

Geographical Differences in Autoantibodies and Anti-infectious Agents Antibodies Among Healthy Adults

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Abstract Much is known about the geoeidemiology of defined autoimmune diseases (AD); however, there is currently limited data regarding the prevalence of autoantibodies among healthy populations of different geographical areas. The aim of this study was to evaluate a large profile of autoantibodies in healthy adults from distinct global regions as well as the prevalence of anti-infectious agents antibodies in those regions. Sera samples from 557 healthy donors were obtained at six centers located in different countries (i.e., Italy, Netherlands, Israel, Mexico, Columbia,

Papua New Guinea (Kitavans)). Sera were tested for the presence of antinuclear antibodies (ANA) and autoantibodies associated with thrombophilia, vasculitis, and gastrointestinal (GI) disease. Sera samples were also screened for antibodies against infectious agents (i.e., EBV, CMV, HBV, *Helicobacter pylori*, *Treponema pallidum*, and *Toxoplasma gondii*). Tests were performed using the BioPlex 2200 or ELISA kits (Bio-Rad Laboratories, USA). We found a significant gradient of ANA positivity among the groups: 45% of Columbians, 38% of Kitavans,

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26% of Mexicans, 12% of Italians, 12% of Dutch, and 11% of Israelis were ANA positive. Geographical differences were also observed regarding the prevalence of specific autoantibodies, namely ANA: anti-dsDNA, chromatin, SmRNP, Ro/SSA, La/SSB, Scl70; GI associated: antigliadin; and thrombophilia-associated: anti- β 2GP1 and prothrombin. Additionally, significant differences were observed regarding serological markers of all infectious agents screened. The observed variance between healthy ethno-geographical distinct populations in prevalence of autoantibodies may represent different genetic or environmental (e.g., prior exposure to infection) influences. Thus may illuminate possible causes of geoepidemiological differences in AD.

Keywords Autoimmunity · Geoepidemiology · Ethnicity · Antinuclear antibodies · Antigliadin · Antiphospholipid antibodies · Infectious agents

Introduction

Autoantibodies and Autoimmunity

The detection of serum autoantibodies has had a central role in the diagnosis and classification of autoimmune diseases (AD) for over 50 years. Some autoantibodies also have a prognostic significance or are used as markers for disease activity. In recent years, it is becoming clear that asymptomatic individuals may carry autoantibodies for many years prior to clinical manifestation of an established AD (Table 1), and that serological detection of these antibodies has a strong predictive value [1–4].

For instance, in systemic lupus erythematosus (SLE) the appearance of antibodies to RNP, Sm, dsDNA, cardiolipin, Ro (SSA), and La (SSB) can precede clinical diagnosis by 7–10 years with a positive predictive value (PPV) of 94–100% [5]. In rheumatoid arthritis (RA), antibodies for rheumatoid factor or anticyclic-citrullinated-peptide (anti-CCP) were detected in serum samples obtained a median of 4.5 years before disease onset, with a PPV of 100% [6]. The presence of autoantibodies is also used for the prediction of future type 1 diabetes mellitus (T1D) [7, 8].

The predictive capacity of AD made possible by serological detection of autoantibodies in the preclinical stage may have a crucial role in disease prevention. Several lines of evidence (e.g., in SLE, T1D, and RA) suggest that intervention in the preclinical period may modify future AD risk [9–11]. Moreover, prediction is especially important when the disease progression can be prevented or postponed by avoiding environmental factors, among them infectious agents, which may trigger or worsen autoimmunity [1, 12–14].

Geoepidemiology and Autoimmunity

Geoepidemiology of AD portrays the burden of these illnesses across various regions and ethnic populations, and yields important clues regarding their genetic and triggering environmental mechanisms. Uneven global distribution of certain AD, on the one hand suggests that population genetics have an important role in disease development. On the other hand, genetic factors cannot completely account for the phenomenon, and it is only by combining evidence of environmental factors that the geoepidemiological picture can be elucidated [15, 16]. The recent Congress of Autoimmunity in Slovenia highlighted environmental factors in geoepidemiology and the reader is directed to the publications that emphasize this issue [16–57].

