are given in column (9) for comparison (*Altman, Dittmer and Grebe*, 1959; *Goodale*, 1955; *Gradwohl*, 1956; *Hawk et al.*, 1954; *Hepler*, 1955; *Tárnoky*, 1958; *Varley*, 1962).

To determine the regression of the clinical indices, taken singly, on age, height, and weight, coefficients of correlation were computed (*Walker and Lev*, 1953). For these data, age was correlated  $-0.070$  with height and  $+0.140$  with weight. These two correlations do not differ significantly from zero. Height and weight were correlated  $+0.467$  ( $P < 0.01$ ). The correlations of each clinical index with age, height, and weight are given in columns (3), (4) and (5) of table II. To partial out the effect of the intercorrelations between age, height and weight on their correlations with the clinical indices, multiple regression analysis was carried out (*Walker and Lev*, 1953). The standard partial regression coefficients, which are pure numbers (independent of the scale of measurement), giving the regression of each clinical index on age, height, and weight are given in columns (6), (7) and (8) of table II. The significance of each regression coefficient has also been indicated. Multiple regression equations for the prediction of clinical indices, given age, height, and weight, are presented in columns (9) to (12) of table II. The constant for each equation is given in column (9), and the regression coefficients, expressed in the units of measurement specified in column (5) of table I, are given in column (10) for age, (11) for height, and (12) for weight. The correlation between the expected value of the clinical index, as estimated by the multiple regression equation, and the observed value, is reported as a multiple correlation coefficient in column (13) of table II for each of the clinical indices. The

Table II  
Multiple Regression Analysis of Age, Height, and Weight as Predictors of Clinical Indices of Blood Biochemistry

Number	Variable	Name	Correlation coefficients		Standard partial regression coefficients		Contact	Regression equation		Multiple correlation coefficient			
			Age (5)	Height (4)	Age (6)	Height (7)		Age (10)	Height (11)		Weights (12)	Weights constant (13)	
4	Glucose		0.211	-0.075	0.101	0.184**	-0.124	0.133	114.695	0.301	-0.387	0.286	0.248**
5	Chlorides		0.381	-0.085	-0.028	0.389**	-0.025	-0.071	512.432	2.136	-0.256	-0.509	0.390**
6	Urea		0.129	-0.054	-0.015	0.128	-0.038	-0.015	27.858	0.058	-0.033	-0.069	0.137
7	Creatinine		0.050	-0.124	0.057	0.177	-0.190**	0.143	3.048	0.0005	-0.010	0.005	0.180**
8	Calcium		0.329	-0.031	0.037	0.330**	-0.005	-0.007	8.544	0.036	-0.001	-0.001	0.329**
9	Uric acid		0.022	0.016	0.082	0.005	-0.034	0.107	3.937	0.0003	-0.004	0.010	0.097
10	Inorganic phosphorus		0.017	-0.065	-0.082	0.025	-0.030	-0.072	4.201	0.001	-0.003	-0.005	0.091
11	Thymol-turbidity		-0.096	0.067	0.148	-0.122	-0.024	0.176*	2.198	-0.013	-0.005	0.025	0.190**
12	Bilirubin		-0.099	-0.007	-0.019	-0.100	-0.015	0.002	0.898	-0.003	-0.001	0.000	0.100
13	Total cholesterol		0.154	0.039	0.315	0.095	-0.122	0.359**	208.908	0.292	-0.707	1.435	0.351**
14	Cholesterol ester		0.043	0.032	0.179	0.009	-0.064	0.208*	125.350	0.019	-0.246	0.547	0.188**
15	Free cholesterol		0.147	0.043	0.266	0.100	-0.087	0.292**	72.359	0.248	-0.406	0.946	0.298**
16	Hemoglobin		-0.061	0.022	0.046	-0.070	-0.012	0.061	14.900	-0.008	-0.002	0.009	0.083
17	Mean corpuscular hemoglobin		0.044	-0.034	0.031	0.033	-0.056	0.053	32.717	0.007	-0.023	0.015	0.071
18	Mean corpuscular hemoglobin concentration		0.079	0.011	0.048	0.075	-0.002	0.089	29.544	0.015	-0.001	0.010	0.087
19	Erythrocytes		-0.220	0.052	0.055	-0.233**	-0.007	0.091	5.014	-0.010	-0.001	0.005	0.237**
20	Leukocytes		-0.032	-0.202	-0.066	-0.055	-0.228**	0.048	16 800.180	-7.843	-62.009	9.045	0.211**
21	Neutrophils		0.254	-0.022	0.020	0.257**	0.004	-0.018	50.966	0.176	0.006	-0.016	0.235**
22	Lymphocytes		-0.151	0.078	0.013	-0.147*	0.067	0.002	23.128	-0.095	0.082	-0.002	0.165*
23	Eosinophils		-0.174	-0.064	-0.066	-0.178*	-0.073	-0.007	24.507	-0.095	-0.074	-0.005	0.190**
24	Sedimentation rate		0.291	-0.044	0.051	0.284**	-0.037	0.029	14.232	0.254	-0.064	0.034	0.293**
25	Volume of packed erythrocytes		-0.274	-0.003	-0.051	-0.223**	-0.012	-0.014	52.437	-0.086	-0.009	-0.007	0.225**
26	Pulse rate (right)		0.148	-0.049	0.007	0.144*	-0.042	0.006	83.619	0.119	-0.066	0.007	0.153*
27	Systolic pressure (right)		0.428	0.038	0.367	0.369**	-0.107	0.365**	112.206	0.427	-0.233	0.551	0.537**
28	Diastolic pressure (right)		0.414	0.050	0.320	0.368**	-0.063	0.298**	63.784	0.296	-0.097	0.314	0.494**

