Journal of Translational Autoimmunity 1 (2019) 100003

Contents lists available at ScienceDirect

Journal of Translational Autoimmunity

journal homepage: www.journals.elsevier.com/journal-of-translational-autoimmunity/

Immunodietica: A data-driven approach to investigate interactions between diet and autoimmune disorders

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ARTICLE INFO

Keywords:
Autoimmunity
Diet
Epitopes
Cross-reactivity
Gershteyn-Ferreira index

ABSTRACT

Autoimmunity is on the rise around the globe. Diet has been proposed as a risk factor for autoimmunity and shown to modulate the severity of several autoimmune disorders. Yet, the interaction between diet and autoimmunity in humans remains largely unstudied. Here, we systematically interrogated commonly consumed animals and plants for peptide epitopes previously implicated in human autoimmune disease. A total of fourteen species investigated could be divided into three broad categories regarding their content in human autoimmune epitopes, which we represented using a new metric, the Gershteyn-Ferreira index (GF index). Strikingly, pig contains a disproportionately high number of unique autoimmune epitopes compared to all other species analyzed. This work uncovers a potential new link between pork consumption and autoimmunity in humans and lays the foundation for future studies on the impact of diet on the pathogenesis and progression of autoimmune disorders.

1. Introduction

Autoimmune disorders, combined, affect an estimated 50 million Americans. The National Institutes of Health (NIH) spend over $500 M annually in research grants to study autoimmunity. Yet, the risk factors, causes, and modifiers of most autoimmune disorders remain poorly understood. Strikingly, genetics has been estimated to account for only 30% of all autoimmune diseases, with environmental triggers being responsible for the vast majority of autoimmune disease incidence [1,31].

1.1. Molecular mimicry

The most well known example of an autoimmunity environmental trigger is infection. Some autoimmune disorders, notably Guillain-Barré syndrome (GBS) and type 1 diabetes (T1D), have been suggested to be triggered by bacterial and viral infections [13,16,23,24]. The mechanism behind these observations is thought to be molecular mimicry. Some bacteria and viruses contain epitopes identical to self epitopes in the host. While most self-reactive T cells are deleted in thymus (central tolerance), some escape to the periphery and can be detected in peripheral blood [32]. Nevertheless, these autoreactive T cells remain anergic in the absence of danger signals and thus pose no threat of autoimmunity at steady state. Infection with bacteria or viruses containing these cross-reactive peptides, however, will lead the host to mount an immune response against them potentially using T cells that recognize pathogenic antigens that match self antigens. When infection subsides, the remaining memory T cells that recognize those shared bacterial and viral antigens are now poised to attack the tissues that express their cognate antigens, leading to autoimmunity [6,22].

A decade ago, a dramatic demonstration that exposure to higher organisms' antigens can also cause autoimmunity occurred when pork abattoir workers processing pig brains by removing and liquefying them using compressed air suddenly developed an autoimmune neurologic disorder [7]. Analysis of 24 exposed workers indicated that aerosolized pig neural antigens induced polyradiculoneuropathy, a normally slow-developing autoimmune neurologic disease, in as little as 4 weeks of exposure. Nerve biopsies revealed demyelination, axonal degeneration, and perivascular inflammation. Moreover, sera from all exposed workers,

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https://doi.org/10.1016/j.jtauto.2019.100003
Received 2 May 2019; Received in revised form 17 May 2019; Accepted 19 May 2019

but not sera from any of almost 200 healthy community controls, contained neural-reactive IgG antibodies. Specifically, an IgG specific to myelin basic protein (MBP) was detected in 75% of patients' sera. Strikingly, anti-MBP antibody titers were inversely associated with the distance of the worker to the point of pig brain extraction and aerosolization. Almost 50% of the exposed workers required immunosuppressive therapy (corticosteroids) to alleviate their symptoms [11,27]. Recent follow-up studies report that the vast majority of patients still experience pain, and tingling, display nerve conduction anomalies, and retain circulating antibodies reactive against sections of distal nerve terminals, peroneal nerve, and ventral roots [19]. Altogether, these data strongly suggest a causal relationship between exposure to aerosolized pig antigens and neurological autoimmunity. The identity of the pig antigens that caused the onset of polyradiculoneuropathy, however, remains unknown.

1.2. Oral tolerance and gut permeability

Can dietary antigens then initiate or exacerbate autoimmune disease? Oral antigen delivery often results in the creation of tolerance to that antigen, a phenomenon known as oral tolerance. Initially described in the mouse, oral tolerance is induced upon the interaction of orally administered antigens with the immune system in the gut, chiefly in the mesenteric lymph nodes, leading to the generation of antigen-specific regulatory T cells [3,5,21]. Yet, oral tolerance can be compromised. Recently, it was found that mechanical injury to the skin promotes anaphylaxis, an acute allergic reaction, to oral antigen. Mechanistically, injury-activated keratinocytes in the skin released IL-33 systemically, ultimately activating gut mast cells and increasing gut permeability [12]. Intermittent gut permeability allows partially digested foreign proteins and commensal bacteria to enter the bloodstream and be presented to the immune system in a pro-inflammatory context. Recognition of cross-reactive diet-derived peptides by autoreactive T cells in this fashion may initiate or help sustain autoimmunity [2,18].

