PROJECT 01956/Evo.Res.Con.Edu

Unraveling the educational potential of the research and concepts of evolution

Evolution of lactose tolerance and intolerance in humans

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Evolution of lactose tolerance and intolerance in humans

Direction 4: Holistic approaches based on evolutionary Theory in the context of wellness, disease, treatment, and biological death.

Lactose tolerance and intolerance

Production of lactase in adults as an evolutionary adaptation

Key concepts:

Lactase, mammals, weaning, neolithic revolution, pastoralism, continuous production of lactase, lactose tolerance, lactose intolerance, selective advantage

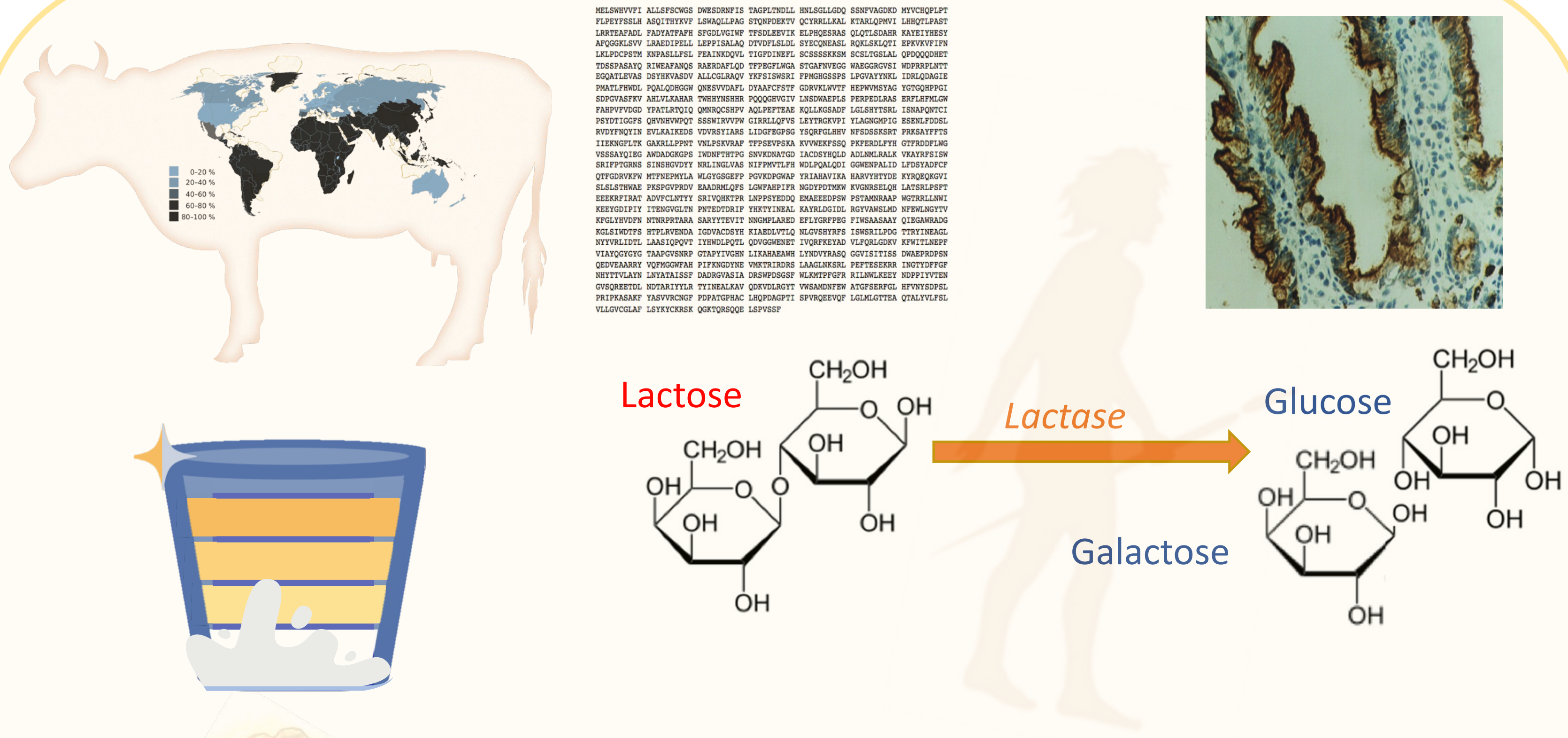
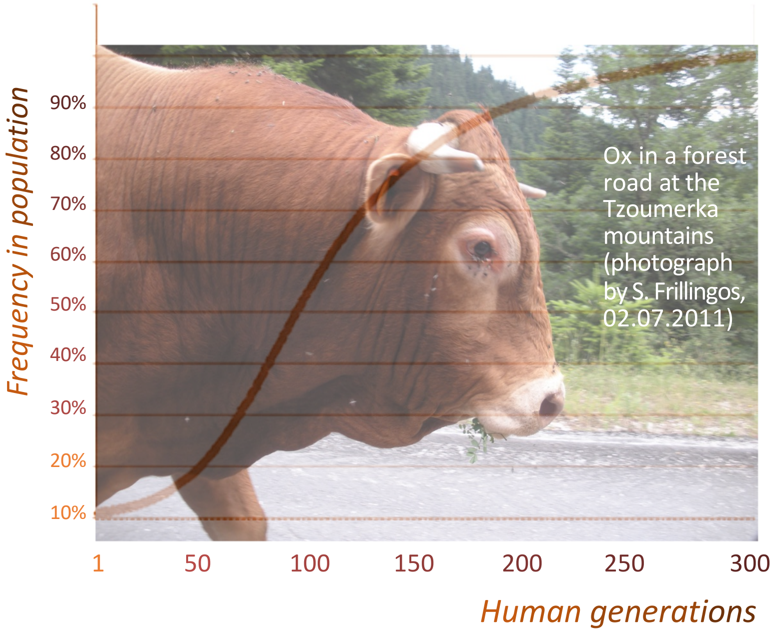