\* P &lt; 0.05

\*\* P &lt; 0.01

Table III  
Observed and Expected Age-Group Means for Physical Variables and Significant Clinical Indices of Blood Biochemistry

Number (1)	Variable (2)	20-29 years Age group (3)		30-39 years Age group (4)		40-49 years Age group (5)		50-59 years Age group (6)		60-69 years Age group (7)		70-79 years Age group (8)		80-89 years Age group (9)		90-99 years Age group (10)	
		Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected	Observed	Expected
1	Age	24.11	-	33.26	-	43.83	-	55.25	-	65.50	-	75.25	-	85.00	-	95.00	-
2	Height	166.51	-	165.67	-	164.74	-	164.45	-	164.04	-	164.45	-	164.04	-	164.45	-
3	Weight	54.58	-	57.49	-	57.78	-	59.58	-	54.24	-	59.58	-	54.24	-	59.58	-
4	Glucose	74.92	73.08	69.51	77.05	78.85	80.68	90.29	84.74	79.82	85.09	90.29	84.74	79.82	85.09	90.29	84.74
5	Chlorides	492.83	495.65	505.90	511.82	532.58	534.49	565.63	558.04	558.04	577.98	565.63	558.04	558.04	577.98	565.63	558.04
7	Creatinine	1.65	1.69	1.79	1.70	1.76	1.72	1.65	1.74	1.63	1.70	1.65	1.74	1.63	1.70	1.65	1.74
8	Calcium	9.16	9.19	9.46	9.52	10.23	9.90	10.44	10.31	10.66	10.61	10.44	10.31	10.66	10.61	10.44	10.31
11	Thymol-turbidity	2.51	2.42	2.27	2.38	2.24	2.23	2.19	2.15	1.63	1.90	2.19	2.15	1.63	1.90	2.19	2.15
13	Total cholesterol	175.61	176.27	185.30	183.99	198.81	188.15	192.04	194.28	175.28	187.90	192.04	194.28	175.28	187.90	192.04	194.28
14	Cholesterol ester	114.72	114.62	115.81	116.70	119.97	117.29	116.57	118.56	114.48	115.41	116.57	118.56	114.48	115.41	116.57	118.56
15	Free cholesterol	61.55	62.13	69.49	67.68	78.86	70.95	75.46	75.60	69.80	71.95	75.46	75.60	69.80	71.95	75.46	75.60
19	Erythrocytes	4.94	4.96	4.90	4.88	4.81	4.78	4.74	4.67	4.59	4.56	4.74	4.67	4.59	4.56	4.74	4.67
20	Leukocytes	6 554.85	6 777.79	6 959.62	6 786.24	6 794.88	6 763.63	6 891.66	6 708.32	5 850.00	6 496.72	6 891.66	6 708.32	5 850.00	6 496.72	6 891.66	6 708.32
21	Neutrophils	55.22	55.29	57.19	56.85	58.83	58.70	61.62	60.69	60.00	62.24	61.62	60.69	60.00	62.24	61.62	60.69
22	Lymphocytes	34.50	34.62	33.61	33.68	33.52	32.60	29.62	31.49	32.95	30.82	29.62	31.49	32.95	30.82	29.62	31.49
23	Eosinophils	9.59	9.66	8.67	8.84	6.71	7.91	7.79	6.84	6.84	5.96	7.79	6.84	6.84	5.96	7.79	6.84
24	Sedimentation rate	10.63	11.60	17.46	14.09	18.50	15.20	17.62	19.83	19.14	21.65	17.62	19.83	19.14	21.65	17.62	19.83