1.3. Diet and autoimmune disease

Indeed, several instances of an impact of diet on autoimmunity have been reported. Rheumatoid arthritis (RA) patients have long been known to have elevated levels of IgG and IgA antibodies in serum, with those with vasculitis displaying even higher increases in these classes of antibodies, as well as in IgM antibodies [28,29]. Deposition of soluble antigen-antibody immune complexes and concomitant fixation of complement, a type III hypersensitivity reaction, in joints and small blood vessels cause inflammatory tissue damage in RA [25]. Patients often report an association between food intake and RA symptom severity. Indeed, a randomized single-blinded clinical trial with 27 experimental patients and 26 control patients found that fasting, followed by one year of vegetarian diet (experiment), significantly reduced pain, morning stiffness, swollen joints, and several other RA symptoms when compared to a regular diet (control) [9]. In line with these observations, all three immunoglobulin classes (IgG, IgA, and IgM) were found to have heightened food-specific activities both systemically (serum) and in the intestine (jejunal perfusion fluid) in most of the RA patients analyzed as compared to healthy controls. Recognized antigens originated chiefly from cow's milk, cereals, hen's eggs, cod, and pork [8]. Remarkably, a strong correlation ($r^2 = 0.795$) was found between cow milk consumption and multiple sclerosis (MS) incidence across 27 countries. Of note, no such correlation was found with cheese consumption, suggesting the involvement of molecules not present in processed dairy [14]. Of note, introducing certain foods early in life has been associated with T1D development. Yet, studies in this field have yielded contradictory results, warranting the use of bigger cohorts and more mechanistic studies in the future [10,20,30].

Porcine neural antigens contain epitopes recognized in human neural autoimmunity [11,27]. Are there porcine peptide epitopes recognized in autoimmune diseases in other tissues? More broadly, what is the degree of similarity between common food epitopes and those implicated in human autoimmune disorders? Are specific animals or plants enriched for cross-reactive epitopes in certain diseases? We sought to systematically address these questions by mapping the epitope similarity between animals and plants commonly consumed and different human autoimmune diseases.

2. Materials and methods

To investigate the overlap of epitopes between human autoimmune disease and commonly consumed foods, we aggregated all epitopes implicated in 77 human autoimmune diseases available at www.iedb.org and cross-referenced them with every epitope in 14 organisms. Those organisms were Atlantic salmon (taxid:8030), Chicken (taxid:9031), Cow (taxid:9913), Duck (taxid:8839), Goat (taxid:9925), Japanese rice (taxid:39947), Turkey (taxid:9102), Nile tilapia (taxid:8128), Pig (taxid:9823), Quinoa (taxid:63459), Rye (taxid:4550), Sheep (taxid:9940), Soybean (taxid:3847), and Wheat (taxid:4564) (Table S1). We utilized Node.js, AWS EC2, and PostgreSQL database technologies in order to automate data gathering and allow for expedient analysis. Data gathering automation was broken into two custom built processes running concurrently.

2.1. Process #1

The first process (Process #1) queried the National Center for Biotechnology Information (NCBI) blastp system. The query request consisted of the unique epitope ID and the list of organisms that were considered for the study. All the queries went against the NCBI refseq_protein database. Initially, the process made a query for a single epitope and a single organism. However, this approach would not scale for our use case, as the final data set would consist of ~18000 epitopes in combination with 14 organisms (in excess of 250,000 queries). Additionally, depending on the query and its position in the job queue on the NCBI servers, each result would vary in time spanning from under a minute to multiple hours. Fortunately, the API supports a list of organisms as a valid query criterion, significantly cutting down the round trip of each query. Once a request was made for a particular epitope in the

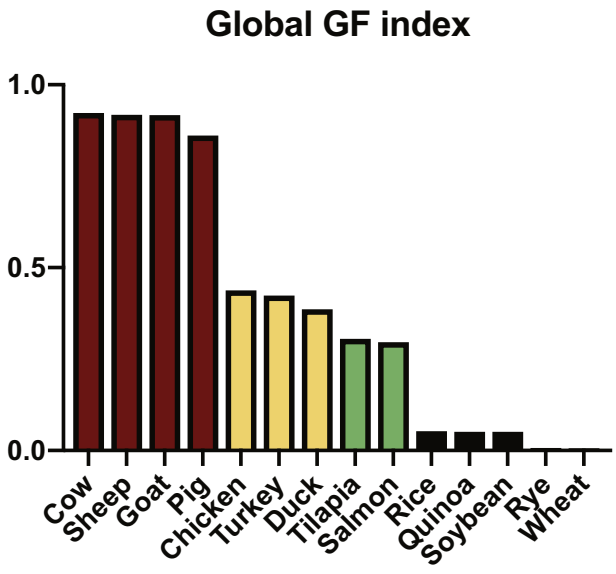

Fig. 1. Global Gershteyn-Ferreira (GF) index for different species. Species are colored according to food group: red meat, poultry, fish, and cereals. The global GF index ranges between 0 and 1 for the analyzed species. Rye and wheat have very low non-zero GF indices.

database, Process #1 created a record with the Request ID where the uniqueness of the record was the epitope ID and the organism. The record was marked as pending to indicate that it is ready to be retrieved for processing by Process #2. The process continued until all the epitopes had been queried and marked as pending.

Once both processes were fully executed, we had compiled a fully comprehensive database of autoimmune human-organism epitope overlap. To assess the epitope similarity between the species analyzed and human autoimmune epitopes, we created the Gershteyn-Ferreira (GF) index (Table S1), obtained using the formula below:

$$\text{Global Gershteyn-Ferreira (GF) index} = \left(\frac{\textit{Organism-specific Overlapping Epitopes}}{\textit{All Human Autoimmune Epitopes}}\right) * 2$$

2.2. Process #2

The second process (Process #2) retrieved the results of the queries made by using the Request ID from Process #1. As the query could take a certain amount of time to finish, the process would iterate through the list of all the records that had a valid Request ID to obtain the results. In the query result data, the top hit for each organism was selected and then, if the Query Coverage and Identity Percentage values were both 100, the record result was considered as a hit (total match), otherwise a miss (Table S1). The record was marked as complete so the process would ignore it on the next pass. The system continued polling NCBI servers until all results were gathered.

