

Change in Referral Diagnoses and Diagnostic Delay in Hypogammaglobulinaemic Patients during 28 Years in a Single Referral Centre

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Key Words

Common variable immunodeficiency • X-linked agammaglobulinaemia • Immunoglobulins

Abstract

Background: The classical clinical manifestation of untreated immunoglobulin deficiency comprises predominantly recurrent and complicated respiratory tract infections. Before the 1980s, little was known about the clinical manifestation of immunodeficiency in the general medical population, and also the availability of serum immunoglobulin laboratory determination was not sufficient, leading to a significant diagnostic delay. **Methods:** We have analysed the diagnostic delay and referral diagnoses in patients in whom any form of primary hypogammaglobulinaemia had been diagnosed at our department, which was established in 1981. **Results:** Comparing the diagnostic delay in the 1980s (19 patients, median 5.5 years), the 1990s (37 patients, median 3.5 years) and the years 2001–2008 (33 patients, median 1 year), a significant decrease was observed ($p < 0.05$, Spearman's correlation coefficient). Also, the median number of pneumonia episodes during the diagnostic delay decreased from 5 in the 1980s, to 1 in the 1990s and to 0 in the period of 2001–2008 ($p < 0.05$, Spearman's correlation coefficient). While in the 1980s 17 of the 19 patients had pneumonia in

their past history, in the period of 2001–2008 only 13 of the 33 patients were concerned. **Conclusions:** Our observation documents improved awareness of immunodeficiencies among physicians. It is supposed that earlier diagnosis will prevent complications, improve the quality of life and even survival of hypogammaglobulinaemic patients.

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Introduction

Primary immunodeficiencies are a group of relatively rare diseases affecting a limited number of patients. They frequently face the situation that the appropriate immunological investigation is delayed although clinical symptoms raise the suspicion of primary or secondary immunodeficiency. This is particularly true for the comparatively frequent primary antibody deficiencies where recurrent and severe respiratory tract infections are often erroneously supposed to represent just only non-significant, common infections. Despite the fact that the information about symptoms in immunodeficiencies is broadly available, the diagnostic delay in hypogammaglobulinaemic patients is still relatively long and remains a significant problem [1].

Two major groups of primary hypogammaglobulinaemia exist. In the first group the symptoms begin after disappearance of placenta-transferred maternal immunoglobulins, usually after the 6th month of age. This group mainly includes X-linked agammaglobulinaemia (X-LA), autosomal recessive forms of inherited agammaglobulinaemia and various forms of immunodeficiencies with increased or normal IgM (hyper-IgM syndromes). In the second group the symptoms of immunodeficiency develop almost at any age. This group mainly includes common variable immunodeficiency (CVID) with a marked variability of the beginning of clinical symptoms from the age of 2, which is a suggested age limit for stating the diagnosis of CVID [2], to +80, with a mean of 26.3 years in one of the largest studies [3]. Contrary to this, in Good's syndrome, when hypogammaglobulinaemia is associated with the presence of thymoma, manifestations begin later with a mean of 56 years (range 41–75) in another study [4]. In all these diseases the most frequent initial manifestation includes recurrent and complicated bacterial infections of the respiratory tract typically with pneumonia episodes, the typical long-term complication being bronchiectasis formation [3].

The goal of this study was to evaluate how the situation in the diagnosis of primary hypogammaglobulinaemia had changed in a single referral centre during the last 28 years.

Methods

A retrospective analysis of the records of patients with primary hypogammaglobulinaemia ever followed up or diagnosed at the Department of Clinical Immunology and Allergology in Brno, Czech Republic, was performed. This department is a referral centre for the South Moravian region including approximately 2,237,000 inhabitants, but occasionally patients from other regions were also referred. The diagnosis of CVID was made on the fact of markedly decreased serum immunoglobulin levels (at least IgG and IgA); from the late 1990s the antibody response to polysaccharide and protein antigens was evaluated as well. The diagnosis of Good's syndrome was based on marked hypogammaglobulinaemia in a patient with thymoma. All other diagnoses were confirmed by adequate molecular genetic analysis when it became available.