25	Volume of packed erythrocytes	48.73	48.54	46.13	47.74	47.17	46.84	46.74	45.85	44.50	45.17	46.74	45.85	44.50	45.17	46.74	45.85
26	Pulse rate (right)	75.75	75.93	79.11	77.10	78.45	78.42	79.00	79.82	78.86	80.66	79.00	79.82	78.86	80.66	79.00	79.82
27	Systolic pressure (right)	115.00	113.58	115.20	119.40	120.33	124.29	131.67	130.22	134.71	130.43	131.67	130.22	134.71	130.43	131.67	130.22
28	Diastolic pressure (right)	72.29	71.85	71.65	73.62	79.62	76.89	83.39	82.90	85.60	83.52	83.39	82.90	85.60	83.52	83.39	82.90

Table IV

Expected Values of Significant Clinical Indices According to Age, Height, and Weight: Chemical Variables

Variable Number	Name	Height (cm) Weight (kg) Age (years)	160				170			
			(3)	(4)	(5)	(6)	(7)	(8)	(9)	(10)
4	Glucose	25	73.90	75.33	76.76	78.19	70.03	71.46	72.89	74.32
		35	76.91	78.34	79.77	81.20	73.05	74.47	75.90	77.33
		45	79.93	81.36	82.78	84.21	76.06	77.49	78.92	80.35
		55	82.94	84.37	85.80	87.23	79.07	80.50	81.92	83.36
5	Chlorides	25	500.71	498.17	495.62	493.08	498.16	495.61	493.06	490.52
		35	522.08	519.53	516.98	514.44	519.52	516.97	514.43	511.88
		45	543.44	540.89	538.35	535.80	540.88	538.33	535.79	533.24
		55	564.80	562.25	559.71	557.16	562.24	559.69	557.15	554.60
7	Creatinine	25	1.70	1.73	1.76	1.78	1.60	1.63	1.66	1.68
		35	1.71	1.74	1.76	1.79	1.61	1.64	1.66	1.69
		45	1.71	1.74	1.77	1.79	1.61	1.64	1.67	1.69
		55	1.72	1.74	1.77	1.80	1.62	1.64	1.67	1.70
8	Calcium	25	9.24	9.23	9.23	9.22	9.23	9.22	9.22	9.21
		35	9.60	9.59	9.59	9.58	9.59	9.58	9.58	9.57
		45	9.96	9.95	9.95	9.94	9.95	9.94	9.94	9.93
		55	10.31	10.31	10.30	10.30	10.30	10.30	10.29	10.29
11	Thymol-turbidity	25	2.27	2.39	2.51	2.64	2.22	2.34	2.47	2.59
		35	2.14	2.26	2.38	2.51	2.09	2.21	2.33	2.46
		45	2.00	2.13	2.25	2.37	1.96	2.08	2.20	2.33
		55	1.87	2.00	2.12	2.24	1.82	1.95	2.07	2.19
14	Cholesterol ester	25	112.48	115.21	117.95	120.68	110.02	112.75	115.49	118.22
		35	112.66	115.40	118.14	120.87	110.21	112.94	115.68	118.41
		45	112.85	115.59	118.33	121.06	110.40	113.13	115.87	118.60
		55	113.04	115.78	118.52	121.25	110.59	113.32	116.06	118.79
15	Free cholesterol	25	58.48	63.22	67.95	72.68	54.42	59.15	63.88	68.62
		35	60.96	65.69	70.43	75.16	56.90	61.63	66.36	71.09
		45	63.44	68.17	72.90	77.64	59.37	64.11	68.84	73.57
		55	65.92	70.65	75.38	80.11	61.85	66.58	71.32	76.05