Next, we sought to represent the degree of epitope similarity between different species and epitopes implicated in specific human autoimmune disorders. We aligned all species' epitopes with human autoimmune epitopes and created disease-specific GF indices to express that similarity. Lastly, to isolate potential species-restricted mechanisms of contribution to autoimmunity pathogenesis and severity, we created the unique autoimmune GF index, which only takes into account autoimmune epitopes uniquely present in each species and not anywhere else in our sample. Of note, pig had the highest number of unique autoimmune epitopes (Table S2) and thus we used the inverse of the number of unique pig autoimmune epitopes as a normalizing factor, with the goal of getting a theoretical maximum of 1. Formulas for these two specific GF indices are as follows:

$$\text{Disease-specific GF index} = \left(\frac{\textit{Organism-specific Overlapping Epitopes}}{\textit{Total Disease-associated Epitopes}}\right) * \textit{Normalizing Factor}$$

$$\text{Unique autoimmune GF index} = \left(\frac{\textit{Unique Organism-specific Overlapping epitopes}}{\textit{Total Disease-associated epitopes}}\right) * \textit{Normalizing Factor}$$

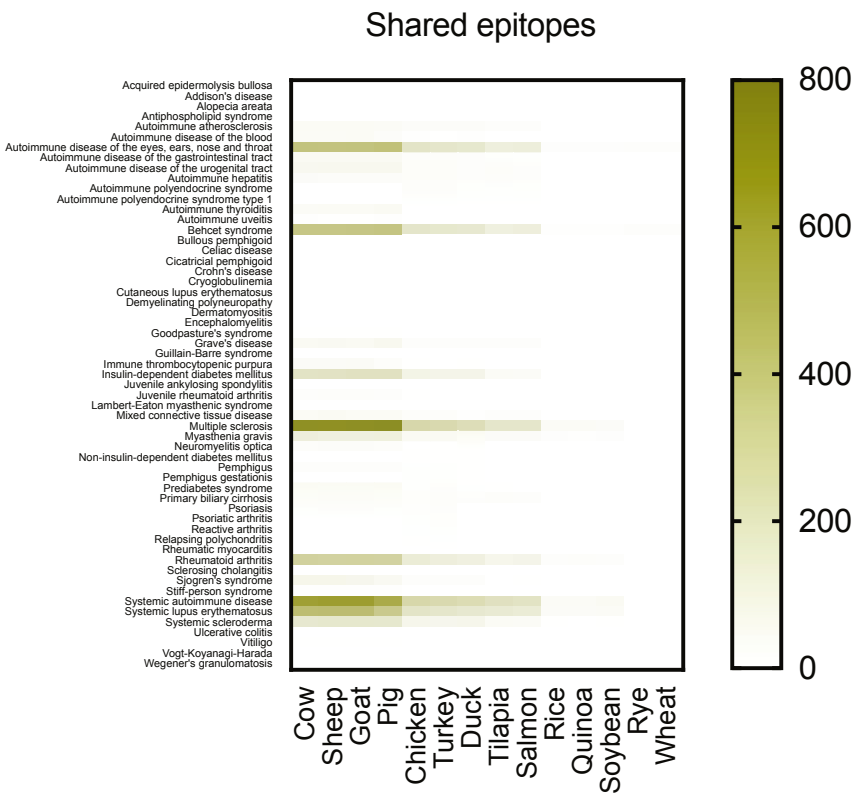

Fig. 2. Shared epitopes per autoimmune disease. Autoimmune diseases are listed on the y axis and analyzed species on the x axis.

