REFERENCE RANGES OF 17 SERUM BIOCHEMICAL CONSTITUENTS IN A SINGAPORE POPULATION

K.S. CHUA
E. JACOB
C.W.K. LAM
I.K. TAN

**Clinical Biochemistry Laboratories
Department of Pathology, Outram Road,
Singapore 3.**

Chua Kim Seng, B.Sc.Hons., M.Sc., M.C.B., M.S.N.I.C.
Biochemist

Edward Jacob, B.Sc.Hons., Ph.D., M.R.I.C., C.Chem., M.C.B., M.S.N.I.C.
Biochemist

Christopher Lam, M.Sc., Ph.D., M.R.I.C., C.Chem.
Biochemist

Tan It Koon, B.Sc.Hons. Ph.D., C.Chem., F.R.I.C. M.C.B., F.S.N.I.C., M.R.C. Path.
Head

SYNOPSIS

This paper describes an investigation that was carried out to establish reference ranges for 17 serum biochemical constituents. The approach, instrumentation, and procedures used are described and the reference ranges obtained for the local population are described. The precisions of analysis for all the constituents studied are also presented. A discussion is made on the selection of the reference population, the statistical methods employed, and the importance of good analytical precision. Finally, a comparison of reference ranges obtained from several population groups is made.

INTRODUCTION

An important function of a clinical biochemistry laboratory is to provide, for the practising clinician, baseline biochemical data which can serve as a valid basis for the interpretation of his patients' biochemical test results. The use of the term 'reference range' as opposed to 'normal' range to describe these baseline data has been convincingly argued (Grasbeck, 1972), and reference range or reference interval is now the accepted terminology.

The conditions under which a 'reference range' for any biochemical constituent is established need to be specified, as many other factors other than disease may influence the results of a laboratory investigation. The main variables that can effect a marked shift in the range are:

(1) The type of population sampled e.g. ethnic group, sex, age, dietary habits
(2) The procedures used for the collection and handling of specimens
(3) The analytical methods employed
(4) The statistical techniques employed

As the reference ranges for blood constituents reported in our laboratory manual were established by methods and

instrumentation no longer in current use (Handbook of Clinical Biochemistry, 1971), it becomes necessary to replace them with new reference ranges obtained by the more precise methods and sophisticated instruments used today.

It is the object of this paper to present the new reference ranges for 17 serum constituents together with the approach, procedures and instrumentation used to establish them. The reproducibility of each analytical method used is also discussed and a comparison is made between the reference ranges thus established with those obtained by other workers for their population.

MATERIALS & METHODS

The Reference Population

The population sampled comprised 400 apparently healthy adults who were blood donors, laboratory personnel or individuals who presented themselves for health screening at the Singapore General Hospital. There were 350 males and 50 females and the age range of the population was between 20 and 60 years.

Serum Samples

No attempt was made to standardise the time at which blood was collected. However, samples collected for glucose, cholesterol and triglycerides were from fasting subjects. Serum was separated within two hours of blood taking and the analysis performed on the same day or within 48 hours. Bicarbonate analysis however were all performed within 3 hours of blood collection. Prior to their analysis, no special precaution was taken to prevent loss of CO_2 from blood samples. Specimens awaiting analysis were stored at 4°C.

Instrumentation & Methodologies

The instruments used included one single channel Generation 1 Technicon Autoanalyser, two Technicon SMA 6 Plus Autoanalysers, a Beckman Glucose Analyser, a Technicon PBI Autoanalyser and a dual channel Generation II Technicon Autoanalyser. The methodologies employed are given in Table 1.

Precision of analyses

To determine the precision of analysis for the serum constituents studied, pooled sera were included in every batch of analysis of each constituent. At the end of the investigation period, the 'between batch' values obtained for the pooled sera were used for the calculation of mean, standard deviation and coefficient of variation values.

Analysis of data

Frequency histograms were generated from the raw data of each of the constituents analysed (Fig. 1). Visual inspection of the histograms reveals different types of distribution. Some of the histograms approach a Guassian distribution whereas others are skewed. In order to further characterise the frequency distributions of the parameters being investigated cumulative percentage frequencies were calculated on the data (Hoffmann, 1963). The results were initially plotted on normal probability paper and those biochemical data which resulted in a straight line graph were considered to follow a Gaussian distribution. The results which gave a curve on normal probability paper were replotted on log-normal probability paper and those parameters resulting in a straight line graph on such a transformation were considered to have a log-normal distribution. Some of the data failed to give a straight line graph on either type of probability paper and these biochemical parameters indicated a more complex type of distribution. The results which had either a Gaussian or a log-Gaussian distribution were further subjected to parametric analysis while those with more complex distributions were subjected to non-parametric analysis.

Parametric Analysis

A sample from an apparently healthy population could include a number of extreme values or outliers which could result from analytical errors, subclinical disease or other causes. It is important to exclude obviously aberrant values, as inclusions of outliers in the calculation of the reference range will distort the magnitude of the range. For their exclusion we used a criterion suggested by Payne & Levell, 1968 and originating from Pierce, 1852. The mean and standard deviation (s.d.) of the set of values is first calculated using all the observed values. If a small number of figures amounting to less than 5% of the total, fall outside the range of three standard deviations from the mean, they are treated as outliers and rejected from further calculation. These outliers would apply to values falling on the plus or minus side of the range, three standard deviations from the mean. The remaining values are then recalculated for their

TABLE I. METHODOLOGIES AND PRECISIONS*

Constituent	Method	Precision (between batch)		
		Mean	s.d.	c.v. = $\frac{\text{s.d.}}{\text{mean}} \times 100$
Albumin	Technicon Mtd. No. SF4-0080 FES (modified) using Bromocresol green and citrate buffer	2.48 g% 4.30 g %	0.12 0.14	4.70 3.25
Alkaline Phosphatase	Technicon Mtd. No. SF4-0006 FCA using p-nitrophenol phosphate as substrate	114.56 IU/1 71.19 IU/1	5.20 3.94	4.54 5.54
Bicarbonate	Technicon Mtd. No. SF4-0008 FF5 using cresol red	19.69 mmol/l	2.57	13.07
Bilirubin	Technicon Mtd. No. SF4-0018 FFS using caffeine and sulphanilic acid	0.99 mg % 1.75 mg %	0.08 0.09	8.01 5.25
Calcium	Technicon Mtd. No. SF4-0003 FS4 using cresolphtalein complexone	9.87 mg %	0.20	2.04
Chloride	Technicon Mtd. No. SF4-0005 FC4 using mercuric thiocyanate	108.38 mmol/l 83.42 mmol/l	2.35 2.50	2.17 3.0
Cholesterol	Technicon Mtd. No. SE40016 FH4 using isopropanol extraction	193 mg % 186.94 mg %	8.41 7.25	4.36 3.88
Creatinine	Technicon Mtd. No. SF4-0011 FH4 using picric acid mercuric thiocyanate	1.3 mg % 7.5 mg %	0.16 0.30	12.30 4.01
Glucose	Beckman Glucose Analyzer Model No. ERA-2001	96.8 mg % 197.7 mg %	4.13 10.05	4.27 5.08