Patients with severe combined immunodeficiency (8 patients diagnosed between 1996 and 2008), Nijmegen syndrome (diagnosed in 2 patients), Wiskott-Aldrich syndrome (3 patients), agammaglobulinaemia associated with 18 ring chromosome (1 patient) [5], and 1 patient with an unexplained combination of hypogammaglobulinaemia, pancytopenia, cutis laxa, Dandy-Walker syndrome and vascular abnormalities [6] were not included. Only in 1 patient with IgG₂ deficiency had the IVIG treatment been initiated, but owing to no effect on her increased frequency

of non-complicated respiratory tract infections and fatigue it was discontinued after 3 years; this patient was not included into the study either. Mild forms of hypogammaglobulinaemia in which immunoglobulin substitution was not initiated were also excluded from this study, such as patients with hypogammaglobulinaemia associated with lymphoid malignancies.

The clinical beginning of the immunodeficiency was supposed to be the time of the first pneumonia or the time of a marked increase in the frequency of respiratory tract infections – sinusitis, bronchitis or otitis media.

Serum immunoglobulin levels were determined by radial immunodiffusion (until 1993) turbidimetry (between 1993–1995) or nephelometry (since 1995).

The study was approved by the Ethics Committee of the St. Anne's Faculty Hospital in Brno under the protocol number 36V/2009.

Statistical Analysis

Normality of the observed values was tested using the Kolmogorov-Smirnov test. As the observed data significantly differed from the normal distribution, non-parametric tests were used. The testing of difference in continuous variables was performed using the Mann-Whitney test. For correlation analysis, the Spearman rank correlation coefficient was used. Categorical variables were analysed using Fisher's exact test. $p < 0.05$ was considered to be statistically significant.

Results

Characteristics of the Patients

Prior to 1981 a diagnosis of severe hypogammaglobulinaemia was made in 7 patients in various paediatric and internal medicine departments; they became consequently patients of our department but were not included in further computation because of lack of information about the early stages of the disease. Consequently, a diagnosis of X-LA (6 patients) and of CVID (1 patient) was made. Two of these patients are already dead.

Between 1981 (when the Department of Clinical Immunology in Brno was founded) and 2008, altogether 89 patients with severe forms of primary hypogammaglobulinaemia were diagnosed. The sample consisted of 77 patients with CVID, 4 patients with X-LA, 4 patients with Good's syndrome; the remaining diagnoses were μ -chain deficiency, CD40L deficiency, activation-induced cytidine deaminase deficiency (as previously described [7]) and non-X-linked hyper-IgM syndrome (as previously described [8]). Four of them were from a different region than South Moravia. Sixty-six of the whole sample (51 with CVID) were followed at our department at the time of completion of the study, so the computed prevalence of primary hypogammaglobulinaemia in South Moravia is 1:34,000 (1:44,000 for CVID). Twenty of the patients died

Table 1. Diagnostic delay and number of pneumonia episodes during diagnostic delay in hypogammaglobulinaemic patients referred between 1981 and 2008

Period	All patients			Patients with CVID or Good's syndrome		
	patients	diagnostic delay years	pneumonias	patients	diagnostic delay years	pneumonias
1981–1990	19	5 ± 9.6 (0–38)	5 ± 6.3 (0–25)	18	5.5 ± 9.7 (0–30)	5 ± 6.3 (0–25)
1991–2000	37	3 ± 11.7 (0–44)	1 ± 2.2 (0–10)	32	3.5 ± 12.3 (0–44)	1 ± 2.4 (0–10)
2001–2008	33	2 ± 6.6 (0–36)	0 ± 2.6 (0–15)	31	1.0 ± 6.7 (0–36)	0 ± 2.6 (0–15)

Expressed as median ± SD (with range in parentheses). Using the Mann-Whitney test, the following statistical significances were obtained:

All patients: diagnostic delay: 1980s vs. 1990s: $p = 0.628$; 1980s vs. 2000s: $p = 0.005$; 1990s vs. 2000s: $p = 0.035$; number of pneumonias before the diagnosis was made: 1980s vs. 1990s: $p < 0.001$; 1980s vs. 2000s: $p < 0.001$; 1990s vs. 2000s: $p = 0.019$.

