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June 1993

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## Recommended Citation

Molla, A., Khurshid, M., Manser, W. T., Lalani, R., Alam, A., & Mohammad, Z. (1993). Suggested reference ranges in clinical chemistry for apparently healthy males and females of Pakistan. *Journal of Pakistan Medical Association*, 43(6), 115-115.

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# Suggested Reference Ranges in Clinical Chemistry for Apparently Healthy Males and Females of Pakistan

Pages with reference to book, From 113 To 115

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## Abstract

Seven hundred and eighty six apparently healthy males (418) and females (368) aged 0-69 years were randomly selected for estimation of reference ranges of 24 serum analytes at the clinical chemistry laboratory of The Aga Khan University Hospital (AKUH). Of the total study samples, 56% (439/786) were in the paediatric age group (0-14 years) and 44% (347/786) in the adult (15-60 years) group. Beckman Astra Ideal Autoanalyzer was used for all the estimations. Mean and standard deviations (SD) were calculated for each of the age groups. Reference ranges were calculated following standard methods of the International Federation of Clinical Chemistry (IFCC) and International Committee for Standardization in Haematology (ICSH) (JPMA 43:113, 1993).

## Introduction

In a Clinical chemistry laboratory, established normal reference range of serum biochemical analytes is an essential information needed for comparison of the estimated values obtained from the patients. The validity of a reference range depends on various factors, e.g., subject, age, sex, methodologies, instruments used and overall laboratory environment. Previously some of the studies in Pakistan have reported normal ranges for some of the analytes<sup>1,2</sup> estimated from a limited number of apparently healthy population. The objective of the present study is to estimate and establish a complete set of widely used reference range values of the serum analytes following a standardized criteria of IFCC and ICSH<sup>3-6</sup>.

## Material and Methods

Seven hundred and eighty six healthy male and female volunteers aged between 0-60 years were selected for the study. The population was stratified into two groups, e.g., paediatric aged between 0-14 years and adults aged between 15-60 years. The adults were mostly employees, medical students, friends and relatives of the employees of AKUH. The paediatric group was selected mainly from a local private school and from the volunteering children of the employees of AKUH. A questionnaire form including questions on dietary habits, recent and past history of illness, current use of medication and ethnic origin were asked for revealing any obvious reason (diabetes or liver disease) for not including their estimated results in the final data analysis. After an overnight fast, 10 ml of blood samples were collected from the selected subjects in neutral glass tubes. Serum was separated within an hour and all the analytes were estimated within 12 hours of collection. Beckman Astra 24 channel autoanalyzer (Beckman Instrument Inc., USA) was used for estimation of 24 biochemical analytes: glucose, cholesterol, triglycerides, BUN, creatinine, sodium, potassium, chloride, bicarbonate, direct and total bilirubin, gamma glutamyl transferase (GGT), glutamate pyruvate transaminase (SGPT), alkaline phosphatase, total protein, albumin, globulin, glutamate-oxaloacetate transaminase (SGOT),

lactate dehydrogenase (LDH), creatinine phosphokinase (CPK), calcium, phosphorus, amylase and uric acid. Regular maintenance of the instruments is done daily, weekly, bi-weekly, monthly and yearly. Equipment calibration for each of the chemistry is done 8 hourly, using standardized calibrator samples supplied by Beckman Instruments. Internal quality control samples obtained from commercially prepared serum (Beckman) with three levels (low, medium and high) of concentrations of each constituent are used four times daily. In between hours, home made standardized pooled serum samples are also used as internal quality control sample. External quality control samples supplied by Wellcome External Quality Assessment Scheme, UK<sup>7</sup> are run twice monthly. All quality control results are scrutinized carefully before releasing patient's results. Reporting of results are carried out according to the regulations for laboratory certification by the College of American pathologists<sup>8</sup>.

### Statistical Analysis

Means and standard deviations were calculated for each of the male and female groups. From any set of results, values above or below 2SD were excluded as outliers. Means and standard deviation were calculated for the second time following which lower and higher ranges were calculated by using the formula  $\text{mean} \pm 2\text{SD}$ . This formula is only valid for those analytes, the distribution of which are of the Gaussian type. For the skewed pattern of distribution, following recommendation of IFCC, a non-pancreatic test was performed<sup>3-6</sup> to set lower and higher range values.