Table I — *cont'd*

Constituent	Method	Precision (between batch)		
		Mean	s.d.	c.v. = $\frac{\text{s.d.}}{\text{mean}} \times 100$
Inorganic Phosphate	Technicon Mtd. No. SF4-0004 FH4 using ammonium molybdate, stannous chloride and hydrazine sulphate	2.55 mg % 4.27 mg %	0.07 0.10	3.01 2.44
Potassium	Technicon Mtd. No. SF4-0007 FH4 using Flame Photometer	2.54 mmol/l 3.65 mmol/l 4.73 mmol/l	0.06 0.06 0.10	2.34 1.78 2.13
P B I	Technicon Mtd. No. N-56 by automatic digestion	6.05 ug % 8.30 ug %	0.40 0.33	6.50 3.60
Sodium	Technicon Mtd. No. SF5-0007 FH4 using Flame Photometer	145.88 mmol/l 126.83 mmol/l 118.09 mmol/l	1.69 1.52 1.44	1.16 1.20 1.22
Total Protein	Technicon Mtd. No. SF4-0014 FC4 using Biuret reagent	7.03 g % 4.73 g %	0.22 0.21	3.40 4.47
Triglycerides	Technicon Mtd. No. SE4-0023 FF3 using isopropanol extraction	142.5 mg %	10.30	7.30
Urea	Technicon Mtd. No. SF4-0001 FC4 using diacetylmonoxime and thiosemicarbazide	60.29 mg % 111.12 mg % 205.96 mg % 27.88 mg %	2.27 2.22 4.83 2.59	3.77 2.00 2.35 9.28
Uric acid	Modified Technicon Mtd. No. TF4-0013 FH4 using hydroxylamine and phosphotungstate	4.03 mg % 6.02 mg %	0.14 0.16	3.50 2.70

*(Number of analyses on each test carried out once daily over a period of 4 months totalled 94)

mean and s.d. values. For biochemical parameters having a log-Gaussian distribution, the individual values are first converted to their logarithmic values before calculations for the mean and s.d. values are made. The reference range is then derived by taking the set of values spanning the mean plus or minus twice the s.d. and then anti-logging the values obtained.

Non-parametric analysis

Outliers are excluded from further calculations by using the technique proposed by Reed, Henry and Mason (Reed, Henry, Mason 1971). By this method, an extreme value is rejected if the difference between it and its closest neighbour is more than one third of the entire range of observed values. The reference range is then calculated from the remaining values by the method of percentiles as described by Reed et al (Reed, Henry, Mason, 1971).

RESULTS

Precision of analyses

Table I gives the precisions of analyses for the 17 serum constituents investigated.

Frequency distributions

Fig. 1 shows the frequency distributions obtained for the 17 serum constituents studied.

Cumulative percentage frequency plots

Fig. 2 gives the plots of data with Gaussian/log-Gaussian/non-parametric distribution on normal/log-normal probability paper.

Reference ranges

Reference ranges (in conventional and SI units) for the 17 serum constituents studied are given in Table II & III.

DISCUSSION

Selection of the Reference Population

There are both practical and theoretical problems in deriving a reference range (Robinson, 1971; Zilva & Pannall, 1973). The choice of the reference population is of paramount importance and the most common practice is to use a healthy non-hospitalised population as a reference. The best choice, however, would be a group of healthy individuals who had rested some days in hospital prior to having their blood taken for analysis, but this would present administrative difficulties. An alternative approach would be to use patients themselves as the reference population and derive a reference range by statistical analysis of their results (Hoffmann & Waid, 1966; Becktel, 1970; Pryce, Haslaw & Wootton, 1969; Wootton and King, 1953; Hoffmann, 1963; Newmann, 1968). This however has been critisized (Amador & Hsi, 1969). A more selective approach has been the use of 'non-renal' outpatients (O'Halloran et al, 1970) or general practitioner patients attending for haematological but not biochemical tests (Little et al, 1974) as the reference population.

Workers in this field have not satisfactorily defined the optimal reference population, and blood donors have historically formed the most convenient and commonly used reference population. Our local blood donors usually comprise people within a narrow age range, and the reference values obtained for their serum constituents are applicable for comparison, only to a small proportion of the patient population seeking medical care. In order to extend the age range of our reference population, we included laboratory personnel and individuals who presented themselves for health screening with the population of blood donors.

Choice of statistical method

There are several statistical approaches for the calculation of reference ranges. They can be broadly classified into parametric and non-parametric methods. For parametric methods to apply, the raw data obtained from a reference population should be distributed in a Gaussian fashion or they should be mathematically transformable to fit a Gaussian distribution. Non-parametric methods do not make any assumption about the type of distribution of the raw data.

There are many advantages in dealing with a Gaussian distribution of values, the most important of which is the fact that with such a distribution, multiples of the standard deviation mark certain limits on the scatter of the observations. Two standard deviations above and two standard deviations below the mean of the set of values mark the points within which 95% of the observations lie. We used this mathematical fact to set the limits of our reference ranges for those biochemical parameters which gave a Gaussian or log-Gaussian distribution. For frequency distributions which were neither Guassian nor Log-

