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## Perspective

# Curious case of the history of fermented milk: tangible evidence

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Humans are often curious about why and how early humans had given up raw food in diet? And which came into being first, cooking or fermentation? It was posited, based on the skull and jaw structure, that early hominids spent a lot of time eating food vis-a-vis *Homo sapiens*. Around 2 million years ago, a significant change occurred, and *Homo* species started spending less time chewing food, implying that they abandoned eating raw food and started utilizing fire as a tool [1]. One possibility could be that they started eating cooked food because it was relatively tender and easy to digest. However, it would require a pot, an oven, or a pit to cook or live near volcanoes. If these were not available then, fermentation could be a robust possibility for this change as it tenders the food, develops fascinating flavor, and stymies putrefaction.

Origin of fermented milk. Fermented milk products are widely produced and consumed across the globe. For thousands of years, milk fermentation has been practiced. It is one of the oldest techniques to extend the shelf-life of milk. Although it is insurmountable to directly unravel the exact origin of fermented milk, evidence from some indirect methods (ancient rock arts and fat analysis of prehistoric potsherds) suggested that these could date back as far as 10,000 years ago, when humans transformed their lifestyle from hunter-gatherers to farmers [2]. This change led to the domestication of several ruminants, and this transition likely occurred at different times in different regions. However, mounting archaeological evidence suggests that various civilizations were adept at agriculture and fermented milk production. It is very likely that milk fermentation might be originated from the Middle East and the Balkans, and the evolution of fermented milk might be attributed to the culinary skills of local inhabitants.

Andrew Sherratt's model [3] about the secondary product revolution had unraveled that the exploitation of animals for use of secondary products would have taken place in Mesopotamia and later on spread to Europe, North Africa, and Asia in the early Neolithic era. However, several food historians argued against the model. They argued that cattle could not be milked without having a calf. Moreover, in the early ages, it was unlikely that there was enough milk both for maintaining continuous lactation and human consumption due to limited breeding knowledge. However, recent advancements in archeology have refuted these suppositions. The

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oldest reported chemical evidence for the processing of milk into fermented milk products dates back to the Early Bronze Age in Xinjiang, China [4]. Other indications, including actual preserved pieces of cheese, whey strainers, and recipes for cheese production, were found in northern Europe, the Middle and Near East, and the Mediterranean basin [5]. The development of lipid biomarkers and stable isotope proxies that allow the identification of lipid sources from absorbed organic residues in pottery vessels allows direct analyses of specialized uses of vessels, the antiquities of dairying, and related processing activities [6], enabling the detection of dairy or carcass fat residues in pottery sherds. Evidence from faunal assemblages showed a good match between the incidence of dairy fat in pottery (implying a strong dietary dairy fraction) and a milking herd, inferred from the animal bones. Moreover, dairy fat was more likely to be found in the smaller pots while carcass fat was found in the larger ones [7].

Recently, archeologists have unearthed 5000-year-old curd molds in the Middle East. A Sumerian bas-relief of 5500-year-old, found in El Obeid, depicted the milking of cows and milk curdling [8]. In ancient Egypt, 4000-year-old bas-reliefs on tombs also depicted the milking of cows [5].

In Pakistan, Harapan civilization excavations revealed 5000-year-old vases pierced with holes containing residues of goat milk products. Minimal presence of dairy in Harapan vessels compared with other lipid residues found elsewhere was incongruous. The reason for this could be that dairy consumption was limited to few people and/or used vessels were made from organic materials, unlikely to survive. Lipid analysis of 5000-year-old sherds from El Portalón de Cueva Mayor archaeological site (Burgos, Spain) revealed evidence of cattle farming. Novel archeozoological analyses revealed that the most abundant lipid residues identified were dairy fats, followed by ruminant/non-ruminant adipose fats [9].

In northern Africa, the identification of milk fats in ceramics suggested that early pastoralists were utilizing milk around 7200 and 6600 years ago in Libya and Sudan, respectively. And around 5000 years ago, in eastern Africa, early herdsmen were utilizing milk, as proven by a lipid residue study of ceramics in Kenya and Ethiopia [6].