Patients with CVID or Good's syndrome: diagnostic delay: 1980s vs. 1990s: $p = 0.723$; 1980s vs. 2000s: $p = 0.004$; 1990s vs. 2000s: $p = 0.018$; number of pneumonias before the diagnosis was made: 1980s vs. 1990s: $p < 0.001$; 1980s vs. 2000s: $p < 0.001$; 1990s vs. 2000s: $p < 0.032$.

and 6 of them moved to other regions of the Czech Republic.

Diagnostic Delay and Number of Pneumonias during the Diagnostic Delay

The diagnostic delay and number of pneumonias during the diagnostic delay are shown in table 1; figures for all patients and also for those with CVID and Good's syndrome are expressed. A separate calculation for CVID and Good's syndrome was performed because their manifestation usually starts in adulthood when the diagnosis of primary immunodeficiency is frequently (but erroneously) supposed to be less probable than in infancy and childhood. As shown in table 1, the length of the diagnostic delay gradually decreased in all 3 observation periods, although the difference between the 1980s and the 1990s was not significant. This decrease is also documented in figure 1, which shows a negative correlation between the time of diagnosis and the length of diagnostic delay. Also, the number of pneumonias during the diagnostic delay decreased, reaching a median of zero in the period 2001–2008, although even in this period there were patients who had undergone a significant number of pneumonias before the diagnosis of primary hypogammaglobulinaemia was made (table 1). Figure 2 shows a highly significant negative correlation between the year of diagnosis and the number of pneumonias prior to establishing the diagnosis of hypogammaglobulinaemia.

When enumerating the percentage of patients who had suffered any pneumonia before the diagnosis of hypogammaglobulinaemia, a significant decrease in those

patients was observed (fig. 3), while the significant increase in patients who were referred without a past history of serious or frequent respiratory tract infections is shown in figure 4.

There was a trend in the increase in the percentage of patients who had been referred with previously documented hypogammaglobulinaemia (6/19 patients in the 1980s, 20/37 patients in the 1990s and 18/33 patients in the 2000s), although it did not reach statistical significance (Fisher's exact test). Considering only patients with CVID and Good's syndrome (5/18 patients in the 1980s, 18/32 patients in the 1990s and 17/31 patients in the 2000s), we observed a significant increase in those patients when comparing the 1980s with the 1990s ($p = 0.037$) and the 1980s with the 2000s ($p = 0.046$, Fisher's exact test).

Referring Symptoms

Before 1980, all patients were referred for recurrent infections, but in 3 of them diagnoses of X-LA were made together (twice) or subsequently (once) with their affected brothers.

From 1981 to 1991, 17 of 19 hypogammaglobulinaemic patients were referred for 1 or more pneumonia episodes or frequent respiratory tract infections. One was referred for gonitis, but pneumonia was in her past history. One female patient with hyper-IgM syndrome was investigated because of marked lymphadenopathy, but in her past history an increased frequency of respiratory tract infections had also been noted.

Fig. 1. Diagnostic delay in hypogammaglobulinaemic patients referred between 1981 and 2008. Time trend in the length of diagnostic delay was estimated using the exponential regression method. Spearman's correlation coefficient $R = -0.359$ ($p < 0.001$).