TABLE II. REFERENCE RANGES FOR 15 COMMONLY ESTIMATED SERUM CONSTITUENTS

Constituents	Frequency* Distribution	Number	Mean	s.d.	Ref. range (conventional units)	Ref. range (S. I. units)
Albumin	G	326	4.4	0.34	3.7 — 5.1 g %	37 — 51 g/l
Alkaline phosphatase	LG	332	—	—	32 — 105 IU/l	32 — 105 IU/l
Bicarbonate	G	141	25	2.8	19 — 31 mmol/l	19 — 31 mmol/l
Bilirubin	LG	383	0.53	—	0.2 — 1.4 mg %	3 — 24 umol/l
Calcium	G	398	9.4	0.5	8.4 — 10.4 mg %	2.1 — 2.6 mmol/l
Chloride	G	183	102	3.2	96 — 108 mmol/l	96 — 108 mmol/l
Creatinine	LG	175	1.1	0.25	0.5 — 1.6 mg %	44 — 141 umol/l
Glucose	G	177	87	16	55 — 119 mg %	3.1 — 6.6 mmol/l
Inorganic phosphate	G	400	3.3	0.47	2.4 — 4.3 mg %	0.8 — 1.4 mmol/l
Potassium	G	182	4.15	0.36	3.3 — 4.9 mmol/l	3.3 — 4.9 mmol/l
Protein Bound Iodine	LG	151	5.5	—	3.8 — 8.0 u g %	299 — 630 nmol/l
Sodium	G	183	140	2.6	135 — 145 mmol/l	135 — 145 mmol/l
Total Protein	G	325	7.2	0.5	6.2 — 8.2 g %	62 — 82 g/l
Urea	G	184	31.5	7.4	17 — 46 mg %	2.8 — 7.7 mmol/l
Uric acid	G	165	6.1	1.1	3.9 — 8.3 mg %	232 — 494 umol/l

* G = Gaussian
LG = Log-Gaussian

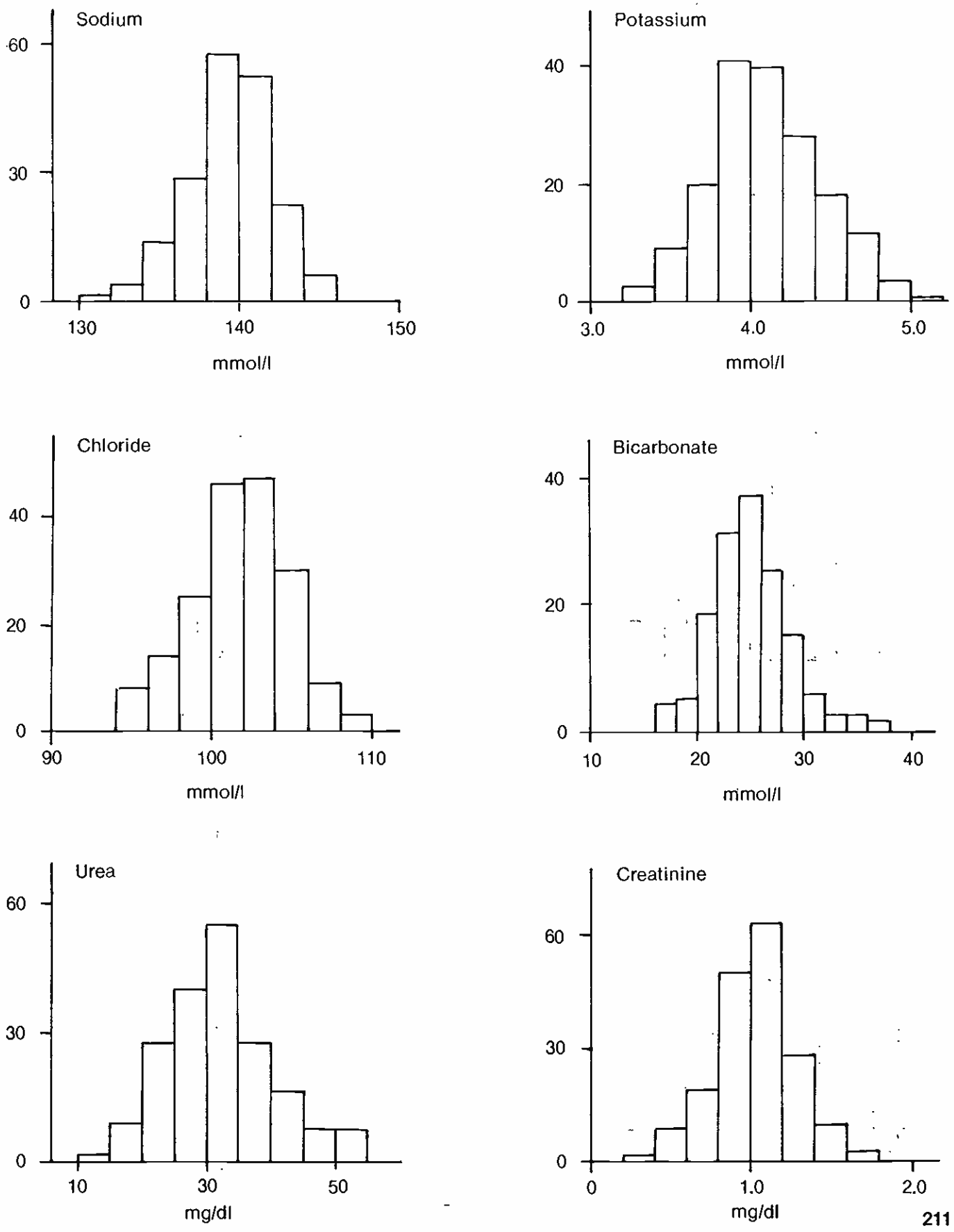
TABLE III. REFERENCE RANGES FOR CHOLESTEROL AND TRIGLYCERIDES

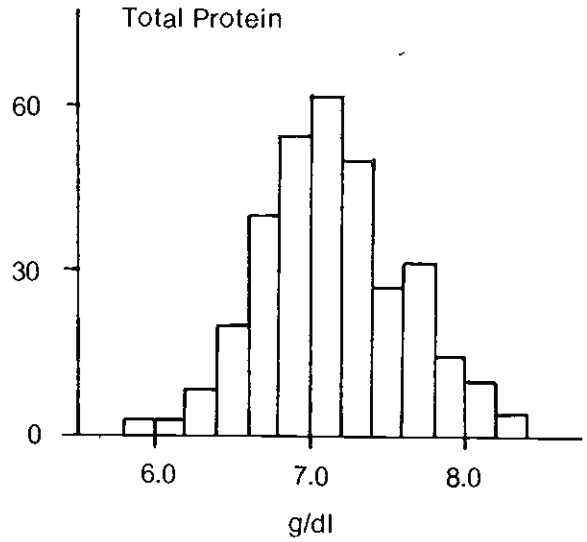
Constituent / Age	Frequency Distribution	Number	Ref. range (conventional units)	Ref. range (S. I. units)	Wilding, 1972	Henry, 1974
			mg %	mmol/l	mg %	mg %
Cholesterol	Non-parametric					
20 — 29 years		43	150 — 260	3.9 — 6.7	136 — 276	144 — 275
30 — 39 "		53	160 — 290	4.1 — 7.5	149 — 321	165 — 295
40 — 49 "		44	170 — 290	4.4 — 7.5	162 — 326	170 — 315
50 — 59 "		42	185 — 290	4.8 — 7.5	164 — 328	177 — 340
Triglyceride	Non-parametric					
20 — 29 years		42	40 — 140	0.45 — 1.58	—	) 30 — 135
30 — 39 "		48	50 — 170	0.56 — 1.92	—	) for the
40 — 49 "		31	75 — 170	0.84 — 1.92	—	) entire
50 — 50 "		31	50 — 185	0.56 — 2.09	—	) age range

