



Reference Intervals for Serum Immunoglobulins IgG, IgA, IgM and Complements C3 and C4 in Iranian Healthy Children

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Abstract

Background: Determination of reference ranges of each serum protein in normal population of each country is required for studies and clinical interpretation. The aim of this study was defining reference range values of immunoglobulins and complement components in Iranian healthy children.

Methods: This study was conducted from June 2003 to June 2006 in Immunology, Asthma and Allergy Research Institute, Tehran University of Medical Sciences. Serum levels of IgG, IgM, IgA, C3 and C4 in 800 Iranian healthy children from newborn to 18 years of age in four population were measured by nephelometry. Kolmogorov-Smirnov tests and Pearson correlation tests were used for analysis.

Results: Our results mainly agree with previous reports, except for some discrepancy that might be due to the ethnic and geographic variety. There was a significant difference between two sexes only with IgA in the group of 1-3 months old, which was higher in male group and IgM in groups of 3-5, 6-8 and 9-11 years old that were higher in female groups. Mean of other serum immunoglobulins and complements was not significantly different between male and female groups.

Conclusion: These results can be considered as a local reference for use in laboratories, clinical interpretations, and research for Iranian children.

Keywords: Reference ranges, Immunoglobulin, Complement components, Iran

Introduction

Reference values of immunoglobulins (Igs) and complement components (CCs) might be different in each population. These data are essential for researches and making clinical diagnosis in every population (1). The determination of serum Igs constitutes one of the diagnostic approaches in immunological as well as infectious disease. The levels of Igs and CCs might vary in diverse geographical regions due to gender and racial differences. Although these differences are generally not significant, sometimes knowledge of these subtle varieties is critical for clinical explanation

(2). To our knowledge, this is the first normal range study for serum IgG, IgM, IgA, C3 and C4 of healthy children in Iran. Three studies on serum Igs in healthy adults exist (two first were performed by means of the RID Mancini and the last by using nephelometry method) (3-5) and also two studies evaluated normal range of serum total IgE in Iranian healthy adults and children (6,7). In addition, in 1997, quantitative serum level measurements of C3, IgG, IgA and IgM were carried out on 102 patients with esophageal carcinoma, in comparison with 66 healthy controls in Iran (8).

Many studies have been done for the determination of reference ranges for serum proteins such as Igs in the world (9-15). Newly, a study has been done to evaluate serum Ig G, A, M, G1, G2, G3, and G4 in 148 healthy Thai children aged 2-15 years by nephelometry (16).

The aim of the present study was to determine local reference values for serum IgA, IgM, IgG and complement components C3 and C4 by nephelometric methods in healthy Iranian children.

Material and Methods

Subjects

This study was conducted from June 2003 to June 2006 in Immunology, Asthma and Allergy Research Institute, Tehran University of Medical Sciences. A total of 800 healthy children and adolescents from newborn to 18 years of age were included in this study. They were chosen from healthy cases admitted for minor surgical procedures in the surgical ward of Children's Medical Center and Imam Khomeini Hospital of Tehran University of Medical Sciences. They were visited by a physician and were confirmed to have no acute or chronic diseases. Their nutritional and developmental status was normal at the time of sampling. Furthermore, a questionnaire was completed for them, which included health status, age, and sex. All of them were from Iranian population and ethnicities such as Fars, Turk, Kurd, and other population such as Arab and Turkmen throughout Iran. From all subjects or their guardians informed consent was obtained. One ml sterile serum was separated from the peripheral blood sample and was frozen at -80°C immediately. The research committee and Ethical Committee of Immunology, Asthma & Allergy Research Institute, Tehran University of Medical Sciences approved the study's protocol.

Measurement of IgG, IgM, IgA and complement components C3 and C4

Serum Igs, C3 and C4 concentrations were determined by nephelometry methods, according to the instructions described by the manufacturer (Mini-

nephTM, The binding site Ltd., Birmingham, UK). Highly lipaemic, turbid or hemolyzed samples were unsuitable for nephelometric measurements and were excluded from the study.

Statistical Analysis

Normal distribution analysis was done by Kolmogorov-Smirnov tests. Differences in Igs and CC concentrations between males and females and between each population were analyzed with the Mann-Whitney U-test. P-values of less than 0.05 were considered significant. Correlation between age and each factor was concluded by means of the Pearson's correlation test.

