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What caused all these troubles, anyway? Epstein Barr virus in Sjögren's Syndrome re-evaluated

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The inflammatory autoimmune and autoinflammatory rheumatic diseases share the persistent, pernicious, and currently depressing feature that they are largely idiopathic. Their true environmental origins remain unknown - or at least there is no general convincing and agreed upon consensus that explains their causation – and this is despite the extraordinary efforts made to date and the awesome capabilities of modern science. Of course, there are exceptions to such a gross generalization. Knowing that the host response to *Streptococci* is responsible for rheumatic fever, for example, seems to have contributed to our therapeutic success with this disease and its demise as a public health scourge, despite the many important details that remain unexplained about the relationship of this bacterium to the host response and the vagaries of disease expression.

In more recent times, we have revolutionized the therapy of many rheumatic diseases without knowing how these diseases are initiated. Indeed, our situation with autoimmune and autoinflammatory rheumatic diseases is reminiscent of the history of antibiotics, many of which were discovered decades before an understanding of how they were differentially toxic to bacteria in preference over toxicity to the host. As a consequence, our 21st century rheumatoid arthritis patients do not have the "natural history of disease" experience that was the fate of the vast majority of our 20th century patients. Having therapies that change the suffering from this unwanted disease process is an extraordinary achievement, but with an incomplete description of the steps leading from health to disease, we operate with the handicap of conceptual darkness.

In April 2014, the PubMed website lists 117,077 studies of rheumatoid arthritis; 56,984 on systemic lupus erythematosus; 22,262 on systemic sclerosis; 14,178 on ankylosing spondylitis; and 12,513 on Sjögren's syndrome, yet there is no consensus in our community of investigators for the original environmental causes of these disorders. This is a brutal truth, despite a multitude of animal models, genetics galore, and the mass of molecular

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insights that begin to decipher the incredibly complicated molecular and cellular interactions that govern cellular responses.

We are awestruck by all the work that is being done for how these diseases unfold, how they are expressed, or how they respond to the latest invented therapy. And yet the relative failure to emphasize an understanding of their beginnings, from investigators and funding agencies, despite the estimated 100,000,000 afflicted worldwide by only the disorders cited above, is incomprehensible. Identifying the origin(s) of these disorders would provide a yet unknown foundation upon which to structure the tens of millions of hours of work behind the 193,633 publications listed above. In our view there is little, except a miraculous, inexpensive cure that could possibly be more important to uncover or would empower us more to make greater progress in our struggle to diminish the suffering these diseases cause.

Some herpes viruses infect the host for life. In the chronic latent stage they are among the "usual" suspects for etiological roles in pathogenesis of idiopathic chronic inflammatory disorders. They percolate under the limits of detection of the innate and adaptable systems built to identify and destroy them. Yet, the latent and emerging lytic phases are, in concert, incredibly immunogenic. For example, Epstein-Barr virus (EBV, HHV-4) commands ≥2% of the T cell repertoire in normal Epstein-Barr virus infected individuals (1, 2). With the tens of thousands of organisms against which the ordinary person must continuously defend themselves, this is an astonishingly high proportion of the host's T cell repertoire.

Many naively argue that these herpes viruses, Epstein-Barr virus in particular, are too ubiquitous to cause such rare, devastating diseases. However, we know that infectious mononucleosis and Burkitt's Lymphoma, both uncommon compared to the number of humans infected worldwide, are indeed caused by Epstein-Barr virus infection. Specifically, work has been done to correlate and mechanistically link Epstein-Barr virus to a number of autoimmune diseases including rheumatoid arthritis, lupus, Sjögren's syndrome (3), multiple sclerosis (4) and even Parkinson's disease (5).

Of note, much work now links Epstein-Barr virus to the etiology of lupus with over 150 papers to date devoted to the topic. Indeed, there is an increased prevalence of Epstein-Barr virus infection in pediatric lupus patients compared to control pediatric subjects, a consistent and predictable course of anti-Epstein-Barr virus antibody-to-autoantibody development through molecular mimicry, and an ever present anti-Epstein-Barr virus immune dysregulation in lupus patients (6–8).