### Results

Age and sex distribution of the study population is given in Table I.

**Table I. Age and sex distribution of the study population.**

Age (years)	Male (No.)	Female (No.)	Total (No.)
0-4	17	18	35
5-9	111	82	193
10-14	124	87	211
15-19	34	28	62
20-24	22	20	42
25-29	16	24	40
30-39	26	31	57
40-49	24	40	64
50-59	26	25	51
≥60	18	13	31
<b>Total</b>	<b>418</b>	<b>368</b>	<b>786</b>

A relatively larger number of children aged between 5-14 years were selected as compared to other groups mainly because students of a private school were included in the study. The number of children in the age groups of 0-4 years and ≥60 years were relatively smaller as compared to the other groups. Suggested reference ranges for the serum analytes for Pakistani paediatric and adult population are given in Tables II and III respectively.

**Table II. Suggested reference ranges for Pakistani paediatric males and females (aged 0-14 years).**

Analytes (serum)	Males	Females
Glucose fasting	69-104 mg/dl	62-105 mg/dl
Cholesterol	110-219 mg/dl	113-233 mg/dl
Triglyceride	18-150 mg/dl	22-157 mg/dl
BUN	6-15 mg/dl	6-14 mg/dl
CRE	0.3-0.8 mg/dl	0.3-0.8 mg/dl
Na <sup>+</sup>	135-145 mmol/l	132-148 mmol/l
K <sup>+</sup>	3.4-5.0 mmol/l	3.7-5.2 mmol/l
Cl <sup>-</sup>	101-112 mmol/l	101-114 mmol/l
HCO <sub>3</sub> <sup>-</sup>	14-26 mmol/l	14-25 mmol/l
T. bilirubin	0.1-1.0 mg/dl	0.1-1.0 mg/dl
D. bilirubin	.07-0.1 mg/dl	.07-0.1 mg/dl
GGT	5-21 IU/l	5-19 IU/l
SGPT	9-30 IU/l	5-29 IU/l
Alk. phosphatase	91-359 IU/l	64-346 IU/l
Total protein	5.4-8.4 g/dl	5.6-8.5 g/dl
Albumin	3.5-5.3 g/dl	3.8-5.2 g/dl
Globulin	1.7-3.3 g/dl	1.8-3.3 g/dl
SGOT	13-49 IU/l	5-57 IU/l
LDH	244-827 IU/l	299-749 IU/l
CPK	16-240 IU/l	15-198 IU/l
Ca <sup>++</sup>	7.9-11.0 mg/dl	6.7-11.9 mg/dl
Inorganic phosphate	3.9-5.5 mg/dl	3.8-5.7 mg/dl
Amylase	49-142 IU/l	43-130 IU/l
Uric acid	2.1-5.7 mg/dl	2.4-5.6 mg/dl

**Table III. Suggested reference ranges for Pakistani adult males and females (aged 15 ≥ 60 years).**

Analytes (serum)	Males	Females
Glucose fasting	61-115 mg/dl	67-106 mg/dl
Cholesterol	114-273 mg/dl	107-272 mg/dl
Triglyceride	24-321 mg/dl	17-269 mg/dl
BUN	6-17 mg/dl	5-15 mg/dl
CRE	0.7-1.5 mg/dl	0.6-1.1 mg/dl
Na <sup>+</sup>	134-150 mmol/l	134-150 mmol/l
K <sup>+</sup>	3.5-4.9 mmol/l	3.6-4.9 mmol/l
Cl <sup>-</sup>	98-115 mmol/l	99-117 mmol/l
HCO <sub>3</sub> <sup>-</sup>	17-28 mmol/l	16-28 mmol/l
T. bilirubin	0.1-1.3 mg/dl	0.2-1.0 mg/dl
D. bilirubin	0-0.15 mg/dl	0-0.11 mg/dl
GGT	7-50 IU/l	6-37 IU/l
SGPT	2-30 IU/l	23-38 IU/l
Alk. phosphatase	19-146 IU/l	24-109 IU/l
T. protein	5.4-8.4 g/dl	5.3-8.2 g/dl
Albumin	3.2-5 g/dl	2.9-5.0 g/dl
Globulin	1.7-3.5 g/dl	1.7-3.9 g/dl
SGOT	9-43 IU/l	9-37 IU/l
LDH	165-640 IU/l	217-542 IU/l
CPK	17-197 IU/l	22-129 IU/l
Ca <sup>++</sup>	8.5-10.5 mg/dl	8.2-10.4 mg/dl
Inorganic phosphate	2.7-4.6 mg/dl	3.0-4.6 mg/dl
Amylase	36-140 IU/l	39-128 IU/l
Uric acid	3.8-8.2 mg/dl	2.8-6.1 mg/dl