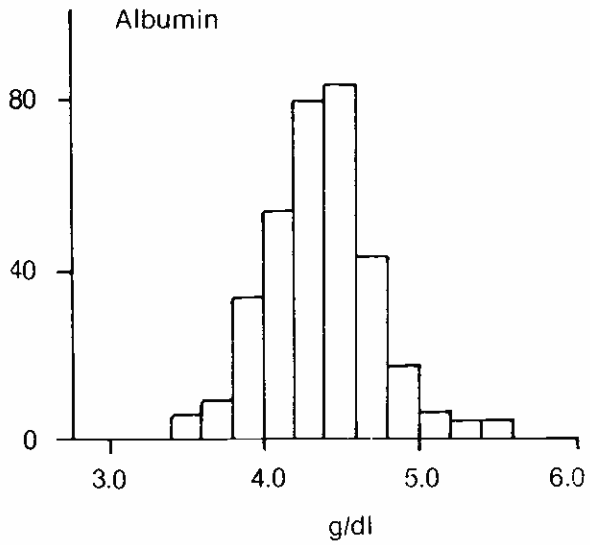
Fig. 1

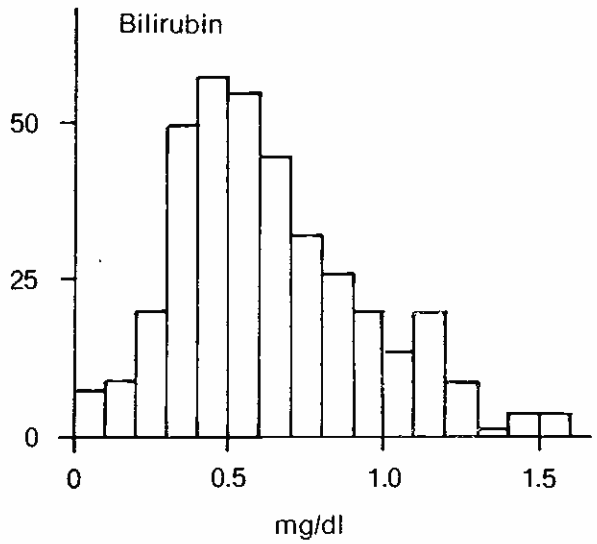
Frequency histograms of serum biochemical constituents

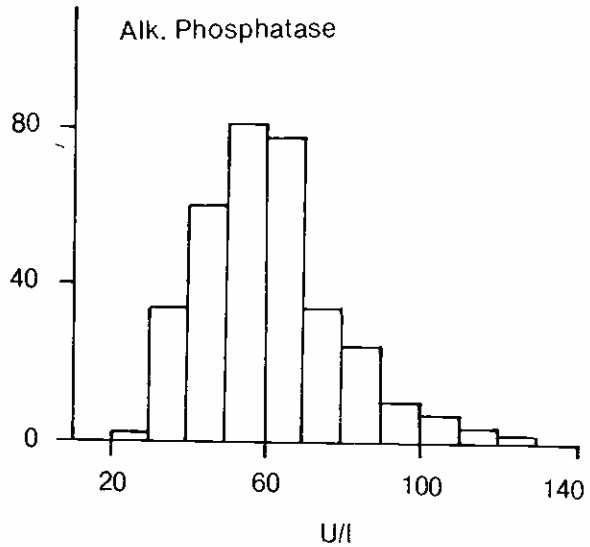
The x-axis shows the concentration of the constituent
The y-axis shows the no. of individuals

