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Editorial

Don't forget the exogenous microbial transglutaminases: it is immunogenic and potentially pathogenic

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Abstract: The exogenous microbial transglutaminase that imitates extensively the functions of the endogenous transglutaminases, is a universal protein cross-linker and translational modifier of peptides. The intestinal microbiome, dysbiome, pathobiome, probiotics and industrial processed food are at the origin of the luminal microbial transglutaminase daily cargo. It is hypothesized that those exogenous enzymes, are potential drivers of neurodegenerative and neuroinflammatory diseases via the gut luminal eco events. The substantial luminal activity of the enzyme, by cross-linking naive proteins, can potentially generate neo-epitopes that are not only immunogenic but may also be pathogenic, activating some harmful pathways in the cascade of chronic central brain diseases induction or progression. The harmful activities of microbial transglutaminase may represent a new pathway in the gut-brain axis and might open new therapeutical strategies to fight neurodegenerative conditions.

Keywords: microbial transglutaminase; transglutaminases; mechanism; antibodies; environment; immunogenicity; pathogenicity

1. Introduction

Tissue transglutaminase (tTg) is a pleiotropic enzyme expressed ubiquitously and abundantly. It has been implicated in a variety of physiological processes, such as growth, differentiation, migration, signaling, cytoprotection, cell death and survival, wound healing, angiogenesis,

inflammation, apoptosis and autophagy. It operates intracellularly in multiple organelles, extracellularly and on cell surface. It plays a role in inflammatory, degenerative-age related, neurodegenerative, malignant, metabolic and hormonal, autoimmune and genetic conditions [1]. In celiac disease, it is the autoantigen whereby anti-tTg or the anti-neoepitope tTg antibodies are directed to [2,3]. tTg is a member of the family of transglutaminases (Tgases) (Enzyme Commission [EC] no. 2.3.2.13, OMIN*190196), i.e., protein-glutamine γ-glutamyltransferase, belongs to the class of transferases. It catalyzes the formation of an isopeptide bond between the group of γ-carboxamides of glutamine residues (donor) and the first-order ε-amine groups of different compounds, for instance, proteins (acceptors of an acyl residue). tTg is the most abundant and most studied of the nine members of the Tgases enzyme family [1]. Recently, more data are accumulating on the role of Tgases in the pathomechanisms of human neurodegenerative and neuroinflammatory diseases. Gatta NG et al. should be congratulated for the most recent review on the subject, highlighting the potential molecular mechanisms of Tgases in central degenerative and inflammatory diseases [4]. Since aberrant Tgases cerebral activity was found in Alzheimer's, Parkinson's and Huntington's diseases, supranuclear palsy and other polyglutamine diseases, and since increased content of cross-linked proteins are found in affected brains and since Tgases are a major peptide cross-linkers and a prototype of post-translational modification of proteins (PTMP), it is logical that they are involved in those diseases. The present editorial will describe another Tgase, but this time an exogenous one: microbial Tgase (mTg), that imitate extensively the functions of endogenous tTg that potentially might be involved in brain diseases and may represent a new pathway in the gut-brain axis.

2. Microbial Transglutaminase

Microbial Tgase can catalyze all three reactions of the Tgases family: acyl-transfer reaction, cross-linking reaction between Gln and Lys residues of proteins or peptides (transamidation) and deamidation [1,2,5,6]. mTg is an extracellular enzyme and is biosynthesized by multiple microbes. It has been isolated from Streptoverticillium sp. Contrary to the tTg, mTg is calcium independent, has a lower molecular weight, has a single structural domain and exhibits a different reactivity to food proteins. These characteristics make mTg a very useful tool for modifying the functionality of proteins in food products [6,7].

3. mTg Usage in Food Industries

The development of bread process was an important event for mankind, resulting in bread becoming a commodity within almost everyone's reach. Introduction of industrial enzymes in the baking process has, over the last 14 years, led to the development of a significant segment of the industry, as reflected by increased market value and the growth predictions for the next 6 years. In fact, the value of the baked goods industrial enzyme market doubled between 2000 and 2010. The prediction for 2015–2020 is an additional increase, mounting to 144% [5,6,8].

mTg occupies a substantial segment of the industrial enzyme markets. Thanks to established bioengineering techniques, the microbial expression of enzymatic genes gave rise to massive microbial transglutaminase production. Improvement of meat texture, appearance, hardness and preservability, increased fish product hardness, improved quality and texture of milk and dairy

products, decreased calories, improved texture and elasticity of sweet foods, protein film stability and appearance and improved texture and volume in the bakery industry are only some of mTg applications in the processed food industry. The demand for baked goods, food and beverage enzymes is forecasted to grow by 0.22 to 0.32 fold per year, between 2000 and 2020 [8]. Altogether, a maximum daily intake of mTg could range up to 15 mg. Dosing for restructuring is about 50–100 mg of mTg for each kilogram of treated food [9].

4. Microbial Transglutaminase Cross-links Products and Forms Complexes that are Immunogenic in Human

Several studies have shown that the mTg cross-linked nutritional products elicited antibodies in human [6]. Transglutaminase-modified gluten proteins were shown to react with immunoglobulin A (IgA) anti-gliadin antibodies in the sera of celiac patients. mTg-treated cereal prolamines are preferentially recognized by IgA from celiac patients in an age-dependent manner. Most recently, it was shown that mTg-treated gluten peptides applied to cultured intestinal biopsies from patients with celiac disease induced a 15-fold increase in interferon-ϒ release and 2.5-fold and 2.1-fold increases, respectively, in mean tissue transglutaminase antibody levels and endomysial antibody positivity [10]. Further studies reinforce the immunoreactivity of wheat products treated with mTg [11,12]. These observations imply that mTg–treated breads induce immunogenic peptides that react with human IgA.