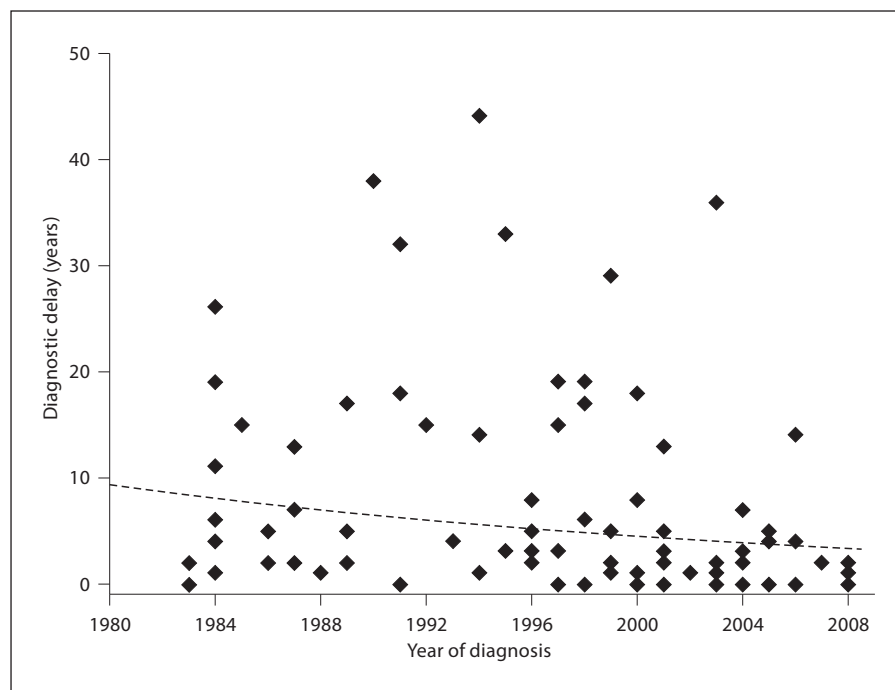
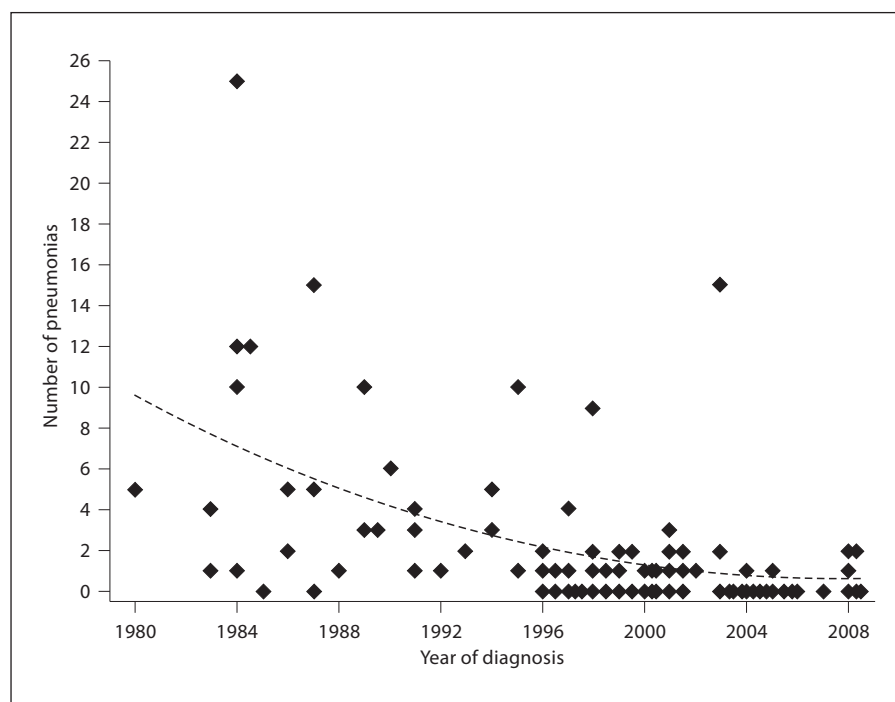


Fig. 2. Number of pneumonia episodes during diagnostic delay in hypogammaglobulinaemic patients referred between 1981 and 2008. Time trend in the number of pneumonias during the diagnostic delay was estimated using the polynomial regression method. Spearman's correlation coefficient $R = -0.520$ ($p < 0.001$).