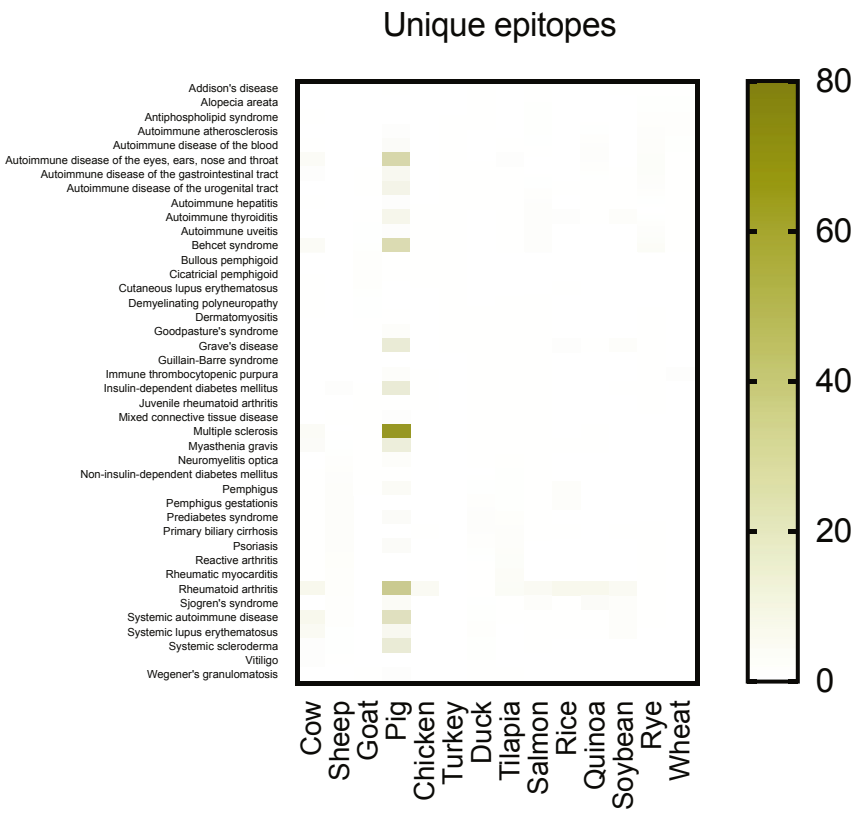

Fig. 3. Unique epitopes per autoimmune disease. Autoimmune diseases are listed on the y axis and analyzed species on the x axis.

Table 1
Diseases with single species epitope matches.

Disease	Species	Number of hits
Alopecia aerata	Salmon	1
Antiphospholipid syndrome	Cow	1
Autoimmune atherosclerosis	Pig	2
Bullous pemphigoid	Pig	1
Cutaneous lupus erythematosus	Rice	1
Demyelinating polyneuropathy	Cow	1
Dermatomyositis	Pig	1
Goodpasture's syndrome	Pig	3
Guillain-Barre syndrome	Cow	1
Non-insulin dependent diabetes mellitus	Rye	1
Reactive arthritis	Salmon	1
Rheumatic myocarditis	Cow	1
Vitiligo	Cow	2

An interactive website for researchers and the public to explore this database will be available at www.immunodietica.com.

3. Results

After screening all available linear epitopes of a total of 14 commonly consumed animals and plants against human linear epitopes, we selected hits with 100% coverage and identity (identical amino acid sequences) and rank ordered the hits for immunogenic similarity (see *Materials and Methods*). We then normalized this index, which we named the Gershteyn-Ferreira index (GF index), to range from 0 to 1 for the 14 organisms in our sample, although mathematically the maximum of the global GF index is 2 in case of Human-Human identity (100% overlap). As can be seen in Fig. 1, the analyzed species fell within 3 major groups with regards to their global GF index. Red meat (cow, sheep, goat, pig) had the highest GF index, poultry and fish (chicken, turkey, duck, tilapia, and salmon) an intermediate GF index, and cereals (rice, quinoa, soybean, rye, and wheat) a low GF index.

In order to dissect the relative similarity of each dietary epitope to specific autoimmune disorders, we isolated autoimmune disease-associated epitopes and cross-referenced each organism's hits with the overlap to each of the 77 immune diseases featured in the www.iedb.org database (Fig. 2).

Similarly to the global GF index, cow, sheep, goat, pig (red meat) had the highest number of epitopes shared with specific autoimmune diseases (Fig. 2). The highest number of shared epitopes was 725; pig has 725 epitopes implicated in MS.

Finally, we isolated the unique hits per animal or plant and found that the amount of unique disease-specific epitopes appearing in pigs is 10–15 times higher than in any other organism (Fig. 3). Of the 68 diseases analyzed (9 diseases were excluded due to the lack of dietary antigen hits), 40 have at least one unique autoimmune epitope found in one of the animals and plants analyzed (Fig. 3).

Strikingly, pig was found to share at least one epitope with 29 of those diseases (72.5%). Thirteen diseases only had epitope matches with one species (Table 1). Pig shared unique epitopes with 4 (30.8%) of these disorders: autoimmune atherosclerosis, bullous pemphigoid, dermatomyositis, and Goodpasture's syndrome. Yet, the species found to possess shared unique epitopes with the highest number of diseases (5) in exclusivity was the cow. These were antiphospholipid syndrome (APS), demyelinating polyneuropathy, GBS, rheumatic myocarditis, and vitiligo (Table 1).

Next, we sought to gain a deeper understanding of the tissue expression patterns of autoimmune epitopes found in food. We focused on the 6 autoimmune neurological disorders for which we found autoimmune epitopes matching epitopes in at least one of the species analyzed: demyelinating polyneuropathy, encephalomyelitis, GBS, MS, neuromylitis optica, and Stiff-person syndrome. The complete list of epitopes implicated in each one of these diseases can be found in Tables S3–S8. Pig possesses 5 epitopes previously implicated

Table 2
Sequence and tissue expression of unique multiple sclerosis epitopes found in pig.