multiple correlation coefficients differ significantly from zero ( $P < 0.01$  or  $P < 0.05$ ) for eighteen of the clinical indices measured. The values of the multiple correlation coefficient required for significance are 0.177 ( $P < 0.01$ ) or 0.147 ( $P < 0.05$ ).

Eighty five % of the subjects were between 20 and 69 years of age. The numbers of subjects belonging to the five decades between 20 and 69 are as follows: 20-29, 173; 30-39, 52; 40-49, 42; 50-59, 24; 60-69, 14. For each age group, mean age, height, and weight

Table V

Expected Values of Significant Clinical Indices According to Age, Height, and Weight:  
Hematological Variables

Variable Number	Name	Height (cm) Weight (kg) Age (years)	160				170			
			47.5 (3)	52.5 (4)	57.5 (5)	62.5 (6)	47.5 (7)	52.5 (8)	57.5 (9)	62.5 (10)
19	Erythrocytes	25	4.92	4.94	4.97	4.99	4.91	4.94	4.96	4.99
		35	4.82	4.84	4.87	4.89	4.81	4.84	4.86	4.89
		45	4.72	4.74	4.77	4.79	4.71	4.74	4.76	4.79
		55	4.62	4.64	4.67	4.69	4.61	4.64	4.66	4.69
20	Leukocytes	25	7112	7157	7203	7248	6492	6537	6583	6628
		35	7034	7079	7124	7170	6414	6459	6504	6549
		45	6955	7001	7046	7091	6335	6381	6426	6471
		55	6877	6922	6967	7013	6257	6302	6347	6392
21	Neutrophils	25	55.52	55.44	55.36	55.28	55.58	55.50	55.42	55.34
		35	57.29	57.20	57.12	57.04	57.34	57.26	57.18	57.10
		45	59.05	58.97	58.89	58.81	59.11	59.02	58.94	58.86
		55	60.81	60.73	60.65	60.57	60.87	60.79	60.71	60.63
22	Lympho- cytes	25	33.99	34.00	34.01	34.02	34.81	34.82	34.83	34.84
		35	33.03	33.04	33.05	33.06	33.85	33.86	33.87	33.88
		45	32.08	32.09	32.10	32.11	32.90	32.91	32.92	32.93
		55	31.12	31.13	31.14	31.15	31.94	31.95	31.96	31.97
23	Eosinophils	25	10.09	10.07	10.04	10.02	9.35	9.33	9.30	9.28
		35	9.14	9.12	9.10	9.07	8.40	8.38	8.36	8.33
		45	8.19	8.17	8.15	8.12	7.46	7.43	7.41	7.38
		55	7.25	7.22	7.20	7.18	6.51	6.48	6.46	6.44
24	Sediment- ation rate	25	12.01	12.18	12.35	12.52	11.38	11.54	11.71	11.88
		35	14.56	14.72	14.89	15.06	13.92	14.09	14.26	14.42
		45	17.10	17.27	17.44	17.60	16.47	16.63	16.80	16.97
		55	19.65	19.81	19.98	20.15	19.01	19.18	19.35	19.51
25	Volume of packed erythrocytes	25	48.57	48.53	48.50	48.46	48.48	48.44	48.41	48.37
		35	47.71	47.68	47.64	47.60	47.62	47.59	47.55	47.52
		45	46.86	46.82	46.78	46.75	46.77	46.73	46.70	46.66
		55	46.00	45.96	45.93	45.89	45.91	45.88	45.84	45.81

