A history into genetic and epigenetic evolution of food tolerance: how humanity rapidly evolved by drinking milk and eating wheat

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Purpose of review
Human exposure to wheat and milk is almost global worldwide. Yet the introduction of milk and wheat is very recent (5000–10 000 years) when compared to the human evolution. The last 4 decades have seen a rise in food allergy and food intolerance to milk and wheat. Often described as plurifactorial, the cause of allergic diseases is the result from an interplay between genetic predisposition and epigenetic in the context of environmental changes.

Recent findings
Genetic and epigenetic understanding and their contribution to allergy or other antigen-driven diseases have considerably advanced in the last few years. Yet, environmental factors are also quite difficult to identify and associate with disease risk. *Can we rethink our old findings and learn from human history and recent genetic studies?*

Summary
More than one million years separate *Homo habilis* to today's mankind, more than 1 million years to develop abilities to obtain food by foraging in diverse environments. One million year to adjust and fine-tune our genetic code and adapt; and only 1% of this time, 10 000 years, to face the three biggest revolutions of the human kind: the agricultural revolution, the industrial revolution and the postindustrial revolution. With big and rapid environmental changes come adaptation but with no time for fine-tuning. Today tolerance and adverse reactions to food may be a testimony of adaptation successes and mistakes.

Keywords
allergy, genetic and epigenetic, human evolution, milk, oral tolerance, wheat

INTRODUCTION

Milk and wheat are today consumed by billions of people. Whether it is through consumption of regular milk, yoghurt or other dairy, whether it is bread, pita, pasta or beer, milk and cereals represent 50% of our global protein supply and is thus an essential part of our today nutrition. Interestingly enough, *Homo sapiens* (and possibly its pets like cats and dogs) is the only mammals drinking milk at the adult age. On a nutritional standpoint, breast feeding is not required after 3 years of age if other sources of protein and other nutrients are provided in sufficient amount. Naturally, young mammals have developed ability to digest the lactose of their mother's milk. Once not required anymore, an epigenetic mechanism turns off this ability by silencing the gene coding for lactase (*LCT*) [1]. *H. sapiens* between 10 000 and 5000 years ago has developed its ability to raise farm animals for their meat, skin and milk. Persistence of milk consumption through the intake of animal milk in childhood and adulthood, in region where climate allowed animal to survive, has led to several major societal changes as animal farming was offering advantage over obtaining food by foraging [2]. Meanwhile, it was leading to persistence of lactose in the human diet [2]. Three main ways to take advantage of this lactose could be envisioned: first, delay the epigenetic turn off; second, create mutation to avoid epigenetic silencing or to enhance its expression; third, the absence of lactase allows to feed our microbiota with undigested lactose and to harvest the

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KEY POINTS

- Wheat and milk appeared in our nutrition 10 000–5000 years ago, it is a relative recent introduction in our diet.
- Humanity has spread mutations acquired 5000 years ago along with farming and sedentarization, allowing lactase persistence and increasing immune competences to survive societal contacts.
- Today, farming exposure early in life is associated with a decrease risk of developing allergic disease such as asthma.
- Environmental changes may increase the pressure on long-acquired existing predisposing genetic variants, increasing risk of developing food allergy.

energy through microbial metabolites. These three mechanisms certainly coexist today. Significantly, numerous mutations leading to lactase persistence have been identified in the population derived from the first dairy farmer regions: Northern Europe, Middle East and Central Africa. These regions are associated with the highest prevalence of lactase persistence in the population [1]. These mutations allow persistence of lactase expression by intestinal cells as they allow better binding or transactivation of transcriptional factors through mutations in the lactase persistence locus (*MCM6* gene) upstream of the *LCT* gene [1]. However, in central and northern Portugal, the lactase variant −13910C>T (rs4988235) and more specifically the genotype CT/TT has been associated with BMI, fat mass and weight showing how a past variant acquisition may be associated with current diseases [3▪▪].

Significantly, those mutations are recent as most of them seem to arise around 7000–6000 years ago and have spread to an allele frequency up to 98% in specific populations [2]. Lactase persistence is a textbook example of genetic convergence in divergent populations, the acquired functionality being directly in adequacy with the environment [1]. This example is for sure one on the most recent and striking genetic signature of natural selection characterized so far in the context of food source being a driver for change [1]; whether lactase persistence truly had a direct evolutive advantage or is just a marker of animal farming benefit is still to be demonstrated.