Epitope	Parent protein	Location
YLVAVAHGDLLELDPPANHTPCVVQVHTLTG	ATP-sensitive Inward Rectifier Potassium Channel 10	Kidney
YIYFNTWTTCQSIAFPSKTSA	Myelin Proteolipid Protein	CNS
WYRSKFADLTDAAAR	Glial Fibrillary Acidic Protein	CNS
WTTCQSIAFPSKTSASIGSL	Myelin Proteolipid Protein	CNS
WSQLLANSAARKKKL	Spectrin Beta Chain, Erythrocytic	CNS, lung, kidney
VTDDSSFLGGGG	E3 Ubiquitin Protein Ligase RNF115	Heart, skeletal muscle, testis
VSLLHFVPTREANGH	Myelin-associated Glycoprotein	PNS, CNS
VSHFFRELAEEKREG	Ferritin Light Chain	Ubiquitous
VRAPAKPRSPPRSER	Myelin-associated Oligodendrocyte Basic Protein	CNS
VLIRHGESAWNLENR	Phosphoglycerate Mutase 1	CNS, liver
VLFSSDFRI	Myelin-associated Glycoprotein	PNS, CNS
VHGNSGAGNNWAKGH	Tubulin Beta-1 Chain	Blood
VCNPIISGLYQGAGG	Heat Shock 70 kDa Protein 1B	Testis
VAVAHGDLLELDPPANHTPCVVQVHTLTG	ATP-sensitive Inward Rectifier Potassium Channel 10	Kidney
TWTTCQSIAFPSKTSASIGS	Myelin Proteolipid Protein	CNS
TFDPHFLRVPCWKITLFVIV	Myelin Oligodendrocyte Glycoprotein Precursor	CNS
SSVEAIEGSHVSLLC	Myelin-associated Glycoprotein	PNS, CNS
SSNAGGGGGGA	SOX4	CNS, heart, stomach
SKTSASIGSLCADARMYGVLP	Myelin Proteolipid Protein	CNS
SDVGEYRAVTELGRP	SLA-DRB1 Precursor	Ubiquitous
SAFEGTCVSIPCRFD	Myelin-associated Glycoprotein	PNS, CNS
RLRGKLRAE	Myelin Oligodendrocyte Glycoprotein Precursor	CNS
RFDFPDELRPAVVHG	Myelin-associated Glycoprotein	PNS, CNS
RCGLQELGPGLFRGL	Reticulon-4 Receptor	CNS
RAEIENLHRTFDPHF	Myelin Oligodendrocyte Glycoprotein Precursor	CNS
QYNSSSGGGG	RNA-binding Protein FUS	Ubiquitous
QNPNLHKSLASS	Zinc Finger Protein Basonuclin-1	Skin
QAGKELEEQHGIHCN	Transaldolase	Ubiquitous
PVGIRHRLRG	Mitochondrial Import Inner Membrane Translocase Subunit TIM17-B	Ubiquitous
PRSPPRSERQPRPRP	Myelin-associated Oligodendrocyte Basic Protein	CNS
PIIEDRHGGYKPSDE	Creatine Kinase B-type	Ubiquitous
PDPGAGAAGGGG	Ring Finger Protein 50	Ubiquitous
NTWTTCQSIAFPSK	Myelin Proteolipid Protein	CNS
NADACFCIDNEALYD	Tubulin Beta-1 Chain	Blood
MVAFKGVWTQAFWKA	Aquaporin-4	Skeletal muscle, stomach, CNS, lung
MESALDQLKQFTTVV	Transaldolase	Ubiquitous
MAAYKLVLIRHGESA	Phosphoglycerate Mutase 1	CNS, liver
LRVPCWKITLFVIVPV	Myelin Oligodendrocyte Glycoprotein Precursor	CNS
LLMAWGQYIDHDIAF	Thyroid Peroxidase	Thyroid
LKKEKINIRVLDPFT	Transketolase	Ubiquitous
LILPEAVGGTVF	Fatty Acid 2-Hydroxylase	Skin, stomach, small intestine, pancreas, CNS
LFALTLPIW	C-X-C Chemokine Receptor Type 2	Blood
LDRKLILDSARATKG	Transketolase	Ubiquitous
KRPSQRHGSK	Myelin Basic Protein	CNS
KLRAEIENL	Myelin Oligodendrocyte Glycoprotein Precursor	CNS
IAFPSKTSASIGSLC	Myelin Proteolipid Protein	CNS
HRKLFEELVRASSHS	Pyruvate Kinase	Ubiquitous
HIILVLSGK	Cytosolic Fe–S Cluster Assembly Factor NUBP2	Ubiquitous
GVVWYLVAVAHGDLLELDPPANHTPCVVQVHTLTGAFL	ATP-sensitive Inward Rectifier Potassium Channel 10	Kidney
GRDHGLPGYNEWREF	Thyroid Peroxidase	Thyroid
GKVNVDEVGGEALGRLL	Hemoglobin Subunit Beta	Blood
GIRKFAADAVKLERM	Transaldolase	Ubiquitous
GHWGAWMPSSISAFEGTCVSI	Myelin-associated Glycoprotein	PNS, CNS
GGHWGAWMPSSISAF	Myelin-associated Glycoprotein	PNS, CNS
GGAAAQSLYIANHAY	Fructose-biphosphate Aldolase C	CNS
FGEEGLTLNLEDVQP	60 kDa Heat Shock Protein, mitochondrial	Ubiquitous
FDPHFLRVPCWKITL	Myelin Oligodendrocyte Glycoprotein Precursor	CNS
ESAWNLENRFSGWYD	Phosphoglycerate Mutase 1	CNS, liver
ENLHRTFDPHFLRVPCW	Myelin Oligodendrocyte Glycoprotein Precursor	CNS
EELGSKAKFAGRNFR	Alpha-enolase	Ubiquitous
CVSIPCRFDFPDELR	Myelin-associated Glycoprotein	PNS, CNS
CVCYNEPKVTTSCPQ	Reticulon-4 Receptor	CNS
CRLEKPAKYDDIKKV	Gyceraldehyde-3-phosphate Dehydrogenase	Ubiquitous
CQSIAFPSKTSASIGSLCAD	Myelin Proteolipid Protein	CNS
ASQKRPSQRHGSKY	Myelin Basic Protein	CNS
APEYRGRTELLK	Myelin Oligodendrocyte Glycoprotein Precursor	CNS
AFKGVWTQAFWKAVT	Aquaporin-4	Skeletal muscle, stomach, CNS, lung

in GBS (Table S9). Not surprisingly, these epitopes were derived from proteins expressed in nervous tissue, namely MBP and tubulin beta-2B chain, found exclusively in the central nervous system (CNS), myelin protein zero, only found in the peripheral nervous system (PNS), and myelin P2 protein, present in both the CNS and the PNS (Table S9). Of the 6 diseases aforementioned, 2 have epitopes uniquely found in pig amongst all the species analyzed: neuromyelitis optica (3 epitopes) and MS (67 epitopes) (Table S9). Interestingly, pig epitopes matching MS epitopes were derived not only from proteins expressed in the CNS and PNS, but also from a variety of other