have been computed; the means are reported in the first three rows of table III [in columns (3), (5), (7), (9) and (11)]. For the eighteen significant clinical indices identified in table II, means for these age groups have also been computed. Entering mean age, mean height and mean weight of an age group in the multiple regression equations [columns (9) to (12) of table II], expected values for the eighteen clinical indices have also been computed [columns (4), (6), (8), (10) and (12) of table III]. The units of measurement are the same as those specified in column (5) of table I.

Tables IV and V have also been computed using the multiple regression equations. For two heights, four weights, and four ages, expected values for the seven significant chemical variables have been presented in table IV. Similarly table V presents the expected values for the seven significant hematological variables. Total cholesterol is composed of two statistically independent components, free cholesterol and cholesterol ester. Their correlation is  $-0.077$  which does not differ significantly from zero. Hence total cholesterol is not included in table IV and expected values are given only for the two components. The three measures of circulatory system performance, pulse rate, systolic and diastolic pressure, also have not been included, detailed data having been reported elsewhere for similar samples (*Das*, 1959, 1960, 1961; *Das* and *Mukherjee*, 1963). The expected values in tables IV and V illustrate the regression of the clinical indices on age, height and weight. The expected values are expressed in the units of measurement recorded in column (5) of table I.

#### Discussion

Clinical indices are required for the diagnosis of pathological and abnormal conditions. If a determination of a clinical index falls within the normal range, it is regarded as an acceptable or normal value. Climatic and nutritional factors may affect the normal range as they are likely to influence physiological processes. A value which indicates successful adaptation in one climate may not necessarily be successful in another climate. These considerations suggest that the normal ranges of clinical indices should be empirically determined for different geographical regions. The data presented in table I summarize observations on male residents of Calcutta, India. The ranges observed for these data [see column (8)] are compatible with the accepted ranges for Europe and North America [see column (9)] for the majority of the clinical indices. Differences apparently exist between the two ranges for the three cholesterol determinations (variables 13, 14 and 15), and have been reported previously for samples from the same population (*Das*, 1959, 1960, 1961). The lower limits of the normal range for these three indices may be attributed to the low weights of some subjects in the sample, since cholesterol and weight are significantly and positively correlated [see columns (5) and (8) of table II]. Differences between the two

ranges for the differential leukocyte count are mainly due to the high percentage of eosinophils (variable 23), a mean value of 8.88% obtaining for the entire sample of subjects. This observation may reflect hygienic conditions, as eosinophils are generally increased in parasitic and allergic conditions (Goodale, 1955). Sedimentation rate (variable 24), corrected for room temperature and volume of packed erythrocytes, also deviates from the accepted range, its mean value being 13.85 mm/h. Whether this result is due to the presence of chronic infectious processes (Goodale, 1955) or some other cause is not clear from the present data. Two further indices may be mentioned, glucose (variable 4) and chlorides (variable 5), both of which exhibit a lower limit than the accepted range and hence may be reflecting climatic and nutritional conditions.

A statistically significant increase with age has been shown for the following clinical indices of blood biochemistry: glucose, chlorides, calcium, neutrophils, and sedimentation rate (variables 4, 5, 8, 21 and 24 in table II). A statistically significant decrease in the clinical index occurring with an increase in age has been observed for erythrocytes, lymphocytes, eosinophils, and volume of packed erythrocytes (variables 19, 22, 23, and 25). The significant positive relationships between age and pulse rate, systolic and diastolic pressure (variables 26, 27, and 28) have been reported previously (Das, 1959, 1960; Hamilton *et al.*, 1954). Although variation with age has been reported in European and North American populations for inorganic phosphorus, hemoglobin and leukocytes, these variables (numbers 10, 16 and 20) have not shown significant relationships to age in the present data.