From 1991 to 2000, 33 of 37 hypogammaglobulinaemic patients were referred for severe or frequent respiratory tract infections. In the remaining patients the referral diagnoses contained: 1 severe herpes zoster, 1 hepatopathy after delivery, 1 chronic fatigue (but 1 episode of

pneumonia was registered in the past history of the patient); in 1 patient CVID had developed from IgA deficiency. At that time we also diagnosed CVID and started IVIG treatment in 2 patients in whom 4 and 5 years prior to our investigation a diagnosis of hypogammaglobu-

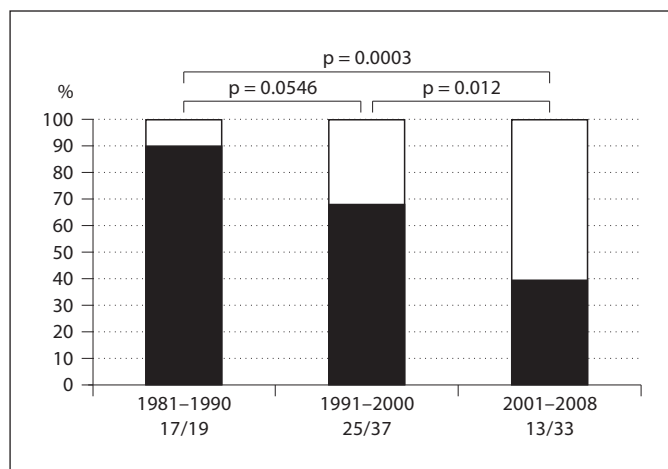


Fig. 3. Number of patients who suffered at least 1 episode of pneumonia during the diagnostic delay. Fisher's exact test was used for statistical comparison.

linaemia had been made, and immunoglobulin treatment with intramuscular immunoglobulin had been initiated but subsequently interrupted.

From 2001 to 2008, 24 of 33 hypogammaglobulinaemic patients were referred because of frequent or complicated respiratory tract infections. In the remaining patients the serum immunoglobulin levels were determined because of bronchial asthma (1), herpes zoster (1), alopecia areata (1), arthralgia (1), incapacity to determine blood group after delivery (1; the patient had had 1 episode of pneumonia in her past history), recurrent infectious parotitis (1) and general investigation of a patient with fatigue (1); in 2 cases a progression from IgA deficiency to CVID was documented.

Discussion

Notwithstanding that the first patient with humoral immunodeficiency was discovered more than 50 years ago [9], the general awareness of these diseases in the general medical population has markedly increased only in the last 20–30 years. Although our computed prevalence of primary hypogammaglobulinaemia is slightly lower than the estimated prevalence of humoral immunodeficiencies (1:25,000) [1], we believe that it reflects the real situation in our region. Even if in the present time the situation when a patient with symptomatic antibody deficiency dies before a diagnosis is made is probably extremely rare, the problem of a delay of immunological

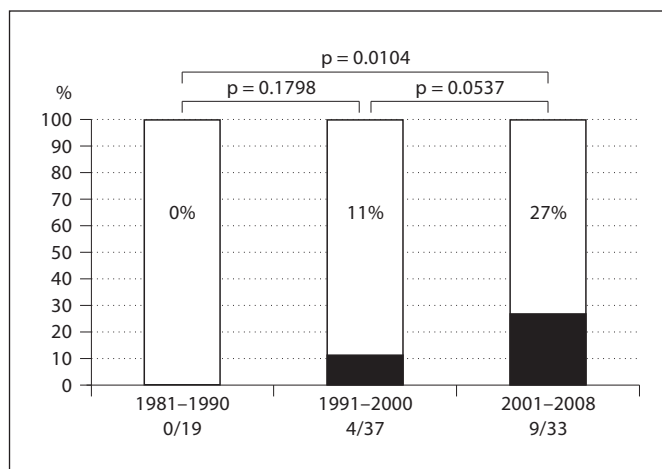


Fig. 4. Hypogammaglobulinaemic patients without severe respiratory tract infections at the time of diagnosis. Fisher's exact test was used for statistical comparison.

diagnosis, and consequently of a delay of adequate treatment, still exists, thus leading to increased morbidity of the patients [10]. On the other hand, the broad spectrum of manifestations different from infectious complications, for example, malabsorption, autoimmunity and lymphoid proliferation, together with wide availability of serum immunoglobulin determination, enabled the diagnosis and consequently the beginning of the treatment even before symptoms of severe or frequent respiratory tract infections occur.

The goal of this study was to assess how the diagnostic delay has changed during the last 28 years in a single referral centre, as well as how the referral diagnoses of those patients have changed.