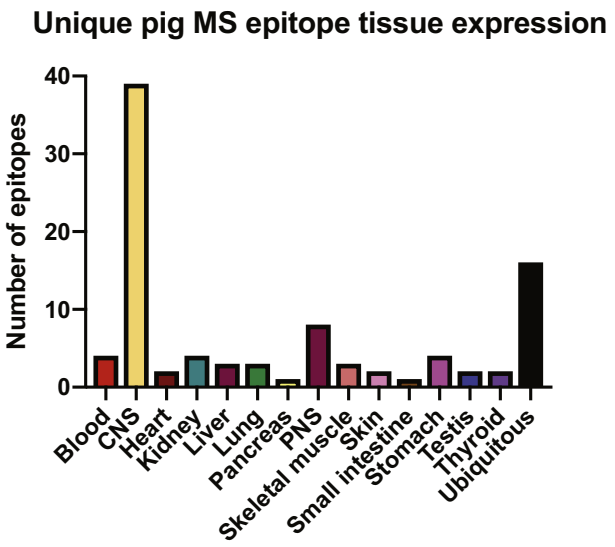

Fig. 4. Tissue expression pattern of unique multiple sclerosis epitopes found in pig. Tissues are listed on the x axis and number of unique MS epitopes in pig on the y axis. CNS, central nervous system; PNS, peripheral nervous system.

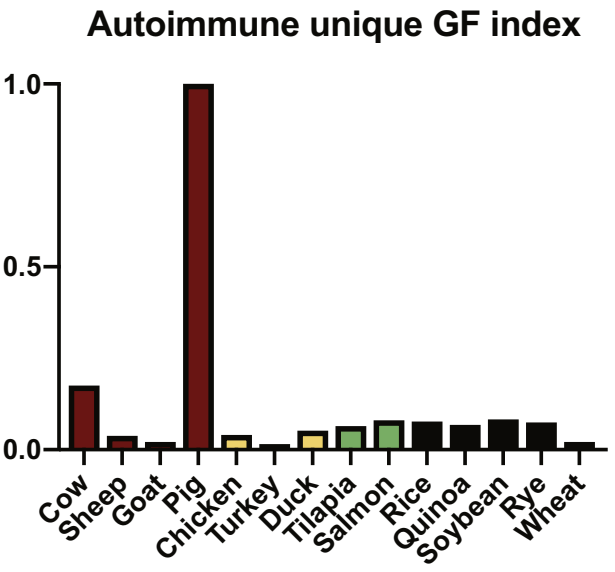

Fig. 5. Autoimmune unique Gershteyn-Ferreira (GF) index. Species are colored according to food group: red meat, poultry, fish, and cereals. The autoimmune unique GF index ranges between 0 and 1.

tissues, including skeletal muscle, heart, and skin (Table 2). In fact, some pig MS epitopes were derived from ubiquitously expressed proteins involved in basic metabolism and other housekeeping processes, with "ubiquitous" constituting the second most common tissue expression category after CNS (Fig. 4).

To better illustrate the unique links between peptide epitopes present in various commonly consumed plants and animals and autoimmune disorders, we created the Autoimmune unique GF index, where we normalized the unique epitope matches to the maximum number observed, i.e. 325 unique epitope matches found in pig (Fig. 5).

4. Discussion

Diet has undergone tremendous changes over the course of history due to globalization and increased life standards. In particular, the consumption of meat and animal products in general has increased over the past decades in both the developed and the developing world [26]. The same time period has also witnessed a noted rise in autoimmune disorders, overwhelmingly due to environmental factors [1]. In addition to many other environmental factors tested, consumption of specific foods has been associated with the genesis and severity of autoimmune disease [8,14]. Through systematic analysis, we found many peptide epitopes present in species commonly consumed as food that have been implicated in human autoimmune disorders. Of note, pig contains the highest number of autoimmune-associated epitopes not found in any other species investigated. Importantly, many pig-derived autoimmune neurological disease-associated epitopes, specifically MS epitopes, were found to be expressed in all tissues in the body and are thus likely to be present in pork.

A caveat in our current approach is the inability to account for immunogenic peptides arising from protein posttranslational modifications inside cells. Three examples stand out: gliadin peptides modified by tissue transglutaminase involved in the pathogenesis of celiac disease [17], "hybrid insulin peptides" (HIPs), fusion peptides resulting from covalent cross-linking between pro-insulin peptides and peptides derived from other proteins present in β cells' secretory granules recently implicated in T1D [4], and citrullinated proteins, thought to be important targets for autoantibody recognition in RA [15].