Clinical indices serve as measures of the effectiveness of physiological functioning. Statistically significant changes in these indices with advancing age may indicate changes in the *milieu interne* causing or reflecting regressive changes in the tissues or in specific organs. Glucose level serves as one measure of liver and pancreatic functioning; the levels of chlorides and calcium are mainly affected by kidney function; the sedimentation rate may reflect both liver and kidney functioning. For each of these variables, a significant increase with age has been noted, suggesting a decreased effectiveness of the liver, pancreas and kidneys with age. The changes might be expected to adversely influence the following tissue and cell functions: nutrition, maintenance of water content, temperature regulation, and protection. Among the hematological variables, percentage of neu-

trophils has shown a significant increase with age and the percentage of lymphocytes has decreased with age; these results are in agreement with accepted clinical findings (Goodale, 1955). They have particular relevance for protection of the tissues, and it may be appropriate that their sites of production differ: neutrophils originating in the bone marrow and the lymphocytes in the lymph nodes. While the action of neutrophils is phagocytic, the precise function of lymphocytes is unknown but they are believed to be concerned with antibody production and reaction to foreign substances. A decreased percentage of eosinophils with age does not seem to have been reported previously. Eosinophils are produced in the bone marrow, as are the neutrophils, but they decrease with age in the present data. Eosinophils and lymphocytes may be affected by the functioning of the adrenal glands and both may be concerned with protection of the tissues and cells (Goodale, 1955). Both the erythrocyte count and the volume of packed erythrocytes have also decreased with increasing age; this trend among adults has not been emphasized in the literature, although high values for infants and lower values for children, relative to adults, have been recorded (Goodale, 1955). Some possible causes of this effect may tentatively be advanced: a possible decreased production of erythrocytes by the bone marrow; alternatively, a cumulative effect of inadequate nutrition with age. Regardless of cause, this trend might affect cellular and tissue respiration. Observed values of the statistically significant variables for subjects according to age are presented in table III, along with expected values computed using the multiple regression equations. These observed and expected values provide illustrations of the effects of the significant relationships reported in table II.

Weight has previously been observed to be significantly related to total cholesterol, cholesterol ester, free cholesterol, systolic pressure, and diastolic pressure (Das, 1959, 1960, 1961; Das and Mukherjee, 1963). In addition to confirming these results, column (8) of table II shows that weight is significantly and positively related to thymol-turbidity (variable 11). The precipitate obtained in the thymol-turbidity test has been reported to contain globulin, phospholipid, cholesterol and thymol (Goodale, 1955). In normal individuals, the increased amount of serum lipids, including cholesterol, associated with increased weight may cause an increased turbidity of reaction in the thymol-turbidity test. In the present data, height has been significantly and negatively related to creatinine and

leukocytes (variables 7 and 20), that is, it appears that measurements of these two indices tend to be smaller for taller individuals.

For the diagnostic application of the findings of clinical tests, it is useful not only to have the normal range, but also to have expected values according to age, height, and weight. The observed value for a specific individual may then be compared with the expected value for persons of the same age, height, and weight. Deviation from the expected value may provide valuable information for the medical practitioner. Table IV presents a series of tables giving expected values for the chemical variables significantly related to age, height, or weight. A similar series is presented in table V for the significant hematological variables.

*Acknowledgment.* The correlation matrix for the twenty-eight variables was calculated by the Computing Laboratory of the Lafayette Clinic, Detroit, Michigan, U.S.A. through the kind courtesy of Dr. A. F. Az and Dr. J. L. Grial.