Table 1 Predictive autoantibodies in autoimmune diseases [2]

Disease	Autoantibodies
Type 1 diabetes mellitus	anti-GAD IA-2 IAA
RA	RF (anti-IgG) Anti-CCP
SLE	Anti-PL Anti-Ro Anti-La Anti-dsDNA Anti-Sm Antinuclear ribonucleoprotein Anti-HS Antinucleosome Antihistone Antiribosomal P protein
Sjogren's Syndrome	Anti-SSA(Ro) Anti-SSB (La)
APS	Anticardiolipin Lupus anticoagulant
PBC	Anti-gp120 Anti-PDC
AIH	ANA
Crohn's disease	ASCA
UC	pANCA
Addison's disease	ACA (anti-21-hydroxylase)
AITD	Anti-TG Anti-TPO
Pemphigus	Antidesmoglein 1

GAD glutamic acid decarboxylase, *IA-2* protein tyrosine phosphatase-like molecule, *IAA* insulin autoantibodies, *RF* rheumatoid factor, *CCP* cyclic citrullinated peptide, *PL* phospholipids, *HS* heparan sulphate, *PDC* pyruvate dehydrogenase complex, *ANA* antinuclear antibodies, *ASCA* anti-*Saccharomyces cerevisiae* antibodies, *pANCA* perinuclear antineutrophil cytoplasmic antibodies, *ACA* adrenal cortex cells antibodies, *TG* thyroglobulin, *TPO* thyroid peroxidase

Unlike the case in several defined AD, data is scarce regarding ethno-geographic differences in prevalence of autoantibodies among healthy individuals, currently limiting our understanding of their predictive utility across different populations (e.g., differential specificity). Moreover, by investigating the prevalence of a variety of autoantibodies and concomitantly infection exposure, in distinct healthy populations, one may reveal unique ethno-geographical interactions. The aim of the present study was thus to investigate the prevalence of a variety of autoantibodies as well as the frequency of anti-infectious antibodies in six different healthy populations.

Materials and Methods

The present study is observational and of comparative cross-sectional design.

Subjects The sera of 557 healthy volunteers (age and sex matched between groups) were collected in six centers located in different countries. The healthy subject groups comprise of: 100 subjects from Italy, 98 from Israel, 50 from Mexico, 49 from the Netherlands, 140 from Columbia, and 120 from the indigenous population of the island of Kitava in Papua New Guinea (PNG).

Autoantibodies Profile A large profile belonging to four groups of autoantibodies has been tested in the sera:

Antinuclear antibodies (ANA IgG): Anti-dsDNA, Sm, chromatin, ribosomal-P, RNP, SmRNP, Ro /SSA, La/SSB, centromer, Scl-70, and Jo-1.

Of note, subjects exhibiting serological positivity to at least one antigen of the entire ANA analytes panel above were considered to be “ANA positive”. Subjects exhibiting serological positivity to at least one of the extractable nuclear antigens group (ENA: anti-Sm, RNP, SmRNP, SSA, SSB, Scl-70, and Jo-1) were considered “ENA positive”.

Vasculitis associated (IgG): Antiglomerular basement membrane, proteinase 3, and myeloperoxidase.

Gastrointestinal associated (IgG, IgA): Anti-*Saccharomyces cerevisiae*, gliadin, and tissue transglutaminase.

Thrombophilia associated (IgG and IgM): Anticardiolipin (CL), β_2 glycoprotein 1 (β_2 GP1), complex of both, phosphatidylserine- β_2 GP1 complex, phosphatidylethanolamine, prothrombin (PT), and phosphatidylserine-prothrombin complex.

Infectious Agents Exposure to various infectious agents has been assessed by testing sera for presence of IgG antibodies directed to viral infection: hepatitis B (IgG and IgM),

cytomegalovirus (CMV), Epstein–Barr virus (EBV); bacterial infection: *Helicobacter pylori* and *Treponema pallidum*; and parasitic infection: *Toxoplasma gondii*.