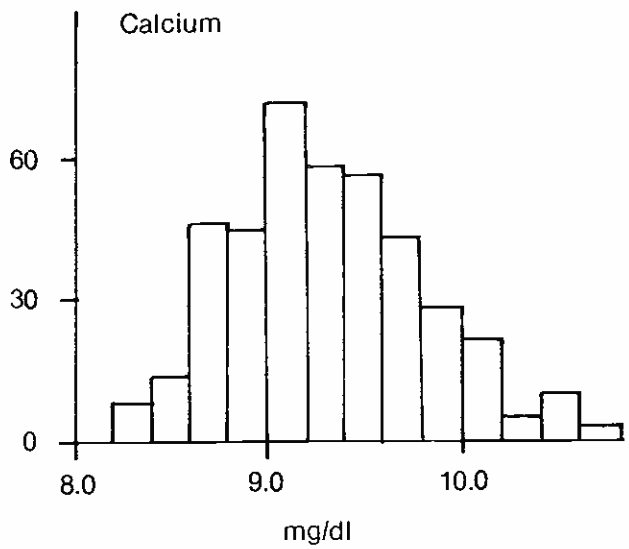


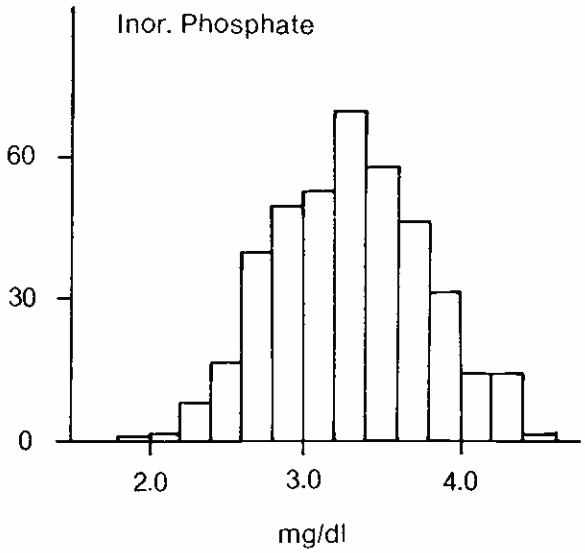

Total Protein
60
30
0
6.0
7.0
8.0
g/dl

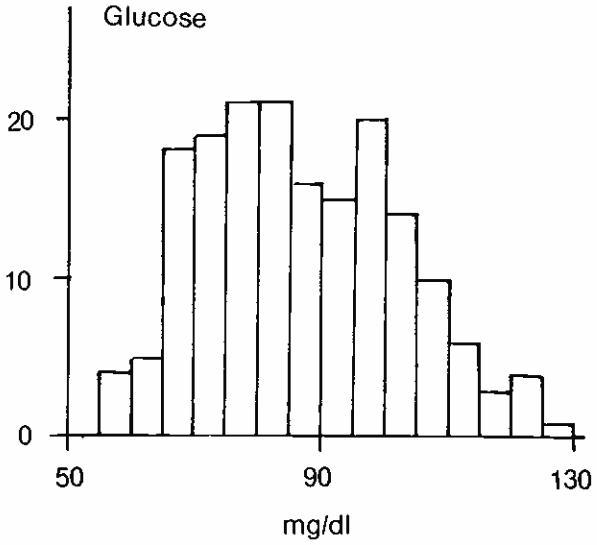

Albumin
80
40
0
3.0
4.0
5.0
6.0
g/dl

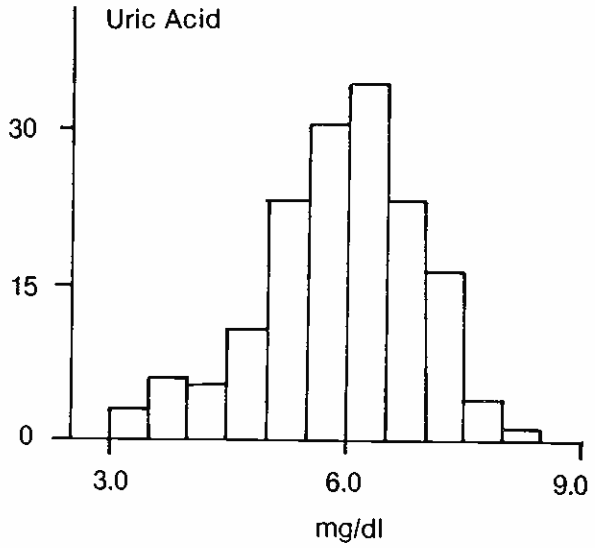

Bilirubin
50
25
0
0
0.5
1.0
1.5
mg/dl

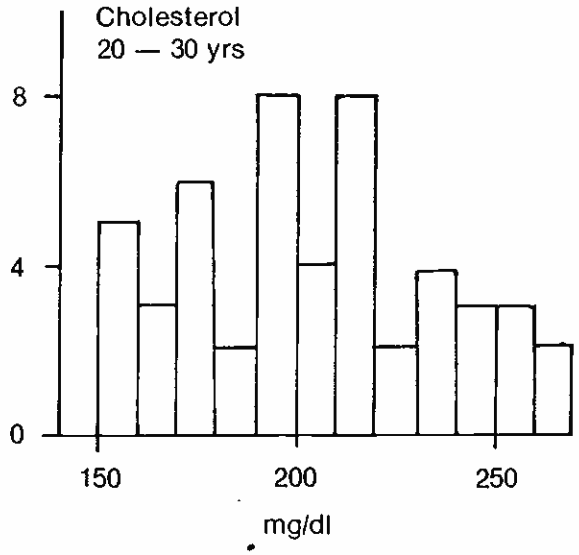

Alk. Phosphatase
80
40
0
20
60
100
140
U/l

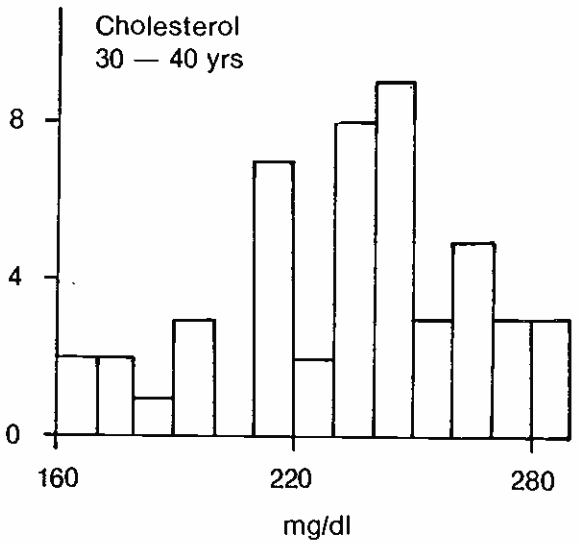

Calcium
60
30
0
8.0
9.0
10.0
mg/dl

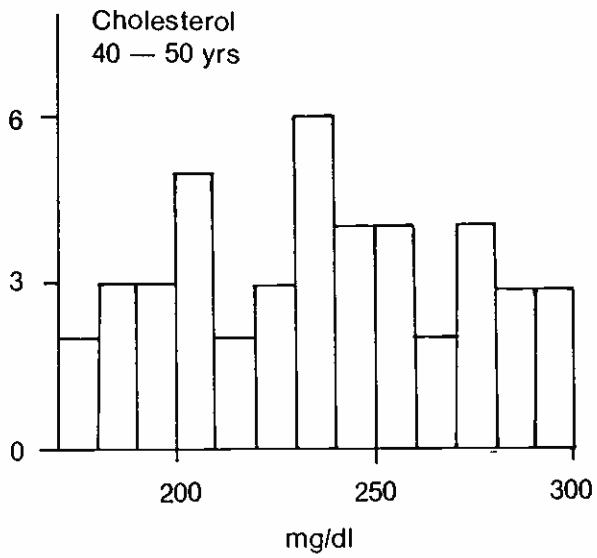

Inor. Phosphate
60
30
0
2.0
3.0
4.0
mg/dl

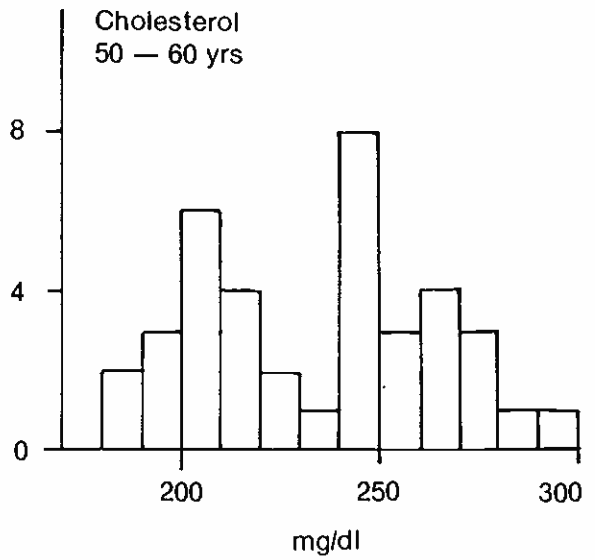

Glucose
20
10
0
50
90
130
mg/dl

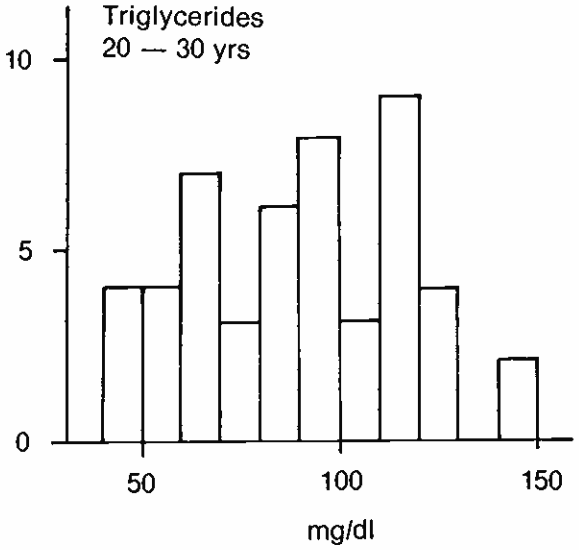

Uric Acid
30
15
0
3.0
6.0
9.0
mg/dl

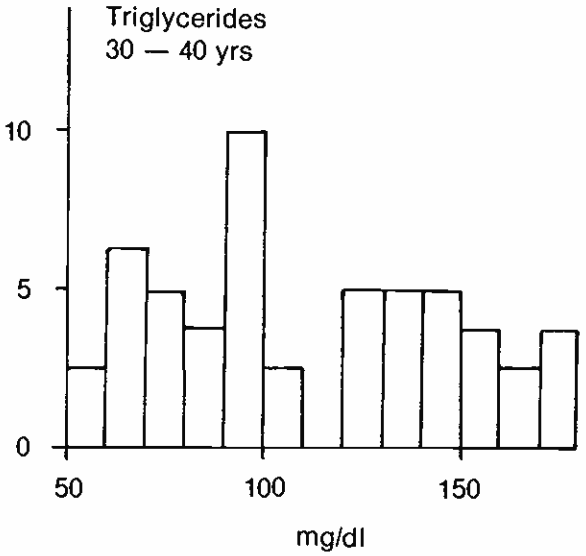

Cholesterol
20 — 30 yrs
8
4
0
150
200
250
mg/dl

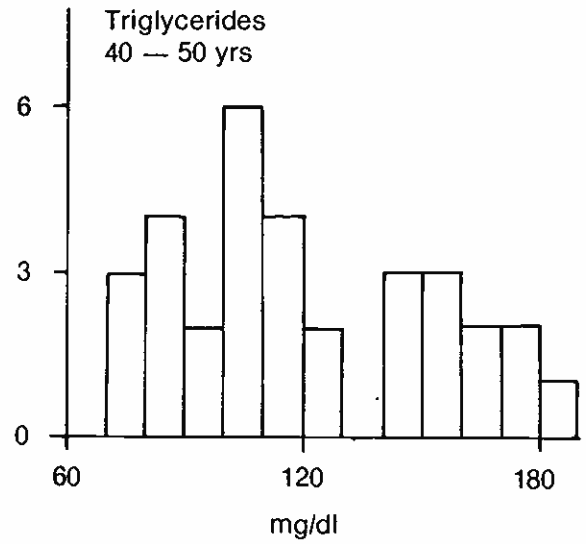

Cholesterol
30 — 40 yrs
8
4
0
160
220
280
mg/dl

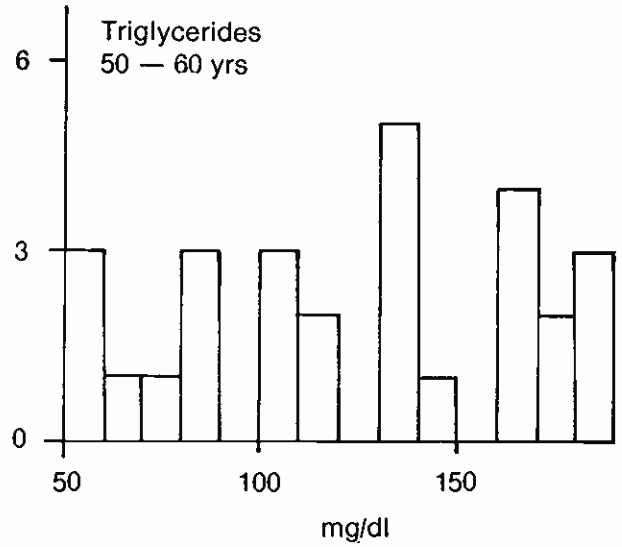

Cholesterol
40 — 50 yrs
6
3
0
200
250
300
mg/dl


Cholesterol
50 — 60 yrs
8
4
0
200
250
300
mg/dl


Triglycerides
20 — 30 yrs
10
5
0
50
100
150
mg/dl


Triglycerides
30 — 40 yrs
10
5
0
50
100
150
mg/dl


Triglycerides
40 — 50 yrs
6
3
0
60
120
180
mg/dl


Triglycerides
50 — 60 yrs
6
3
0
50
100