Most of the serum analyte ranges were similar between the paediatric and adult groups. However, the reference range for alkaline phosphatase and lactate dehydrogenase were higher for the paediatric group compared to the adult group. For the purpose of comparison with the Pakistani values, western reference ranges obtained from two different sources<sup>9,10</sup> are given in Table IV.

**Table IV. Suggested reference ranges for adult western population.**

Analytes (serum)	Reference ranges (9)	Reference ranges (10)
Glucose fasting	70-110 mg/dl	70-105 mg/dl
Cholesterol	120-220 mg/dl	140-310 mg/dl
Triglyceride	40-150 mg/dl	Normal 140-220 mg/dl M 40-160 mg/dl F 35-135 mg/dl
BUN	8-25 mg/dl	7-18 mg/dl
CRE	0.6-1.5 mg/dl	M 0.6-1.2 mg/dl F 0.5-1.1 mg/dl
Na <sup>+</sup>	135-145 mmol/l	136-146 mmol/l
K <sup>+</sup>	3.5-5.0 mmol/l	3.5-5.1 mmol/l
Cl <sup>-</sup>	100-106 mmol/l	118-132 mmol/l
HCO <sub>3</sub> <sup>-</sup>	24-30 mmol/l	22-29 mmol/l
T. bilirubin	upto 1.0 mg/dl	
D. bilirubin	upto 0.4 mg/dl	
GGT		M 9-50 IU/l F 8-40 IU/l
SGPT	1-21 IU/l	8-20 IU/l
Alk. phosphatase	13-39 IU/l; infants and adoles. upto 104 IU/l	
Total protein	6.0-8.4 g/dl	6.4-8.3 g/dl
Albumin	3.5-5.0 g/dl	3.5-5.0 g/dl
Globulin	2.3-3.5 g/dl	2.2-4.0 g/dl
SGOT	7-27 IU/l	8-20 IU/l
LDH	45-90 IU/l	45-90 IU/l
CPK	F 10-79 IU/l M 17-148 IU/l	M 25-130 IU/l F 10-115 IU/l
Ca <sup>++</sup>	8.5-10.5 mg/dl	8.4-10.2 mg/dl
Inorganic phosphate	3-4.5 mg/dl	2.7-4.5 mg/dl
Amylase	4-25 IU/l	25-125 IU/l
Uric acid	3-7 mg/dl	M 3.5-7.2 mg/dl F 2.6-6.0 mg/dl

Pakistani reference ranges for cholesterol, triglyceride, alkaline phosphatase, LDH, CPK and amylase were higher and for other analytes were similar to western values.

## Discussion

Reference range values in clinical chemistry may be defined as the result of a quantitative analysis of a group of healthy individuals selected at random according to clearly specified criteria. It was necessary to establish a reference range value for clinical chemistry which will be essentially applicable in

Pakistani population. Using an internationally recognized and well calibrated equipment, e.g., Astra Autoanalyzer, we have attempted to develop our own values while vigorous quality control procedures were employed for obtaining precise and accurate results. For comparative analysis, the most up to date western reference range values used by the Massachusetts General Hospital<sup>9</sup> and by International Federation of Clinical Chemistry<sup>10</sup> are provided in Table W. In this study, we have stratified our population into two main groups providing separate reference range values for both paediatric males, females (Table II) and adult males, females (Table III) respectively. However, while comparing results between Pakistani and western adults, we obtained comparable results for most of the analytes except cholesterol, triglyceride, alkaline phosphate, lactate dehydrogenase, creatinine phosphatase and amylase values. Reference range values for these analytes are comparatively higher in our population than those of the western population as shown in Table IV. There could be various possible explanations for the discrepancy between these two sets of results obtained from two culturally different populations. Our results on cholesterol, triglyceride and alkaline phosphatase were critically analyzed and reported previously<sup>11,12</sup>.

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