Moreover, in Anatolia and Libya, excavations have revealed milk residues dated back 6000 and 5000 years, respectively. Dairy residues dating back to 5000 years have been detected in large numbers in Britain [7]. Ceramic faisselles that are around 6000 years

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old have been found to contain cheese residues in the Swiss Jura, Romania, and Hungary. Therefore, dairy products consumption was already well-established around 8000 years ago [10]. Contrarily, lipid residue analysis from ceramics has aroused serious concern. Specifically, the presence of milk fats in ceramics does not necessarily mean regular dairy consumption by humans, as dairy products could be stored for medicinal/ritual purposes. Therefore, it is imperative to explore hard evidence for the prehistoric consumption of milk/fermented milk. Proteomics of ancient dental plaque is an advanced technique to exploit milk consumption by humans. Previous studies generally focused on historical archeological sites instead of prehistoric materials. Bleasdale et al. [11] identified dairy proteins from dental calculus of the ancient Africans, proving that milk was being consumed by humans in Africa (Sudan and Kenya) at least 6000 years ago. A joint group of researchers from China and other countries found that the earliest inhabitants of northwest Xinjiang, China, had genomic origins dating back as far as 5000 years [4]. Genomic data collected from mummies found in Xinjiang's Tarim and Junggar Basin of Xiaohe culture exhibited strong evidence of milk proteins (from sheep and goats) in their dental calculus, indicating a reliance on dairy pastoralism at the site since its first inhabitants arrived [12]. Recently, paleofecal (3000 years old) analysis represented the earliest known evidence for cheese consumption as part of diet in Europe, which also indicated the sophisticated culinary traditions in protohistory [13].

Livestock farms were found on the shores of the Mediterranean in the Middle East and Europe that were 8000 and 6000 years old, respectively. They were "hunter-farmers raising" rather than real "farmer-herders", developing techniques appropriate for dairy exploitation of the first cattle that were not domesticated. In fact, it was milk consumption, instead of meat consumption, that led to the domestication and spread of livestocks to the Mediterranean basin [8].

Transmission of fermented milk products. The specific community and place of development of the first fermentation are still unclear. but fermentation conquered the world prehistorically. Dairying practice transitioned parallel to adaptation in pastoralists of Africa. Europe, and the Middle East. Emerging archeological evidence has unraveled that from sub-Saharan Africa, fermented products arrived in Egypt via the Nile Valley and then percolated to Mesopotamia (Iraq) and Asia where they were settled around 6000 years ago. Fermented milk was spread to central Asia through the Silk Road. It is very likely that the technique of cheese production from ruminant milk was learned from descendants of the Afanasievo and later on introduced to Xinjiang, China, via Inner Asia Mountain Corridor neighbors around 5500 years ago [12]. Silk road was of paramount importance for its role in exchanges of food fermentation knowledge, especially cheese and bread making, across Eurasia. This is the reason for the same/similar model of fermentation in different parts of the world. There is tangible evidence supporting that dairying techniques were brought from the Middle East rather than the indigenous domestication of cattle.

Domestication of cattle from Southwest Asia was transitioned to eastwards across the Eurasian steppe into Central Asia. Recently, biomolecular identification of dairy lipids from ceramics (collected from Neolithic Anatolia and eastern Europe, and Kazakhstan) has substantiated the speculation of dairying in Southwest Asia during the Copper and Bronze Ages [14]. Around 10,000 years ago, cheese was developed by the nomadic populations of Central Asia, and after 4000 years of development, the cheese-production tradition was transmitted to Africa and Western Europe. These fermentation techniques were created by nomadic/semi-nomadic peoples of Central Asia, transferred from Northeast Asia to West Africa, with contributions from Persia, the Ottoman Empire, and the Arabs [14].

At first, cheese fermentation was spontaneous, without adding milk curdling enzymes. Milk was autochthonously cultured with bacteria present naturally in the air, on the udders of animals, and in the same area where it was coagulated. The ease of manufacture, transport, and storage made it the ideal food, both among the gypsies of Asia and among pastoralists in Africa. As we also known, in Mongolia, a large part of diet comprises various fermented milk products prepared during summertime [14].