We predict that subpopulations with outsized contributions of certain foods in their diet where there is a shared epitope with an autoimmune disease will be over-represented in the incidence of that disease in the total population, assuming their genetic predisposition is not significantly different from the average in that population. Moreover, avoidance of foods containing autoimmune peptide epitopes could improve ongoing symptoms of the autoimmune disease in question. Epidemiological studies seeking associations between the consistent consumption of certain foods and the incidence and severity of different autoimmune disorders are thus warranted. In addition, mechanistic experiments *in vitro* and in animal models will be required to dissect the contribution of dietary antigens to the pathogenesis and magnitude of autoimmunity. Factors to be tested include quantity, duration, and periodicity of exposure to diet-derived epitopes matching autoimmune epitopes, in isolation and in combination with perturbations in environmental factors, such as stress and microbiome composition. An additional area of interest is the detection and quantification of predicted autoimmune epitopes in different food samples originating from distinct parts of an animal or prepared differently. Finally, for humans in particular, human leukocyte antigen (HLA) genotype, which plays an important role in predisposition to autoimmunity, will also undoubtedly act as a modifier of the impact of different dietary antigens on disease.

Interestingly, many cultures forbid certain foods from consumption. Pork specifically has been considered unclean by a vast variety of cultures and religions. Since these rules have stood the test of time and are likely to remain for many years going forward, it is intriguing that we found pig antigens to contain, by far, the highest number of unique overlapping epitopes with human autoimmune disorder-associated epitopes. Our results support the hypothesis that there are links between specific foods and autoimmune diseases, and that a common denominator is disproportionately porcine in nature.

Conflict of interest

The authors received funding from Ajax Biomedical Foundation, a 501(c)(3) corporation.

Acknowledgements

The authors thank Andrey Burov for his excellent technical and programming contributions, Dr. Andrey Vyshedskiy for his stimulating discussions, and the reviewers for insightful comments. This work was supported by the Ajax Biomedical Foundation (Newton, MA). L.M.R.F. is

a Jeffrey G. Klein Family Diabetes Fellow. Dedicated to the memory of Lev Gershteyn.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jtauto.2019.100003.

References

[1] J.F. Bach, The effect of infections on susceptibility to autoimmune and allergic diseases, N. Engl. J. Med. 347 (2002) 911–920.

[2] S.C. Bischoff, G. Barbara, W. Buurman, T. Ockhuizen, J.D. Schulzke, M. Serino, H. Tilg, A. Watson, J.M. Wells, Intestinal permeability–a new target for disease prevention and therapy, BMC Gastroenterol. 14 (2014) 189.

[3] Y. Chen, V.K. Kuchroo, J. Inobe, D.A. Hafler, H.L. Weiner, Regulatory T cell clones induced by oral tolerance: suppression of autoimmune encephalomyelitis, Science 265 (1994) 1237–1240.

[4] T. Delong, T.A. Wiles, R.L. Baker, B. Bradley, G. Barbour, R. Reisdorph, M. Armstrong, R.L. Powell, N. Reisdorph, N. Kumar, C.M. Elso, M. DeNicola, R. Bottino, A.C. Powers, D.M. Harlan, S.C. Kent, S.I. Mannering, K. Haskins, Pathogenic CD4 T cells in type 1 diabetes recognize epitopes formed by peptide fusion, Science 351 (2016) 711–714.

[5] D. Esterhazy, J. Loschko, M. London, V. Jove, T.Y. Oliveira, D. Mucida, Classical dendritic cells are required for dietary antigen-mediated induction of peripheral T(reg) cells and tolerance, Nat. Immunol. 17 (2016) 545–555.

[6] L. Guilherme, E. Cunha-Neto, V. Coelho, R. Snitcowsky, P.M. Pomerantzeff, R.V. Assis, F. Pedra, J. Neumann, A. Goldberg, M.E. Patarroyo, et al., Human heart-infiltrating T-cell clones from rheumatic heart disease patients recognize both streptococcal and cardiac proteins, Circulation 92 (1995) 415–420.

[7] S.M. Holzbauer, A.S. DeVries, J.J. Sejvar, C.H. Lees, J. Adjemian, J.H. McQuiston, C. Medus, C.A. Lexau, J.R. Harris, S.E. Recuenco, E.D. Belay, J.F. Howell, B.F. Buss, M. Hornig, J.D. Gibbins, S.E. Brueck, K.E. Smith, R.N. Danila, W.I. Lipkin, D.H. Lachance, P.J. Dyck, R. Lynfield, Epidemiologic investigation of immune-mediated polyradiculoneuropathy among abattoir workers exposed to porcine brain, PLoS One 5 (2010) e9782.

[8] M. Hvatum, L. Kanerud, R. Hallgren, P. Brandtzaeg, The gut-joint axis: cross reactive food antibodies in rheumatoid arthritis, Gut 55 (2006) 1240–1247.

[9] J. Kjeldsen-Kragh, M. Haugen, C.F. Borchgrevink, E. Laerum, M. Eek, P. Mowinkel, K. Hovi, O. Forre, Controlled trial of fasting and one-year vegetarian diet in rheumatoid arthritis, Lancet 338 (1991) 899–902.

[10] J.N. Kostraba, K.J. Cruickshanks, J. Lawler-Heavner, L.F. Jobim, M.J. Rewers, E.C. Gay, H.P. Chase, G. Klingensmith, R.F. Hamman, Early exposure to cow's milk and solid foods in infancy, genetic predisposition, and risk of IDDM, Diabetes 42 (1993) 288–295.

[11] D.H. Lachance, V.A. Lennon, S.J. Pittock, J.A. Tracy, K.N. Krecke, K.K. Amrami, E.M. Poeschla, R. Orenstein, B.W. Scheithauer, J.J. Sejvar, S. Holzbauer, A.S. Devries, P.J. Dyck, An outbreak of neurological autoimmunity with polyradiculoneuropathy in workers exposed to aerosolised porcine neural tissue: a descriptive study, Lancet Neurol. 9 (2010) 55–66.