It is worth mentioning that the immunogenicity of the mTg cross-linked dietary products was lately expanded to more endogenous, in-vivo immune reactivity towards mTg-gliadin neo-complexes in active celiac disease patients [13]. It happens that mTg is immunogenic in children with celiac disease and by complexing to gliadin its immunogenicity is enhanced. Anti-mTg neo-epitope IgG antibodies correlate with intestinal damage to a comparable degree as anti-tTg neo IgA. mTg and tTg display a comparable immunopotent epitope. Thus, mTg-neo IgG might represent an additional marker for gluten sensitive patients.

Since bacteria secrete mTg, in the following section the bugs that are associated with the major neurodegenerative conditions are listed.

5. Relationship between Neurodegenerative Diseases and Bugs

Table 1 summarizes the microbes that were associated with major neurodegenerative diseases.

Table 1. The microbes that were associated with major neurodegenerative diseases.

Disease	Bacteria	Reference
Parkinson's disease	Helicobacter pylori	[14,15]
	Mycobacterium paratuberculosis	[16]
	Provotellaceae	[17]
	Enterobacteriaceae	[17]
	Nocardia asteroids	[18]
	Multi-bacterial overgrowth	[19]
	Dysbiosic microbes	[17,20,21]
Alzheimer's disease	Porphyromonas gingivalis	[22]
	Borrelia burgdorferi	[23]
	Chlamydophilia pneumoniae	[23]
	Spirochetes	[24]

6. Potential Pathogenic Pathway of Microbial Transglutaminase in Neurodegenerative Diseases

mTg is expressed and secreted in the human intestinal lumen. It represents one of the microbial protective mechanisms in the evolutionary struggle between bugs and us. Additionally, the recombinant mTgs that are used extensively by food processing industries and consumed as processed food and the swallowed probiotics which possess the mTg-encoding genes, represent an additional substantial source of mTgs lodged into the lumen of the gut. Following are some pathophysiological pathways that potentially connect the luminal mTg load to neurodegenerative and neuro-inflammatory diseases, thus reinforcing the gut-brain axis.

(1) Post-translational modifier of luminal proteins. This results in formation of higher molecular weight conjugates with different conformational, physical, electrical, chemical and immunogenic identities [1,3,5,6,25]. The transformation of naive peptides to immunogenic neo-epitopes is considered as a major driver of inflammation and autoimmunity.

(2) Being an anti-protease, mTg increases the protein stability against proteinases, thus diminishing the capabilities for the digestion of foreign or improperly folded proteins to eliminate them from the gut lumen, increasing the antigenic load [1,6].

(3) Infections as well as the cross-linked nutritional constituents between gluten and mTg, increase the gut permeability. In fact, mTg is a potential enhancer of tight junction permeability [5,6] and increased intestinal permeability is described in Parkinson's disease [26], disrupted behavioral responses [27], stress, trauma and inflammation [28], brain dysfunction [29] and central autoimmune diseases like multiple sclerosis [30]. The resulting leaky gut allows more immunogenic foreign molecules to enter the systemic circulation and induce strong immune responses, including neurodegenerative, neuro-inflammatory and autoimmune diseases [5].

(4) Parkinson's disease is a classical neurodegenerative disease with multiple gastrointestinal dysfunctions [19]. The oral drooling, swallowing difficulties, delays gastric emptying, increased intestinal permeability, the associated dysbiosis and the constipation that precedes motor abnormalities are some of them. Taking in account presence of pervasive α-synuclein deposition in the gastrointestinal tract and the pivotal role played by the enteric glial cells in the intestinal tight junction integrity and regulation [31], one can envision the bidirectional gut-brain cross-talks. The luminal mTg might enhance some of those mechanisms.

(5) Periodontitis or enteritis, intestinal dysbiosis or pathobionts can provide the brain with intact bacterial products or whole bacteria, virulence factors and inflammatory mediators due to daily, transient bacteremias. If predisposed genetic risks meet environmental risk factors in the brain, neuroinflammatory/degenerative disease may strike roots.

(6) Streptococcus suit serotype 2 is an important human zoonotic pathogen, causing septicemia, arthritis, endocarditis, meningitis and even acute human death. Although several virulence factors have been identified, most recently, a new pathogenic pathway was described for the mTg of the bacteria [32]. Notably, this new secreted mTg shares a common feature of active site cavity with eukaryotic Tgases. The antiphagocytic properties of this specific mTg suppress one of the major protective immune mechanism in human. More so, the same microbe was described recently to contribute to neuroinflammation and to enhance blood-brain barrier permeability [33,34].

7. Conclusions

It is hypothesized that the exogenous mTgs, secreted by the microbiota, especially in the dysbiotic configuration, are potential drivers of neurodegenerative and neuro-inflammatory diseases via the gut luminal PTMP. The massive use of mTgs in the processed food and the increasing use of probiotics including active mTg may be the additional contributors to the enhanced luminal PTMP. The substantial luminal activity of the mTgs, by cross-linking naive proteins, can potentially generate neo-epitopes that are not only immunogenic but may also be pathogenic by activating some harmful pathways in the cascade of chronic central brain diseases induction or progression.

Conflict of Interest

The authors declare no conflict of interests.

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