But with mutations appearing 5000 years ago, and less than 50% of the global population being lactase persistent today, one could think that recent changes in disease prevalence in the last decades, like food allergies, cannot be only due to newer rare genetic variants acquisition. On the contrary, if mutations link to allergy are older, increased allele frequency of these variants in the population due to past population bottleneck and recent population growth, [4–6] may alone not explain the rise in food allergy observed only in the last four generations.

A CHANGING WORLD: THE AGRICULTURAL REVOLUTION SHAPED OUR GENES

Less than 10 000 years ago, during the Neolithic revolution, we have domesticated farm animals and cultivated crops. This was the beginning of drastic changes in the way we would eat, with the introduction of milk and cereals in our daily consumption. This represented a huge advantage for the population being able to stock food for less productive seasons and ensure stocks of available proteins. Yet, from family clans, we have grown small cities and even big cities with more than 1000 habitants as early as 8500–5700 years ago. Mureybet tell is a testimony of these changes. Located in Syria, Mureybet, 10 200 Before Common Era, was inhabited by gatherer-hunters who started sedentarization. About 10 000 years ago, this village was seeing the arrival of rye and barley cultivate and animal farming (cheeps and goats), rising population up to 500 inhabitants.

This sudden close interspecies and intraspecies proximity may have led to transmission of diseases whether it is from animal-to-human or human-to-human transmission [7]. The immune system was facing new challenges, meanwhile the genetic code was creating new variants to test. At this time, a strong immune system might have been a positive selection pressure amplifying allele frequency of immune performant variants in the population. A strong and potent immune system may have thus be transferred from 5000 years ago up until now. Yet another big revolution was announced, arriving in the late 16th century: the industrial revolution.

THE INDUSTRIAL AND POSTINDUSTRIAL REVOLUTION: NEW SOCIETAL CHANGES

As the industrial revolution, the human population has not drastically changed its pool of genetic mutation as too recent but definitely has changed what we are exposed to. Hygiene, sterilization and medicine are somewhat new concepts and saved many lives from infections. With industrialization, work force is needed, and cities became bigger to the point that farm animals are now outside of cities and decrease exposure to earth is starting and increase exposure to pollution. Our performant immune system sees less infectious challenges and new type

of exposure possibly leading to a rise in inflammatory and autoimmune diseases: this is what was described as the hygiene hypothesis [8]. Epidemiologic studies have indeed associated exposure to animal farm environment early in life and the risk of developing allergic symptoms in several studies [9,10].

This hypothesis is certainly only one part of the explanation as not fully explaining why food allergy prevalence is rising only in the last generations. What else happened in the last 100 years? The last 100 years have been marked by societal changes including life style changes and more importantly nutritional changes. The westernized world, which exemplified these changes and which first endorsed this new life style, was also the first to suffer from the rise in none communicable diseases such as food allergy.

Our recent ancestors were not eating as much food originating from other continents. Yet, today the apparent increase in food diversity is a lure on a worldwide scale. Although export–import have exposed population to news type of food, the actual worldwide amount of diverse cultivate have dropped drastically in the last 5 decades. This concept of a decrease food diversity on a worldwide scale is certainly important as on an individual scale, diversity of food introduction early in life in infant has been associated with a decrease risk of developing food allergy and atopic dermatitis [11,12,13▪▪], alighting also the role of the environmental exposure in our genetically predisposed population.

According to Food and Agriculture Organization of the United States (www.fao.org): 'Today, 75 percentage of the world's food is generated from only 12 plants and five animal species. Of the 4 percentage of the 250 000 to 300 000 known edible plant species, only 150 to 200 are used by humans. Only three – rice, maize and wheat – contribute nearly 60 percentage of calories and proteins obtained by humans from plants'. First cooked 10 000–8000 years ago, wheat is the perfect example. Culture techniques, environmental changes and crop selection have allowed to increase yield of production allowing to decrease the Spanish culture area between 1925 and 1997 while keeping a constant, if not increased, production [14]. Wheat consumption is unsurprisingly associated with occurrence of celiac disease. Some of the variant responsible for celiac disease predisposition are part of our immune system and are thus highly present (up to 30%) in today healthy population. However, only 1% of the population will develop celiac disease suggesting again that the onset of the disease is either linked to other variants and/or environmental triggers. Similarly, genetic study of the recently increasing in non-coeliac-wheat-sensitivity will certainly teach us more on how variants, possibly acquired after wheat was introduced, are associated with disease susceptibility. The dissection of what were the advantages for the population to acquire and/or maintain such variants and what are there consequence for the population in the current environment will be key for the understanding of the cause of diseases.