Sjögren's syndrome is so closely related to systemic lupus erythematosus that a shared etiology would be plausible. However, the effort to make such a Sjögren's syndrome / Epstein-Barr virus link has resulted in conflicting reports over the past few decades (9–15). In this issue of *Arthritis and Rheumatology*, Bombardieri and colleagues (16) explore compelling evidence of this herpes virus/auto-immune system interplay and present tantalizing data that, along with the many studies in lupus, bolster the idea that immune responses to Epstein-Barr virus and self arise in concert.

The current study focuses on structures of the salivary gland called ectopic lymphoid structures, a formation relatively unique to Sjögren's patients, which mimic many

characteristics of B and T cell follicles allowing for a germinal-center-like process to unfold. Their data suggest that ectopic lymphoid structures serve as a unique home for Epstein-Barr virus latency and subsequent reactivation driving proliferation of plasma cells. This makes sense. Such structures probably facilitate Epstein-Barr virus growth and persistence through plasma cell generation.

Previous studies examining a possible Epstein-Barr virus / Sjögren's syndrome link did not appreciate the presence of these ectopic germinal center-like formations nor did they focus on B cells, instead looking mostly at epithelial cells. This specific focus may explain the conflicting conclusions of the past literature.

Importantly, in the current study, authors show that Epstein-Barr virus tends to infect disease-specific autoantibody-producing plasma B cells surrounding the B cell follicles of the salivary gland ectopic lymphoid structures. That the disease-specific, autoreactive plasma cells located in ectopic lymphoid structures frequently featured reactivated Epstein-Barr virus is made more striking by the significant presence of anti-Epstein-Barr virus antibody producing plasma cells in the same salivary glands, as the authors showed in mice with transplanted human salivary glands. The well-documented (17, 18) cross-reactive nature of these anti-Epstein-Barr virus antibodies (anti-EBNA1 and anti-VCA) and anti-Ro/La antibodies support a mechanism of molecular mimicry. While the current study does not address a direct causal nature between Epstein-Barr virus and Sjögren's Syndrome, these results are consistent with the hypothesis that, not only is Epstein-Barr virus driving specific dysregulation and survival of autoreactive B cell clones, as the data strongly suggest, but also perhaps driving the very failure of tolerance to self in the first place.

Would this be an expected result given the random nature of most relationships? Given that Epstein-Barr virus resides in the rare B cell in the ~95% of the human adult population infected with this herpes virus for life, finding any subset of B cells selectively enriched for Epstein-Barr virus infestation is unexpected. That a B cell trophic virus, here Epstein Barr virus, would have the moxie to be found in the cells generating the autoantibodies relatively specific for the disease is a connection to possible mechanism and pathogenesis that could be so important that, if independently confirmed, warrants mechanistic elucidation.

Before we become enamored by the possibility that Epstein-Barr virus is a prominent player in the etiology of Sjögren's Syndrome, we should remember that the sequence of events is not addressed here. Though we judge it unlikely, we must formally accede that no order of related phenomena is established in this study. These patients are evaluated at one point in time and the relationships between the phenomena observed are not addressed. Maybe, the specific autoantibody production precedes Epstein-Barr virus infection. Perhaps, the coincident presence of Epstein-Barr virus has nothing to do with the autoantibody specificities observed. The data supporting the direct pathogenicity of autoantibodies subsequently leading to Sjögren's Syndrome are not sufficient to escape the challenge by many serious investigators. Despite these misgivings, the intriguing possibility that this is an insight into causation gives hope that we might one day be able to make clinical decisions based on the understanding between the relationship of Epstein-Barr virus and Sjögren's syndrome.

If Epstein-Barr virus really plays a pathogenic role in the generation of Sjögren's syndrome, then the host and viral cellular and molecular mechanisms that bring the process to this incredible point of convergence would be expected to provide innumerable therapeutic targets, some likely to be unique to Sjögren's syndrome pathogenesis. These are the kinds of therapies that would have the potential to be disease-specific with much less systemic risk than the general inhibition of effector molecules or of specific signaling pathways that is the present rage.

No matter what they will ultimately be found to mean, the observations herein presented by Bombardieri and colleagues (16) are compelling for anyone interested in understanding Sjögren's syndrome. They deserve to be fully vetted. If confirmed, they need to be explained at every level technologically available to us in the second decade of the 21st century.

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