A recently published study [15], based on fossilized teeth of Bronze Age skeletons, suggested that more than 5000 years ago, Yamnaya nomads migrated out of Russia and Ukraine on their ox-drawn wagons, and left genetic signatures in populations from Mongolia to Hungary while expanding across Eurasia. Protein analysis of dental calculus suggested that dairying and herding were the key strategies adopted to transform rapidly from hunter-gatherers into nomadic herders. The main powerful weapon for their successful expansion was fermented milk along with the use of wagons to transport long distances.

These populations, possibly, spread to Asia (e.g., China) around 4000 years ago, as evidenced by the uncovering of dairy proteins of goat, sheep, and cattle in dental plaque of mummies from cemeteries in Tarin Basin. Such findings suggested that the earliest settlers in Tarin Basin (Xinjiang, China) were consuming dairy products, and they had adopted new ideas and culture, such as irrigation system and herding ruminants for milking [12]. However, a connecting thread is still missing regarding how, where, and from whom they acquired these techniques. In the future, further intercontinental and regional studies should consider addressing the complex relationship between cultural exchanges and mechanisms of food choices.

Lactase persistence (LP) conundrum. There is an over 4000-year gap between the earliest evidence of dairying and the first evidence of lactase gene mutation (to digest lactose) in the world, hitherto. DNA analysis of prehistoric human bone samples suggested that, around 7500 years ago, a genetic mutation took place in the European population, allowing persistent lactase secretion in adulthood. Almost 90% of Europeans have inherited this mutation. which is almost absent among East Asians. It was hypothesized that this mutation occurred only after cattle domestication, and it was first occurred in Africa, followed by the Middle East and Europe [16]. The most common form of the mutation was of Caucasian origin in northern European populations, occurring around 6000 years ago. Genes on which this mutation took place are also present in Central Asia, from the Caucasus to Pakistan and as far as Mongolia, the Middle East, and sub-Saharan Africa. All these regions have a very old tradition of fermented milk consumption. Populations with the same mutation share the same dairy culture throughout their history (domesticate the same animals) and belong to the same cultural sphere. Presumably, some unknown reasons caused the mutation in humans, enabling lactose digestion as a natural consequence and initiating the practice of animal breeding and domestication for milk production. Conversely, recent evidence suggested that lactase alleles first appeared in the African population, and Africans have the highest diversity of LP alleles in the world. Interestingly, many Europeans and Mongolians were consuming dairy products thousands of years before the emergence of LP alleles [17].

For example, Mongolian pastoralists typically obtain more than 30% of their energy from dairy products. Surprisingly, 95% of them are lactose intolerant. Moreover, molecular analysis of proteins from 9000 years old sherds from Turkey revealed the domestication of cattle; however, LP in this region appeared around 5000 years ago. In tandem with this, DNA and dental plaque analysis of 5000-year-old mummies found in Xinjiang, China, also confirmed that, genetically, they were not lactase persistent [12]. Thus,

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prehistorically dairy pastoralism in Asia spread without the LP genotypes [12], contradicting the hypothesis of a causal link between dairying and LP.

Another hypothesis is that the warm climate in Africa was conducive for milk fermentation, and humans were consuming fermented milk rather than fresh milk. Fermented milk has a low/ negligible quantity of lactose that did not require lactase for digestion. Eventually, this usual consumption of fermented milk favored the genetic mutation. Ancient DNA analysis found that the Yamnaya had no genetic ability to metabolize milk. Therefore, there is a strong possibility that the Yamnaya also consumed fermented dairy products with no/low lactose, like modern Mongolians [15]. Otherwise, it is questionable that humans were continuously consuming milk that sickened them. Genetic changes would not have happened overnight but through thousands of years of evolution. Therefore, it could be hypothesized that humans utilized the process of fermentation long before the mutation took place. In addition, some milk residues found in sherds appeared to have been heated, suggesting some form of milk processing. In summary, early humans consumed yogurt and cheese-like products before drinking unprocessed milk. Fermentation was found from the beginning among cattle farmers, otherwise milk could not be consumed in adulthood [10].

In conclusion, more direct archeological evidence from Africa and the Middle East is needed to clarify the origin of fermented milk consumption. It remains unclear what drove LP gene mutation to high frequencies and how external fermentation and/or microbiome adaptations influenced the LP evolutionary process. More chronological information for early milk drinking and high-quality ancient molecular data on LP are required to explain the global spread of LP.

#### **Conflict of interest**

The authors declare that they have no conflict of interest.

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