#### Summary

Clinical indices of blood biochemistry were determined for 358 male residents of Calcutta, India, ranging in age from 17 to 83 years. Twenty-eight variables were measured for each subject: physical variables of age, height, and weight; chemical variables for blood or serum, specifically, glucose, chlorides, urea, creatinine, calcium, uric acid, inorganic phosphorus, thymol-turbidity, bilirubin, total cholesterol, cholesterol ester, and free cholesterol; hematological variables, including hemoglobin, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, erythrocytes, leukocytes, neutrophils, lymphocytes, eosinophils, sedimentation rate, and volume of packed erythrocytes; and circulatory system variables of pulse rate and systolic and diastolic pressure. For each of the chemical, hematological, and circulatory system variables, mean, standard deviation, and normal range have been computed. Multiple regression analysis of the contribution of age, height, and weight to each of these variables has been carried out. Values of the following variables increased significantly with age: glucose, chlorides, calcium, neutrophils, sedimentation rate, pulse rate, systolic pressure and diastolic pressure. A significant decrease was observed for the following variables when age increased: erythrocytes, lymphocytes, eosinophils, and volume of packed erythrocytes. Increased weight was associated with a significant

increase in thymol-turbidity, total cholesterol, cholesterol ester, free cholesterol, systolic pressure, and diastolic pressure. Expected values of these variables at different ages, heights, and weights are presented, as are observed and expected values for subjects from 20 to 69 years of age.

### *Zusammenfassung*

Bei 358 Männern aus Calcutta, Indien, im Alter von 17-83 Jahren wurden Blutanalysen gemacht. Die folgenden Untersuchungen wurden bei allen Versuchspersonen durchgeführt. Physikalische Messungen: Alter, Größe und Gewicht. Biochemische Messungen im Blut oder Serum: Glucose, Chloride, Harnstoff, Kreatinin, Calcium, Harnsäure, anorganischer Phosphor, Thymoltrübungstest, Bilirubin, Totalcholesterol, Cholesterolester, freies Cholesterol. Hämatologische Bestimmungen: Hämoglobin, Hämoglobingehalt pro Erythrozyt, Erythrozyten, Leukozyten, Neutrophile, Lymphozyten, Eosinophile, Senkungsgeschwindigkeit, Hämatokrit. Zirkulationsmessungen: Herzfrequenz, systolischer und diastolischer Blutdruck. Für die biochemischen, die hämatologischen und die Zirkulationsmessungen wurden die Mittelwerte, die Standardabweichung und die Streuung berechnet. Die Zusammenhänge von Alter, Größe und Gewicht mit den obigen Daten wurden mittels multipler Regressionsanalysen berechnet. Folgende Messungen ergaben im Alter eindeutig höhere Werte: Glucose, Chloride, Calcium, Neutrophile, Senkungsgeschwindigkeit, Herzfrequenz, systolischer und diastolischer Blutdruck. Eine deutliche Abnahme der Werte wurde bei folgenden Messungen beobachtet: Erythrozyten, Lymphozyten, Eosinophile, Hämatokrit. Bei Gewichtsanstieg waren die Werte des Thymoltrübungstestes erhöht sowie das Totalcholesterol, die Cholesterolester, das freie Cholesterol, der systolische und der diastolische Blutdruck. Die gefundenen Werte der Messungen bei verschiedenem Alter, Größe und Gewicht stimmen für die Versuchspersonen von 20-69 Jahren mit den erwarteten überein.