Laboratory Testing The various antibodies have been tested by one of two methods. For both methods utilized in this study, the positive sera were calculated according to manufactures' equations for cut off value determination.

Multiplexed Assay All autoantibodies (IgG, IgM, and IgA types), as well as (IgG) anti-infectious antibodies directed to toxoplasma, treponema, CMV, EBV nuclear antigen 1, EBV viral capsid antigen (VCA), and EBV early antigen diffuse, were tested utilizing the Bio-Rad BioPlex 2200 system (Bio-Rad Laboratories, Hercules, CA), a fully automated random-access analyzer built on a synthesis of multiplex, magnetic beads, and flow cytometry technologies, as previously described [58–60].

Enzyme-linked immunosorbent assay Antibodies directed to *H. pylori* (IgG) and hepatitis B virus core (IgG and IgM) were tested by PYLORI DETECT IgG and MONOLISA anti-HBc PLUS enzyme-linked immunosorbent assay (ELISA) kits, respectively (Bio-Rad Laboratories, CA, USA), according to manufacturer's protocols as described elsewhere [61].

Statistical Methods The StatSoft-STATISTICA (8.0) software was used for statistical analysis. Pairwise comparisons between the groups were performed by Chi-square test or Fisher's exact test (as appropriate) for categorical variables (e.g., antibody positivity frequencies). The false discovery rate method was used for correction of the multiple comparisons. For continuous variables (e.g., antibody levels) Duncan's post hoc test was used for pairwise comparisons between all groups. Statistically significant results were considered at $p < 0.05$. Due to the extensive application of multiple comparisons correction in this investigation, all post correction comparisons reaching statistical significance are expressed as $p < 0.05$ (i.e., original, pre-correction p values were disregarded).

Results

Differences in Prevalence of Various Autoantibodies

Variance between the groups from the 6 regions was evident in several specific autoantibodies.

Antinuclear Antibodies (IgG) There was a clear gradient of general ANA positivity among the groups: 45% of

Columbians, 38% of Kitavans (PNG), 26% of Mexicans, 12% of Italians, 12% of Dutch, and 11% of Israelis were ANA positive. The rate among Columbians was significantly elevated when compared with all other subject groups ($p < 0.05$), followed by PNG ($p < 0.05$, for comparisons with Italy, Netherlands and Israel), and Mexico ($p < 0.05$, for comparison with Israel). Two specific ANAs were especially notable. Anti-dsDNA antibodies were particularly frequent among Columbians, followed by PNG subjects, compared with the rest of the subject groups (Fig. 1; $p < 0.05$). Antichromatin antibodies were detected in high proportion of Columbian subjects, while detected in comparably low proportion among all other subject groups (Fig. 1; $p < 0.05$).

We further detected general ENA positivity in 15% of Kitavan, 14% of Mexican, 9% of Columbian, 6% of Dutch, 5% of Israeli, and 4% of Italian subjects. However, this gradient did not reach statistical significance after multiple comparisons adjustments ($p < 0.05$, only for comparison between Papua New Guinea and Israel). There was also a trend (NS) of comparatively high frequencies of several specific antibodies directed to ENAs among Mexicans, namely anti-SmRNP, Ro/SSA, La/SSB, and Scl70 (Fig. 1). The mean levels of antibodies directed to these four ENAs were significantly higher in sera of Mexicans when compared with all other subject groups ($p < 0.05$). In contrast, frequency or levels of anti-Sm, RNP, Centromere, Jo-1, and Ribo P antibodies did not differ across the groups, as these were generally detected in a negligible proportion of subjects.

Vasculitis Associated (IgG) The occurrence of these antibodies was rare across all healthy subject groups from the different regions (Table 2). No significant variation was observed.

Gastrointestinal Associated (IgG and IgA) Were tested in three groups of healthy subjects from different regions, namely Italy, Israel, and Columbia (prevalence data presented in Table 3). Out of the six analytes compared, only antigliadin of the IgG type exhibited significant intergroup variance, as it was more frequently detected among Israeli and Italian subjects than among Columbian subjects ($p < 0.05$).