[12] J. Leyva-Castillo, C. Galand, C. Kam, O. Burton, S.F. Ziegler, I. Chiu, F. Austen, R.S. Geha, Mechanical skin injury promotes food anaphylaxis by driving intestinal mast cell expansion, Immunity 50 (2019) 1–14.

[13] M. Lonnrot, K. Korpela, M. Knip, J. Ilonen, O. Simell, S. Korhonen, K. Savola, P. Muona, T. Simell, P. Koskela, H. Hyoty, Enterovirus infection as a risk factor for beta-cell autoimmunity in a prospectively observed birth cohort: the Finnish Diabetes Prediction and Prevention Study, Diabetes 49 (2000) 1314–1318.

[14] D. Malosse, H. Perron, Correlation analysis between bovine populations, other farm animals, house pets, and multiple sclerosis prevalence, Neuroepidemiology 12 (1993) 15–27.

[15] L. Mathsson, M. Mullazehi, M.C. Wick, O. Sjoberg, R. van Vollenhoven, L. Klareskog, J. Ronnelid, Antibodies against citrullinated vimentin in rheumatoid arthritis: higher sensitivity and extended prognostic value concerning future radiographic progression as compared with antibodies against cyclic citrullinated peptides, Arthritis Rheum. 58 (2008) 36–45.

[16] M.A. Menser, J.M. Forrest, R.D. Bransby, Rubella infection and diabetes mellitus, Lancet 1 (1978) 57–60.

[17] O. Molberg, S.N. McAdam, R. Korner, H. Quarsten, C. Kristiansen, L. Madsen, L. Fugger, H. Scott, O. Noren, P. Roepstorff, K.E. Lundin, H. Sjostrom, L.M. Sollid, Tissue transglutaminase selectively modifies gliadin peptides that are recognized by gut-derived T cells in celiac disease, Nat. Med. 4 (1998) 713–717.

[18] Q. Mu, J. Kirby, C.M. Reilly, X.M. Luo, Leaky gut as a danger signal for autoimmune diseases, Front. Immunol. 8 (2017) 598.

[19] S.A. Muley, S. Holzbauer, B. Clark, J. Sejvar, R. Lynfield, G. Parry, A.S. DeVries, Immunohistochemistry and electrophysiological findings in swine abattoir workers with immune-mediated polyradiculoneuropathy, J. Neurol. Sci. 385 (2018) 34–38.

[20] J.M. Norris, K. Barriga, G. Klingensmith, M. Hoffman, G.S. Eisenbarth, H.A. Erlich, M. Rewers, Timing of initial cereal exposure in infancy and risk of islet autoimmunity, J. Am. Med. Assoc. 290 (2003) 1713–1720.

[21] O. Pabst, A.M. Mowat, Oral tolerance to food protein, Mucosal Immunol. 5 (2012) 232–239.

[22] M. Rojas, P. Restrepo-Jimenez, D.M. Monsalve, Y. Pacheco, Y. Acosta-Ampudia, C. Ramirez-Santana, P.S.C. Leung, A.A. Ansari, M.E. Gershwin, J.M. Anaya, Molecular mimicry and autoimmunity, J. Autoimmun. 95 (2018) 100–123.

[23] K. Schopfer, L. Matter, U. Flueler, E. Werder, Diabetes mellitus, endocrine autoantibodies, and prenatal rubella infection, Lancet 2 (1982) 159.

[24] N. Shahrizaila, N. Yuki, Guillain-barre syndrome animal model: the first proof of molecular mimicry in human autoimmune disorder, J. Biomed. Biotechnol. 2011 (2011) 829129.

[25] Y.W. Song, E.H. Kang, Autoantibodies in rheumatoid arthritis: rheumatoid factors and anticitrullinated protein antibodies, QJM 103 (2010) 139–146.

[26] A.W. Speedy, Global production and consumption of animal source foods, J. Nutr. 133 (2003) 4048S–4053S.

[27] J.A. Tracy, P.J. Dyck, Auto-immune polyradiculoneuropathy and a novel IgG biomarker in workers exposed to aerosolized porcine brain, J. Peripher. Nerv. Syst. 16 (Suppl 1) (2011) 34–37.

[28] E.M. Veys, H.E. Claessens, Serum levels of IgG, IgM, and IgA in rheumatoid arthritis, Ann. Rheum. Dis. 27 (1968) 431–440.

[29] E.M. Veys, P.A. Gabriel, E. Coigne, H. Mielants, Rheumatoid factor and serum IgG, IgM and IgA levels in rheumatoid arthritis with vasculitis, Scand. J. Rheumatol. 5 (1976) 1–6.

[30] S.M. Virtanen, M. Knip, Nutritional risk predictors of beta cell autoimmunity and type 1 diabetes at a young age, Am. J. Clin. Nutr. 78 (2003) 1053–1067.

[31] A. Vojdani, K.M. Pollard, A.W. Campbell, Environmental triggers and autoimmunity, Autoimmune Dis. 2014 (2014) 798029.

[32] W. Yu, N. Jiang, P.J. Ebert, B.A. Kidd, S. Muller, P.J. Lund, J. Juang, K. Adachi, T. Tse, M.E. Birnbaum, E.W. Newell, D.M. Wilson, G.M. Grotenbreg, S. Valitutti, S.R. Quake, M.M. Davis, Clonal deletion prunes but does not eliminate self-specific alphabeta CD8(+) T lymphocytes, Immunity 42 (2015) 929–941.