### *Résumé*

Les caractéristiques biochimiques sanguines de 358 hommes de Calcutta d'âge variant de 17 à 83 ans ont été étudiées par l'auteur. 28 variables ont été mesurées sur chaque sujet: caractéristiques physiques, âge, taille et poids; caractéristiques chimiques du sang et du sérum et en particulier glucose, chlorures, urée, créatinine, calcium, acide urique, phosphore inorganique, test au thymol, bilirubine, cholestérol total, cholestérol estérifié, et cholestérol libre; des variables hématologiques, teneur moyenne des globules en hémoglobine, nombre de globules rouges, de globules blancs, de polynucléaires neutrophiles, de lymphocytes et d'éosinophiles, vitesse de sédimentation et volume des érythrocytes sédimentés; et des variables circulatoires, enfin, telle que la fréquence cardiaque et la pression artérielle systolique et diastolique. Dans chaque cas on a calculé la valeur moyenne, l'écart type et les valeurs extrêmes observées. En utilisant la technique de l'analyse des régressions multiples, on a étudié l'effet de l'âge, de la taille et du poids sur chacun des caractères étudiés. Une augmentation statistiquement significative avec l'âge a été notée dans le cas de la glycémie, de la teneur du sang en chlorures et en calcium, du nombre de polynucléaires neutrophiles, de la vitesse de sédimentation, de la fréquence cardiaque, et de la pression systolique et diastolique. Au contraire une diminution avec l'âge a été notée dans le cas du nombre des globules rouges, des lymphocytes, des éosinophiles et du volume des érythrocytes sédimentés. L'augmentation de poids à l'âge constant va de pair avec une élévation de la turbidité au thymol, du taux de cholestérol total, estérifié et libre, ainsi que de la pression artérielle. Les valeurs de ces différentes

variables aux différents âges, aux différentes tailles et aux différents poids sont données, ainsi que les valeurs observées et calculées dans l'intervalle d'âge allant de 20 à 69 ans.

### References

- Altman, P. L.; Dittmer, D. S. and Greb, R. M.*: Handbook of circulation (W. B. Saunders, New York 1959).
- American Heart Association*: Recommendations for human blood pressure determinations by sphygmomanometers (New York 1951).
- Bancroft, H.*: Introduction to biostatistics (Hoebner-Harper, New York 1957).
- Das, B. C.*: Etude sur la tension artérielle et le cholestérol en relation avec l'âge et le poids chez les hommes adultes du Bengale. *Biropologie* 20: 156-175 (1959). - An investigation of the relationships between diastolic and systolic blood pressure, age, weight, and serum cholesterol in adult male Bengalis. *Clin. Sci.* 19: 439-448 (1960). - Relationships between serum cholesterol, blood pressure, age and weight in adult male Bengalis. *Bull. Nat. Inst. Sci. India* 18: 29-35 (1961).
- Das, B. C. and Bhattacharya, S. K.*: Variation in lipoprotein level with changes in age, weight, and cholesterol ester. *Gerontologia* 5: 25-34 (1961). - Changes in human serum protein fractions with age and weight. *Canad. J. Biochem. Physiol.* 39: 569-579 (1961).
- Das, B. C. and Mukherjee, B. N.*: Variation in systolic and diastolic pressure with changes in age and weight. *Gerontologia* 8: 92-104 (1963).
- Geschickter, C. F.*: Some fundamental aspects of the aging process; in *Research in aging*; pp. 83-99 (Veterans Administration, Washington 1959).
- Goodale, R. H.*: Clinical interpretation of laboratory tests. 3rd Ed. (F. A. Davis, Philadelphia 1955).
- Gradwohl, R. B. H.*: Clinical laboratory methods and diagnosis. Vol. I. 5th Ed. (C. V. Mosby, St. Louis 1956).
- Hamilton, M.; Pickering, G. W.; Fraser Roberts, J. A. and Seury, G. S. C.*: The aetiology of essential hypertension. (1) The arterial pressure in the general population. *Clin. Sci.* 13: 11-35 (1954).
- Hauk, P. B.; Oser, B. L. and Summerson, W. H.*: Practical physiological chemistry. 13th Ed. (McGraw-Hill, New York 1954).
- Hepler, O. E.*: Manual of clinical laboratory methods. 4th Ed. (Charles C. Thomas, Springfield, Ill. 1955).
- Macy, J. C. and Kelly, H. J.*: Chemical anthropology (University of Chicago Press, Chicago 1957).
- Tárnözy, A. L.*: Clinical biochemical methods (Hilger and Watts, London 1958).
- Varley, H.*: Practical clinical biochemistry. 3rd Ed. (Heinemann, London 1962).

Author's address: Dr. B. C. Das, Biometry Research Unit, Indian Statistical Institute, 209 Bernettpore Trunk Road, Calcutta 35 (India).