150
mg/dl

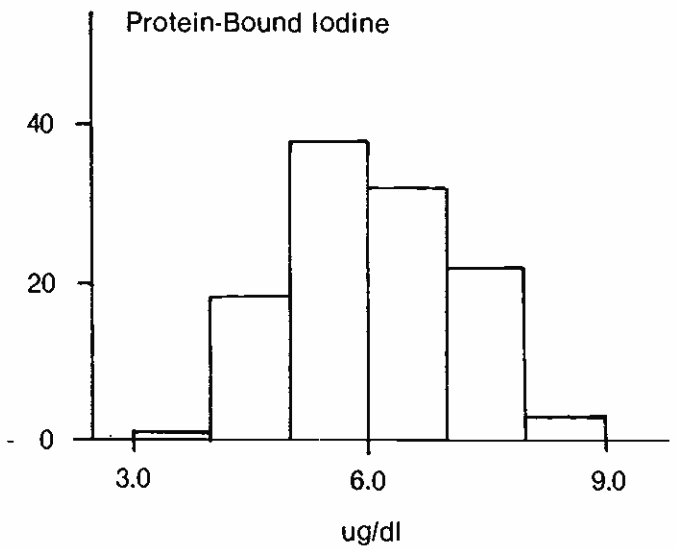

Protein-Bound Iodine
40
20
0
3.0
6.0
9.0
ug/dl

Fig. 2 Cumulative frequency plots

A) Gaussian distribution on normal probability paper

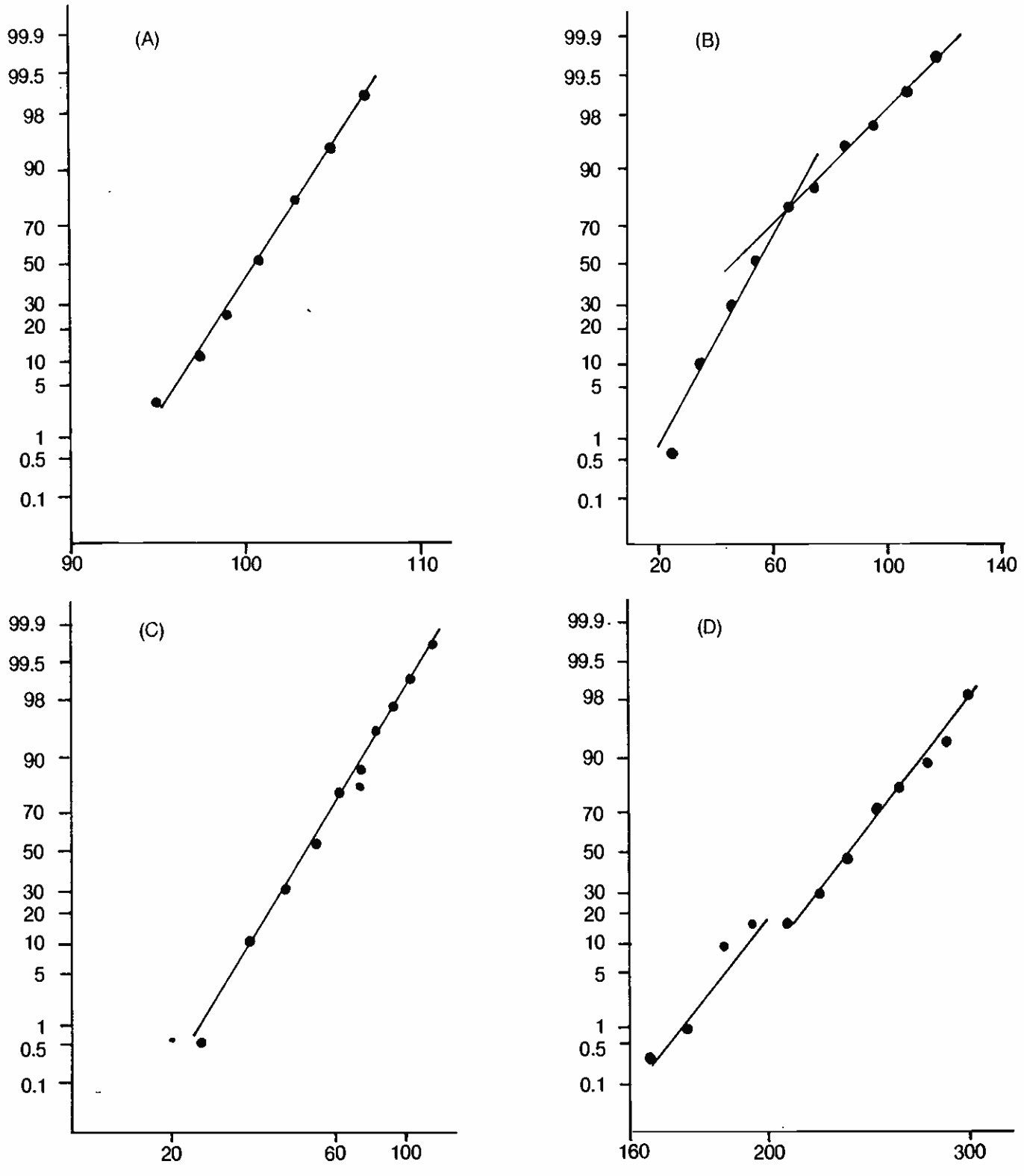
B) Log-gaussian distribution or non-parametric distribution on normal probability paper

C) Log-gaussian distribution on log-normal probability paper

D) Non-parametric distribution on log-normal probability paper

The x-axis shows the concentration of the constituent

The y-axis shows the cumulative percentage



Gaussian, we applied the non-parametric "percentile" method of Reed, 1971 to set the limits of the reference range. Tables II & III indicate the type of analysis used for deriving the reference ranges for the 17 biochemical parameters studied.

The influence of precision on the reference range

In deriving a reference range from a group of subjects, the two most important components to be considered are the individual biological variation (or the within-person variation), and the analytical variation. The observed variation expressed as a standard variation (SD_o) is given as follows:

$$SD_o = \sqrt{SD_b^2 + SD_a^2}$$

where SD_b = standard deviation of the biological variation and SD_a = standard deviation of the analytical variation. Gowenlock, 1969 has shown that small changes in SD_a such as may occur in normal practice, do not affect the range gravely. The observed individual biological variation is made up of pre-instrument errors and the 'true' biological variation within the person. The possible sources of pre-instrument errors may be venepuncture technique, time of day, posture and diet. If these sources of errors are minimised or standardised and the analytical techniques employed have good precision, the resulting reference ranges would clearly improve the diagnostic usefulness of the test value.

Table I summarises the precision of the analytical methods employed in this study. These values were obtained from day to day quality control which was monitored by analysis of five pooled control serum and by the use of assayed commercial quality control sera. In addition, performance in the National Quality Control Scheme (Jacob & Tan, 1977) and in the Wellcome Quality Control Scheme was used as a guide to overall and long term consistency. All these quality control techniques were operative throughout the period of study.