Results

Eight hundred selected subjects, from newborn to 18 years of age, 349 females, and 451 males in 10 age groups were entered in this study. As the raw data of IgA, IgG, IgM, C3 and C4 concentrations did not follow normal distribution ($P < 0.05$), these data were transformed into the natural logarithm for analysis. Normal range for all factors was calculated by the natural logarithm of mean $\pm 1.96 \times \text{SD}$. Ig and CC means with 95% confidence intervals in ten age groups are demonstrated in table 1 (Table 1). IgA in 1-3 months old males was significantly higher compared to the same age females ($P = 0.029$) and IgM in the groups of 3-5 years old, 6-8 years old and 9-11 years old females was significantly higher compared to the same age males ($P = 0.019, 0.001$ & 0.032 respectively). In the other age groups there was no significant difference regarding sex.

Correlation coefficient tests showed a significant positive correlation between age and serum IgA and IgM concentrations ($P < 0.05$). There was no significant correlation between weight and length in each age group and serum factors measured ($P > 0.05$). There was no significant difference between mean of all evaluated factors in each population group.

Table 1: Mean of serum Immunoglobulins and complement components with 95% confidence intervals in ten age groups

Age Groups	n	IgA (mg/dL)		IgG (mg/dL)		IgM (mg/dL)		C3 (mg/L)		C4 (mg/L)	
		Mean	Range	Mean	Range	Mean	Range	Mean	Range	Mean	Range
Newborn	43	5.6	0-22	507.6	133-1037	26	5-120	690	324 - 1127	167	69 - 398
1-3 month	129	18.3	4-79	313.8	55-625	39.6	8-144	705.9	269 - 1280	130.2	51 - 324
4-6 month	45	24.4	7-89	340	49-611	49.8	16-158	743.9	389 - 1321	148	59 - 372
7-12 month	132	22.1	4-110	371.8	11-769	48.8	11-221	723.3	380 - 1313	134	51 - 355
13-24 month	107	27.4	7-115	481.8	127-907	64.8	20-207	775.9	385 - 1375	141.7	58 - 347
25-36 month	112	35.8	9-148	507.4	216-1010	62.1	17-223	817.8	394 - 1465	150	58 - 398
3-5 year	62	73.3	32-178	668.3	280-1108	97.6	42-227	966.7	588 - 1589	171	78 - 372
6-8 year	60	84.3	36-204	751.5	443-1095	96.8	39-240	935.6	588 - 1490	172.7	91 - 324
9-11 year	63	96.8	39-246	881.5	422-1406	100.2	40-251	997.3	624 - 1596	184.7	93 - 363
12-18 year	47	122	60-263	886.5	500-1330	85.5	30-228	1026.4	625-1468	192.6	89 - 417

Discussion

In clinical immunology, the knowledge of immune system components and their normal values is vital. In this field, many studies have been performed (15, 17, 18). In most sciences, such as immunology, an approved, authoritative, global standard reference interval has been determined, which is beneficial in laboratory and clinical tests (9, 15, 19).

Usually, human subjects all over the world show variations, albeit little, in serum immune factor levels. This could be probably due to health, nutritional, environmental and racial variations (2, 20, 21). In our country, previously reported studies mostly include adult age groups and nor children (3-5). Two studies evaluated normal range of serum total IgE in Iranian healthy adults and children (6,7). In this study, we used nephelometry as an automated standard method for measuring serum immune factors (10, 13). As reported by others, we found that concentration of IgA, IgM and CCs increased with age from newborn to 18 years except in the case of IgG. IgG was high in the newborns, due to placental transfer of this immunoglobulin (1, 14, 22). The higher levels of IgM in three age groups agree with previous studies, as some studies found a relationship be-

tween the number of X chromosomes and IgM concentrations (23, 24).

As indicated in results, there is no significant difference between special populations in no one of study factors. Therefore, these results will be usable for all Iranian populations. Results were obtained in this study are similar to other international trustworthy references, except for some differences, which are probably related to ethnic factors (1, 9, 10, 13). In conclusion, these results can be considered as a source of a reliable local reference for use in laboratories, clinical interpretation, and research.

Ethical considerations

Ethical issues (Including plagiarism, Informed Consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc) have been completely observed by the authors.

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