Thrombophilia Associated (IgG and IgM) A large profile was tested in the Italian and Columbian subject groups only, presented in Fig. 2. Two IgM type antibodies were significantly more prevalent among Columbians, namely anti- β 2GP1 and PT ($p < 0.05$ for both).

Differences in Prevalence of Various Anti-infectious Agents Antibodies

We found variance between the groups in prevalence of every one of the eight anti-infectious antibodies (IgG) tested (prevalence data presented in Fig. 3). Variance between the regions was especially prominent in positivity rates of *T. pallidum* antibodies, as 87% of subjects from Papua New Guinea tested positive while the frequency in all other groups was comparably low (range, 0–4%; $p < 0.05$). The highest prevalence of anti-*H. pylori* antibodies was recorded among Columbian subjects (81%) compared with all other groups (range, 30–62%; $p < 0.05$; we did not have data for Papua New Guinea). Anticytomegalovirus antibodies were detected in highest frequency among subjects from Papua New Guinea (100%) when compared with all five other groups of subjects (range, 33–93%; $p < 0.05$). Relatively high prevalence was also detected among

Fig. 1 Prevalence of antinuclear autoantibodies in six groups of healthy subjects. $\star p < 0.05$, significant differences in prevalence and/or in serum levels of these antinuclear antibodies were evident between healthy subject groups

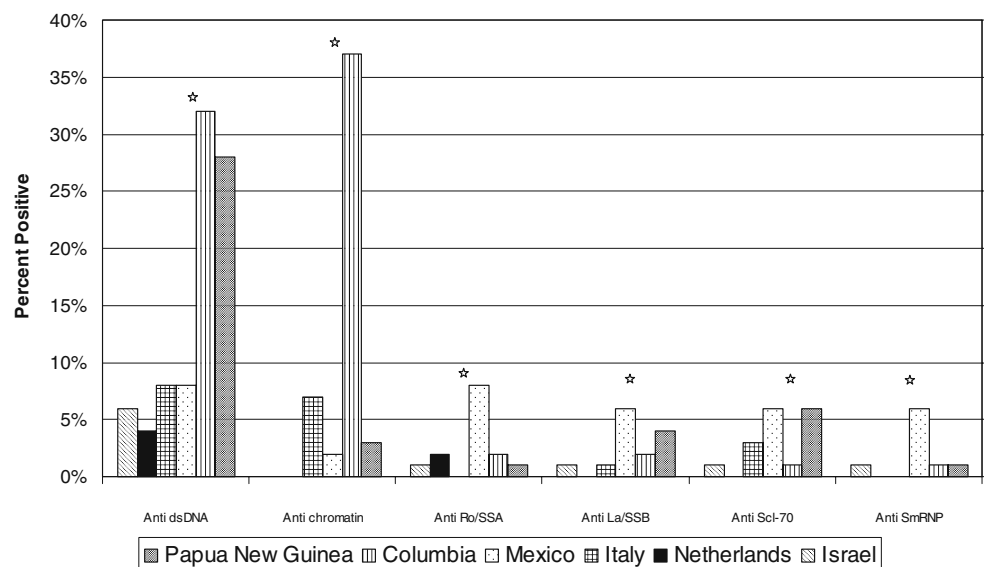


Table 2 Prevalence of vasculitis associated autoantibodies in six groups of healthy subjects

Vasculitis associated autoantibody (IgG)	Italy (%)	Netherlands (%)	Israel (%)	Mexico (%)	Columbia (%)	Papua New Guinea (%)
Anti-GBM	0	0	0	0	0	2
Anti-PR3	1	0	1	0	1	0
Anti-MPO	0	2	0	0	1	0