EPIGENETICS OF TOLERANCE AND FOOD ALLERGY

The definition of 'epigenetics' term is today still debated. Defined in 1942 by Conrad Waddington as 'the branch of biology that studies the causal interactions between genes and their products which bring the phenotype into being', several others will try to define epigenetics, such as Holiday followed by Wu and Morris 'the study of changes in gene function that are mitotically and/or meiotically heritable and that do not entail change in DNA sequence'. As such, the definition has evolved to include the concept of heritability, leading to confusion in the scientific literature of what is or not epigenetics, as nicely described by Deans and Maggert [15]. Only few studies have to date linked allergy to epigenetic marks and the necessity or proof of heritability was not necessarily considered as based on prior definitions. Liang *et al.* [16] have identified in peripheral blood leukocytes that low methylation in 36 loci was associated with IgE levels and the top three loci accounted for 13% of IgE levels. Some of the loci associated with allergic sensitization include genes highly involved to the allergic response such as *IL4* and *IL13* or *IL4R* and *IL5R*, but also genes not directly related to immune function [17]. In a Dutch cohort of cow's milk allergy patients, Petrus *et al.* have recently identified that blood DNA from cow's milk allergic patients was characterized by a general hypermethylation compared with control children, whereas this effect was absent in the tolerant group. Methylation differences were, in the region of genes (*DHX58*, *ZNF281*, *EIF42A* and *HTRA2*) known to be part of immunological pathways and relevant to allergic diseases [18▪▪]. Numerous studies have looked at the contribution of epigenetic changes in forkhead box protein 3 (*FOXP3*) promoter in preclinical experiments. Syed *et al.* have nicely identified epigenetic modification in a pilot human trial with peanut allergic patients undergoing immunotherapy. The authors showed significant 5'—Cytosine—phosphate—Guanine—3' site demethylation of *FOXP3* gene in peanut specific regulatory T cells of tolerant patients compared with the nontolerant patients [19]. These results suggest that sensitization, allergic status and tolerance acquisition also are controlled by nongenetic determinants [17].

GENETIC OF ALLERGIC DISEASES AND ENVIRONMENTAL PRESSURE

Numerous studies have shown association between genetic variants and allergic diseases. Most recent studies have identified multiple loci common to different allergic diseases. In the past, the heritability of allergic diseases in family members have drawn researcher's attention to genetic heritability only and lower attention has been given to the influence of the shared environment [20]. However, with the strong and possibly increasing environmental pressure on the predisposing allergic variants, the genetic heritability (disease expression due to DNA sequence) might increase in the future. It is thus very difficult to dissect out the contribution of the genetic and of the environment but twin studies described below have greatly contributed to this understanding [20].

Most of the data in the field of allergy have focused on the genetic heritability accounting for 60–85% of the disease heritability depending on the studies. In atopic dermatitis, some studies have suggested that genes accounted for 82% and non-shared environmental factors accounted for 18% of the individual susceptibility to develop atopic dermatitis [21]. In food allergic patients, like peanut allergy, heritability of peanut allergy was estimated at 81.6% (95% confidence interval 41.6–99.7%) for a general population prevalence of 0.4% [22].

In asthma and allergic rhinitis [23], consistent results positioned genetic factors as accounting for 60–80% of its susceptibility. Yet in some of the prior studies, the family history of allergy may have been underestimated and the shared environment effect unsuspected most likely, the high prevalence of these diseases complicate the model estimations. Recent twin study in a more rare allergic disease have indeed led to surprising results regarding the contribution of the environment and shared familial environment, compared with the genetics. In the context of eosinophilic esophagitis (EoE), a twin cohort have emphasized the prominent influence of shared environment (81.0%) compared with additive genetic heritability (14.5%) [24]. The low prevalence of the disease in the population and the discordance of prevalence between siblings and dizygotic twins have certainly highly contributed to this finding [25]. This is of particular importance as common loci have been identified between atopic dermatitis and EoE patients and similar environmental factors may influence these loci [26]. In addition, some of the variants associated with specific allergic diseases have a gene expression highly specific for the diseased tissue and have shown to be highly associated with the disease and highly relevant in the cause of allergic disease (filaggrin for atopic dermatitis [27] and *CAPN14* for EoE [28–31]) explaining why a common genetic predisposition may developed into different allergic diseases depending on the presence of other tissue-specific variants.