Comparison of reference ranges

Tables III & IV compare the reference ranges obtained for the local population with those established by foreign workers for their own population. It can be seen that the reference ranges obtained for the local and foreign populations have similar but not identical values. The observed differences are generally known to be due to differences in methodology, race of the population and environmental and physiological factors. The wider range obtained for serum bicarbonate levels for our local population could be attributable to the fact that no special precautions were taken to prevent loss of carbon-dioxide from blood specimens whilst they awaited analysis. For the interpretation of patient laboratory data in routine clinical practice, it is therefore important to compare the value obtained with the appropriate reference range established by the laboratory performing the test.

REFERENCES

1. Amador, E. & Hsi, B.P.: Indirect methods for estimating the normal range, Amer. J. Clin. Path., 52, 538, 1969.
2. Becktel, J.M.: Simplified estimation of normal ranges from routine laboratory data. Clin. Chim. Acta, 28, 119-125, 1970.
3. Gowenlock, A.H.: The influence of Accuracy and Precision on the normal range, Ann. Clin. Biochem., 6, 3, 1969.
4. Grasbeck, R.: Types of reference groups. Scand. J. Clin. Lab. Invest., 29 (Supplementum 126), 192, 1972.
5. Handbook of the Clinical Biochemistry Laboratories, Department of Pathology, 1971.
6. Henry, R.J., Cannon, D.C. & Winkelman, J.W. — Editors. Clinical Chemistry, Principles & Technics. Harper & Row, Publishers, 1974, 2nd Edition.
7. Hoffmann, R.G.: Statistics in the practice of medicine. J. Amer. Med. Assoc. 185, 864, 1963.
8. Hoffmann, R.G. & Waid, M.E.: Normal values from results of testing patients' specimens. In Stefanini, M. (Ed.): Progress in Clinical Pathology, Vol. 1, N.Y. Grune & Stratton, 1966, pp. 11-25.
9. Jacob, E., & Tan I.K.: The need for quality control in Clinical Laboratories, Proc. of the XII Singapore-Malaysia Congress of Medicine, 12, 242-245, 1977.
10. Little, A.J., Williams, R.B., Parker, S.D. & Payne, R.B.: The derivation of biochemical normal ranges from a hospital outpatient population. Clin. Chim. Acta 57, 91, 1974.
11. Neumann, G.J.: The determination of normal ranges from routine laboratory data, Clin. Chem. 14, 979, 1968.
12. O'Holloran, M.W., Studley-Ruxton, J., Wellby, M.L.: Comparison of conventionally derived normal ranges with those obtained from patients' results. Clin. Chim. Acta 27, 35, 1970.
13. Payne, R.B. & Levell, M.J.: Redefinition of the normal range from serum sodium, Clin. Chem. 14, 172-178, 1968.
14. Pierce, B.: Criterion for the rejection of doubtful observations. Astronomical J., 2, 161-163, 1852.
15. Pryce, J.D., Haslam, R.M. & Wootton, I.D.P.: Extration of normal values from a mixed hospital population. Ann Clin. Biochem. 6, 6-11, 1969.
16. Reed, A.H., Henry, R.J. & Mason, W.B.: Influence of statistical methods used on the resulting estimate of normal range. Clin. Chem. 17, 275-284, 1971.
17. Roberts, L.B.: The normal ranges with statistical analysis for 17 blood constituents. Clin. Chim. Acta 16 (1), 69-78, 1967.
18. Robinson, R.: Clinical Chemistry & Automation. London, Griffin, 1971.
19. Whitby, L.G. Percy-Robb, I.W. & Smith, A.F.: Lecture notes on Clinical Chemistry. Blackwell Scientific Pub., 1975. 1st Edition.

TABLE IV. COMPARISON OF REFERENCE RANGES

Constituent		Local	Roberts, 1967	Wootton, 1974	Whitby, 1975	Henry, 1974
Albumin	g %	3.7 — 5.1	3.6 — 4.7	3.6 — 4.8	3.6 — 4.7	3.5 — 5.0
Alkaline phosphatase	IU/l	32 — 105	—	—	—	—
Bicarbonate	mmol/l	19 — 31	—	24 — 30	24 — 30	22 — 30
Bilirubin	mg %	0.2 — 1.4	0.1 — 1.1	0.1 — 0.5	0.1 — 1.0	up to 1.5
Calcium	mg %	8.4 — 10.4	8.8 — 10.4	9.4 — 11.0	8.5 — 10.0	9.2 — 11.0
Chloride	mmol/l	96 — 108	98 — 107	100 — 107	100 — 107	98 — 109
Creatinine	mg%	0.5 — 1.6	0.7 — 1.4	0.1 — 1.4	0.6 — 1.2	0.9 — 1.4
Glucose	mg %	55 — 119	65 — 110	63 — 100	65 — 105	70 — 100
Inorganic Phosphate	mg %	2.4 — 4.3	2.3 — 4.2	2.8 — 4.2	2.5 — 4.5	2.5 — 4.8
Potassium	mmol/l	3.3 — 4.9	3.6 — 4.7	3.8 — 5.2	3.8 — 5.2	3.6 — 5.5
Protein Bound Iodine	u.g %	3.8 — 8.0	—	—	3.9 — 7.5	—
Sodium	mmol/l	135 — 145	134 — 143	136 — 149	136 — 149	135 — 155
Total Protein	g %	6.2 — 8.2	6 — 7.4	6.1 — 7.7	6.0 — 8.0	6.6 — 8.3
Urea	mg %	17 — 46	19 — 43	14 — 38	15 — 40	17 — 56
Uric acid	mg %	3.9 — 8.3	2.9 — 7.4	2.0 — 7.0	2.0 — 7.0	2.8 — 7.5

20. Wilding, P., Rollason, J.G., & Robinson, D.: Patterns of change for various biochemical constituents detected in well population screening. Clin. Chim. Acta, 41, 375-387, 1972.
21. Wooton, I.D.P.: Microanalysis in Medical Biochemistry, Churchill Livingstone, Edin, & London 1974, 5th Edition.
22. Wooton, I.D.P., King, E.J.: Normal values for blood constituents (interhospital differences). Lancet 1, 470, 1953.
23. Zilva, J.F., Pannal, P.R.: Clinical Chemistry in Diagnosis and Treatment, London. Lloyd-Duke, 1973.