GBM glomerular basement membrane, PR3 proteinase 3, MPO myeloperoxidase

Columbian subjects. Conversely, prevalence in subjects from the Netherlands was lowest of all six groups (Fig. 3; $p<0.05$). The presence of antibodies directed to EBV early antigen was also highly variable (range, 8–41%), demonstrating a pronounced gradient (Fig. 3). The highest frequency was registered in the Mexican group ($p<0.05$ for comparison with all groups except Netherlands), and lowest frequency in the Colombian group ($p<0.05$ for comparison with all groups except Italy). The prevalence of antibodies directed to EBV VCA (range, 80–98%) and nuclear antigen 1 (EBNA; range, 72–90%) did not exhibit as prominent a geographic gradient (Fig. 3), nonetheless differences were noted. The highest prevalence of anti-EBV VCA was registered in the Mexican subjects, reaching statistical significance in the comparison to subjects from Israel and the Netherlands ($p<0.05$). Mean serum levels of these antibodies was significantly higher in Mexican subjects compared with groups from all other regions ($p<0.05$). Antibodies directed to EBNA were comparably frequent in the Mexican, Italian and Papua New Guinea subjects, and less frequent among the Colombian subjects ($p<0.05$). The frequency of antibodies directed to *T. gondii* parasite was lowest among subjects from Israel and the Netherlands compared with all other groups (range, 15–43%; $p<0.05$). The mean bacterium antibody levels were also significantly high in sera of Mexican subjects (as well as in Columbians) compared with the other groups ($p<0.05$). Finally, antihepatitis B antibodies were detected in highest frequency among the Colombian subjects (range, 0–11%, we did not have data for Papua New Guinea). However, this reached statistical significance ($p<0.05$) only in comparison to subjects from the Netherlands and Mexico.

Discussion

In the present study, we have compared the frequencies of a large profile of autoantibodies across several healthy, ethno-geographically distinct populations. Unsurprisingly, the majority of the autoantibodies tested negative across most of the subjects in all six geographical groups, subsequently exhibiting no ethno-geographic variability. Nevertheless, we report a significant variability in some particular autoantibodies, warranting further discussion.

The most prominent intergroup variation was shown in antinuclear antibodies. The frequency range of ANA-positive sera was considerable, from approximately 12% comparably in the European groups (including Israel) and up to 26%, 38%, and 45% in the Mexican, PNG, and Columbian subjects (respectively).

There are several discrete reports (e.g., [62, 63]) providing rates of ANA positivity among geographically homogenous healthy populations (i.e., non-comparative point prevalence). The existing data has been recently examined by the American College of Rheumatology Ad Hoc Committee on Immunologic Testing Guidelines, which concluded that ANA can be detected (by immunofluorescence (IIF)) in 5–30% of healthy subjects depending on the dilution of sera (1:40–1:160, respectively). This seroprevalence rises with increasing age and particularly in women [64].

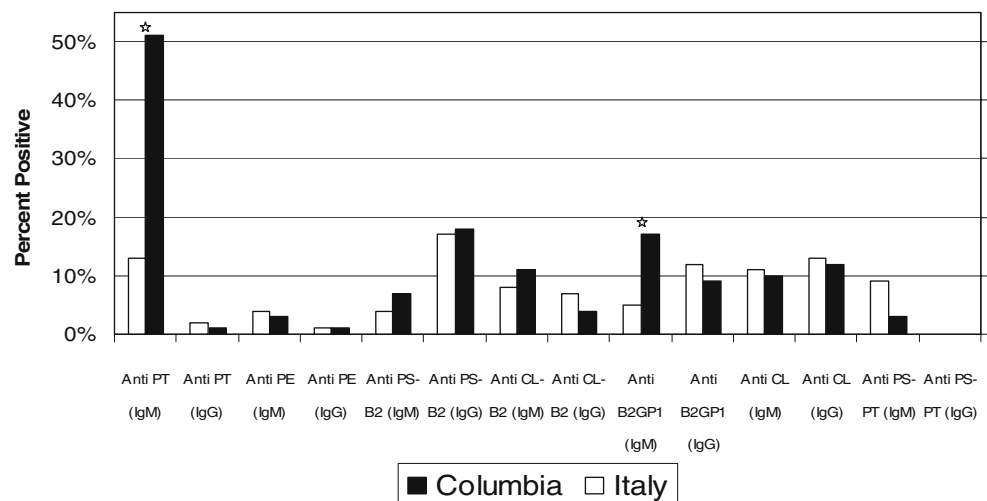
The frequency of ANA in different healthy populations evaluated as controls was reported sporadically utilizing IIF or other methods (e.g., ELISA). However, the detection of autoantibodies was performed in each study employing different techniques, different dilutions and in different laboratories. In the current study we have utilized the same