In allergic diseases, healthy individuals may carry the culprit variants, whereas diseased patients may carry the variant associated with healthy state

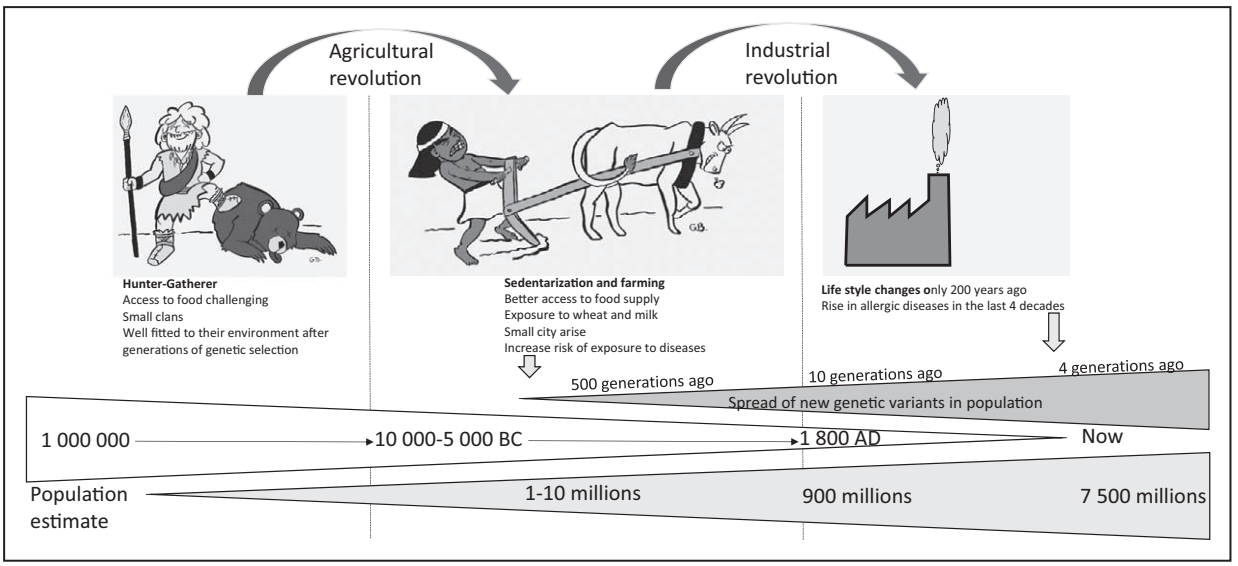

FIGURE 1. How humanity rapidly evolved by drinking milk and eating wheat. From hunters-gatherers to farmers, to home office, the mankind had to adapt to environmental changes extremely rapidly. With the population growth explosion, adaptation acquired 10 000–5000 years ago have spread into current population and may be at least in part responsible of today's health and diseases. Anachronisms may be present in the illustrations.

rendering the genetic cause difficult. These variants today have a poor sensitivity and specificity as biomarkers of allergic diseases, but they highly contribute to the disease pathogenesis understanding. Epigenetics insights will certainly help filling the gap of the missing heritability (e.g. heritability not explained by genetic variants) of the allergic diseases in the near future [20,25].

CONCLUSION

Between the genetic and the environment lays epigenetic allowing accessibility to the genetics. Genetic and epigenetic changes go differently. Genetics is printing in the genetic code long-term changes to be transmitted lastingly. Epigenetics more rapid and labile, tests how changes in the way to interpret this genetic code could be of any advantage rapidly while possibly being transferred to the next generation. For both mechanisms, the constant pressure of the environment allows these changes to be drawn into our genetic and epigenetic roadmap. The last 10 000 years have drastically transformed our environment and nutrition (Fig. 1). Our current genetic code is a testimony of these changes and our capacity to adapt. Current food allergy increased prevalence may just be a representation of how the environment we are currently living in pressures the genes our ancestors acquired to better fit their environment 5000–10 000 years ago. Food allergy may show how mankind is inadapted to our changing world.

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Conflicts of interest

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