Our results have shown that the diagnostic delay has markedly shortened; this change was most significant when comparing the period of 2001–2008 with the previous periods of 1991–2000 and mainly 1981–1990, while the difference between the 1980s and the 1990s was not significant in our study. The shortening of the diagnostic delay is in accordance with similar observations of other authors. Seymour et al. [11] analysed diagnostic delay in a single referral centre in Great Britain between 1989 and 1995, when the median was 3.5 years, compared to 1 year between 1996 and 2002. This delay was even worse in the same centre in patients referred prior to 1989, when the delay had been 5.6 years in adults and 2.5 in children [12]. It should be noted that the authors used a different definition of the clinical onset of the diseases. Also, in a French national study the median of the delay was 15.6

years in those patients with CVID in whom the symptoms had started before 1990 (138 patients), while it was 2.9 years in 114 patients with onset after 1990 [13]. A negative correlation of diagnostic delay and the year of diagnosis in CVID patients was recently observed in a study that included 388 patients from several European centres (also some of our patients were included) [3], and also in Iran [14]. An inverse relationship between the year of birth of patients with X-LA and the age when the diagnosis was made was documented in national surveys from the United States [15], Italy [10] and Iran [16].

It is supposed that a combination of various aspects influence the gradually improving situation in the early diagnosis of primary hypogammaglobulinaemia in our region, and probably in other countries as well. Improved education of medical students and graduated physicians in the subject of immunodeficiency is probably the most important and the most principal. The better availability of serum immunoglobulin determination mainly in the early 1990s probably resulted in the documented significant increased proportion of patients with CVID and Good's syndrome referred with previous serum immunoglobulin determination by their general practitioners or various specialists. The general awareness of patients of their clinical state and general knowledge about frequent infections as the symptom of immunodeficiency might be also taken into account.

Besides the previously documented shortening of the diagnostic delay, the presenting picture of hypogammaglobulinaemic patients has also changed. The most classical manifestation of immunoglobulin deficiency includes respiratory tract infections, among which pneumonias are frequent signs of failure of the immune system. The number of pneumonias occurring before the diagnosis was made can also be supposed as a marker of the depth of immunodeficiency during the diagnostic delay. This can also be documented in the group of our patients, as seen in table 1 and figure 2, which show a marked decline in the number of pneumonias before the diagnosis of primary immunodeficiency disease was made during the observation period. Also, the proportion of patients in whom no pneumonia was documented gradually increased, so that in the period of 2001–2008 more than half the patients with hypogammaglobulinaemia had not had any documented pneumonia before the diagnosis was made. On the other hand, we diagnosed 1 patient in 2003 with a history of approximately 15 episodes of pneumonia; his diagnostic delay was 36 years, which shows the continuous effort of education of doctors in the manifestation of immunodeficiencies.

As mentioned previously, recurrent/severe respiratory tract infections are the most typical clinical manifestation in patients with primary humoral immune deficiency. As our study documents, the proportion of patients who were referred for reasons different from respiratory tract infections has significantly increased. As documented, the presenting symptoms included either some other infections – gonitis, severe herpes zoster or also conditions unrelated to infections – alopecia areata, bronchial asthma, incapacity to determine blood group and progression from IgA deficiency.

In 3 of our patients, previously followed for selective IgA deficiency, CVID developed. This is a well-established and repeatedly described situation [17, 18]. In all these patients a significant decrease in IgG together with a markedly disturbed response to both polysaccharide (non-conjugated 23-valent vaccine) and protein (tetanus toxoid) was documented, even at the time when none of those patients experienced pneumonia and the frequency of minor respiratory tract infections was only mildly increased. There is a discussion on whether it is worth it to follow up patients with selective IgA deficiency, especially if they do not have significant clinical problems. Our experience supports the idea of the value of follow-up for IgA-deficient patients, even if they do not have any clinically significant manifestations.

In conclusion, our retrospective study shows improved awareness of immunodeficiency symptoms in the general medical population, leading to earlier referral of the patients for immunological investigation. It is supposed that earlier treatment will improve the life expectancy of the patients. Despite this success there have been patients even in this decade in whom the diagnostic delay has been inadequately long.

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