Table 3 Prevalence of gastrointestinal associated autoantibodies in 3 groups of healthy subjects

Gastrointestinal associated autoantibody	Italy (%)	Israel (%)	Columbia (%)
Antigliadin (IgG)	11	10	2 ^a
Antigliadin (IgA)	2	1	4
Antitissue transglutaminase (IgG)	2	0	0
Antitissue transglutaminase (IgA)	0	2	0
Anti- <i>Saccharomyces cerevisiae</i> (IgG)	1	0	4
Anti- <i>S. cerevisiae</i> (IgA)	2	0	2

^a Higher frequency of antigliadin (IgG) in Italy and Israel subject groups when compared with the Columbia subject group ($p<0.05$)

Fig. 2 Prevalence of thrombophilia-associated autoantibodies in healthy subjects. *PT* prothrombin, *PE* phosphatidylethanolamine, *PS-B2* phosphatidylserine- β 2GP1 complex, *CLB2* cardiolipin- β 2 glycoprotein 1 complex, *CL* cardiolipin, *PS-PT* phosphatidylserine- prothrombin complex. $\star p < 0.05$



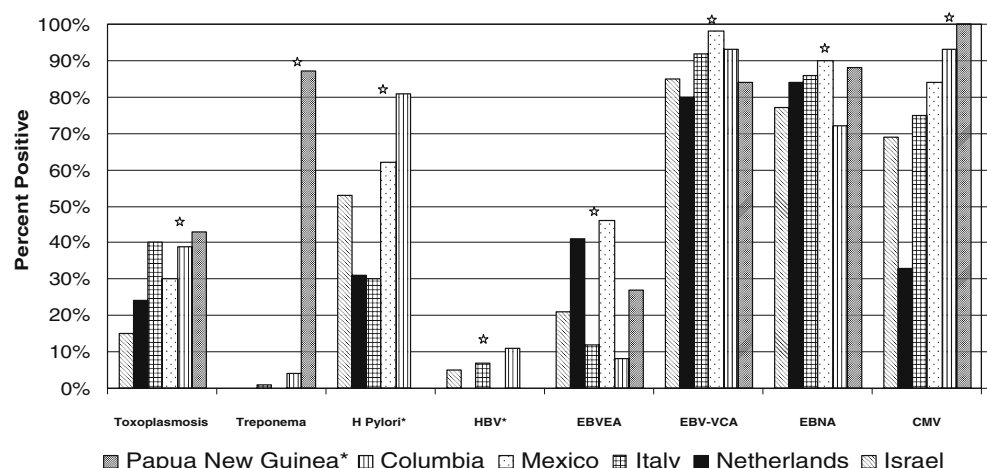
testing method across all six groups of healthy subjects, therefore in a sense rendering comparisons across different cohorts plausible, and suggesting that the observed differences account for true ANA detection variance rather than technical bias. The multiplexed assay we employed, namely the BioPlex 2200 ANA Screen, has been previously validated by our group as well as by others [59, 65]. In a total of 510 healthy European blood bank donors, we have previously found that the frequency of positive results was 7.5%, 5.5%, and 15.5% obtained by using BioPlex 2200 ANA Screen, ANA ELISA, and IIF, respectively [59]. A more recent evaluation of the BioPlex 2200 ANA Screen in a European hospital population found that 221 out of the 1,104 sera (20%) were ANA positive. The most frequently detected autoantibody in this cohort was antichromatin [65]. The conclusions from both evaluations were that this multiplexed ANA screen is of comparable specificity to other routine tests (namely ELISA and double immunodiffusion),

has a lower positive rate than IIF, and is suitable as a sensitive screening test to confirm or to exclude the presence of a large number of autoantibodies simultaneously and rapidly [59, 65].

Truly detected geographical differences notwithstanding, it is challenging to explain the exceptionally high ANA positivity rates herein observed among the non-European populations. The soaring ANAs rates we found in the Colombian subjects group are attributable to the elevated prevalence of two specific autoantibodies, namely anti-chromatin and anti-dsDNA. The latter was also the most prominently prevalent ANA in subjects from PNG. In the Mexican subject group, it was in fact several antibodies directed to ENAs which seem to have elevated the overall ANA positivity.

On the one hand, given the strong predictive value of autoantibodies regarding the future progression (and in some cases prognosis) of an established AD, it may be

Fig. 3 Prevalence of anti-infections antibodies in six groups of healthy subjects. *H* helicobacter, *HBV* hepatitis B virus, *EBVEA* Epstein-Barr virus early antigen, *EBV-VCA* viral capsid antigen, *EBNA* nuclear antigen, *CMV* cytomegalovirus. $\star$, *Helicobacter pylori* and HBV—no data for Papua New Guinea subjects group. $\star p < 0.05$, significant differences in prevalence of these anti-infections antibodies were evident between the subject groups



hypothesized that the detected ANA gradient should in part manifest true predictive risk of AD in these populations. Moreover, it implies that autoantibodies screening specifically tailored to different populations may be a valuable tool for future AD risk stratification.

However, while most patients with an established AD have marking autoantibodies, most people with autoantibodies do not have and likely will never have an overt AD. Therefore, one must consider that while on the one hand geographical differences in autoantibodies detection may manifest true AD risk variance, on the other hand unique non-pathological serological mechanisms should also contribute to the detection rates. In fact, both alternatives together may ultimately account for extraordinarily high autoantibodies (e.g., ANA) detection rates in sera of healthy subjects from these particular regions.

The intriguing relationship between the mechanisms of *molecular mimicry* and *cross-reactivity* may, in theory, hold the key. In recent years, many investigations into the pathogenesis of autoimmunity elaborated on the definition of these mechanisms [66–68]. For example, several human monoclonal antibodies were found to react specifically with infectious agents as well as with human antigens [13, 69]. These antibodies could represent merely cross-reactivity that will not result in any clinical or pathological outcome, or alternatively, they can initiate a pathogenic process that may eventually result in autoimmunity, hence defining molecular mimicry. The process of molecular mimicry between microbial and self-components (i.e. proteins, carbohydrates or DNA epitopes) is postulated to be the most likely mechanism of post-infectious autoimmunity [68–73].

In fact, infectious agents have been repeatedly and for a long time implicated as a major environmental trigger of AD in general, and in the induction of autoantibodies specifically [13, 14, 60, 68–79]. In addition to molecular mimicry, there are other underlying mechanisms which are complex and vary with different agents; among them are potentiation and diversion of innate immunity, and exposure or spillage of intracellular autoantigens [3, 14]. There is a wide unevenness in the geographic distribution of infectious agents, thus it seems plausible that in interaction with population genetics the infection gradient should influence global gradients of AD and autoimmune manifestations (e.g., autoantibodies) [15, 16, 80, 81].

In the current study we have indeed identified a generally elevated evidence of infection exposure (i.e., anti-infectious agents antibodies) in the subject groups from the Latin American region (i.e., Columbia and Mexico) and in the indigenous Kitavan population (from PNG), when compared with the European and Israeli subject groups. It may be suggested, that in these populations the elevated infection exposure over lifetime should indeed have a significant role in the elevated detection of autoantibodies.

Furthermore, the particular combination of infectious agents prominent in each population may account for the distinct autoantibody profiles detected in these groups.

One salient example, in the current study the frequency of anti-*Treponema* antibodies was exceptionally high among the (PNG) Kitavans (87%), while almost undetectable in all other groups. The geographical region of PNG is indeed known to be endemic to a subspecies of *T. pallidum* and *Treponema pertenue*, which causes Yaws disease [82]. We and Frostegård J et al. have previously studied the same individuals from PNG, and found elevated levels of antiphosphorylcholin antibodies in 100% of the Kitavans, plausibly induced by the treponemal exposure [83–85]. Ultimately, the high presence of antiphosphorylcholin antibodies may in turn be responsible for the high rates of anti-dsDNA antibodies observed in the same population, on account of cross-reactivity between these two autoantibodies [84, 86].

Taken together, the observed link between generally high infection exposure and elevated ANA prevalence among Columbians, Kitavans and Mexicans, perhaps represents molecular mimicry implying future AD risk, and/or detection due to serological cross-reactivity unique to each population.

In the present study three additional specific autoantibodies exhibited marked intergroup differences, namely antigliadin (IgG), IgM β 2GPI, and IgM PT, meriting a brief elaboration. The current model of the natural history of celiac disease (CD) recognizes that in many cases there is a preclinical stage, in which there are no intestinal and/or extraintestinal symptoms (including no findings in a small bowel biopsy), however autoantibodies can often be detected [87]. Several series conducted in Europe and the US have reported a rather comparable prevalence of around 0.5% of preclinical CD (based on presence of autoantibodies alone), while the estimates of combined preclinical and diagnosed (i.e., including clinical disease) ranged between 0.5–2.5% in the different populations. In Africa, and in East Asian populations who lack the disease associated HLA haplotype, the disease is considered to be rare [87]. Our finding of approximately fivefold lower antigliadin antibodies in Columbians than in Israelis and Italians seems to be rather in agreement with a US population based series reporting a three-fold lower incidence of a positive serological test to CD in Hispanics than in non-Hispanic whites [88]. In fact, it has been suggested that this may be attributable to the low frequency of the HLA-DR3, DQB1*0201 haplotype in the studied Hispanic population [88]. Genetic determination notwithstanding, we previously reported that certain infections, CMV among them, may have a protective role in the etiopathogenesis of CD [89]. We therefore propose that in the present study the elevated exposure to several infectious agents (including CMV) in the healthy Columbians group may in this case have a protective role in the appearance of disease associated antibodies.

Nonetheless, while such an association may be hypothesized, a protective (causal) relationship may not be established based on this observational report.

Antiphospholipid antibodies (aPL) were previously reported in 1–5% of asymptomatic healthy subjects, with a highly variable point prevalence when comparing different reports from various ethno-geographic populations [21, 90, 91]. The variability has been attributed to both genetic and environmental factors [21, 90]. We herein report a significantly higher frequency of IgM anti- β 2GPI and anti-PT antibodies in Columbian compared with Italian healthy individuals. These specific aPL account for most of the global antiphospholipid binding activity [92, 93]. aPL have been shown to be induced by many viral, bacterial and parasitic infections [94]. We have previously detected elevated levels of EBV early antigen IgG, toxoplasma IgM, and rubella IgM antibodies in sera of primary APS patients compared with controls [95]. Although in the present study we did not find differences in prevalence of EBV early antigen IgG or toxoplasma (IgG) between Columbians and Italians (rubella was not tested), other infectious or environmental influences (herein not tested) may not be ruled out.

Conclusions

Of note, due to the investigative nature and broad scope of this study, we did not apply a specific analysis of the numerous demographic variables which may serve as confounders in the observed intergroup variability. Nonetheless, we applied the same valid laboratory method across geographically distinct, age and sex matched populations, thereby revealing marked geoepidemiological variance in autoantibodies positivity and infectious agents exposure. Our results warrant further prospective investigations to establish whether indeed the AD rates will eventually be elevated in regions found to be more prevalent in autoantibodies. This should ultimately validate the predictive value of population-specific autoantibodies screening, and help establish unique causal factors. Furthermore, in the ongoing quest of autoantibodies testing methods standardization, it is becoming clear that population variance should be significantly factored in.

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