Figure composition from the sources:
https://www.freepikcompany.com
http://www.evo-ed.com/lactase-persistence/biological-processes/
https://www.ucl.ac.uk/life-sciences/gee/gee-research-centres/research-groups/global-lactase-persistence-association-database-glad



INTRODUCTION

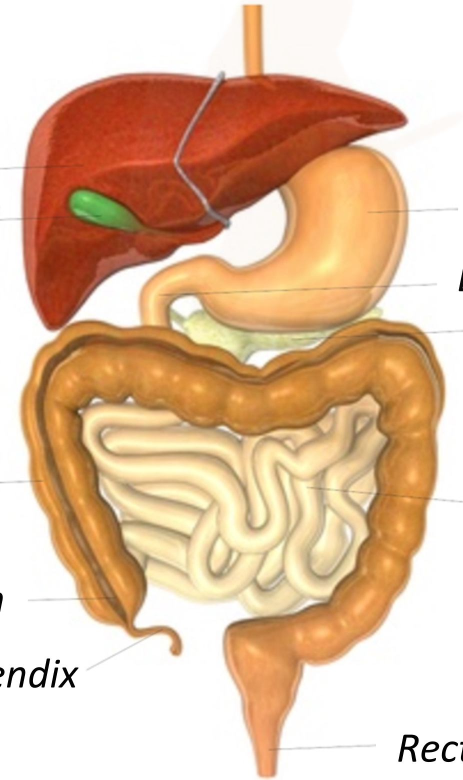
Humans are one of the few mammalian species in which many adult individuals retain the ability to absorb lactose, a milk sugar.

This ability is the ***result of a mutation*** that occurred millennia ago, which must have granted some adaptive advantage to individuals with this specific trait.

Let us examine how, when, and for what reasons that evolutionary event took place.

Biology of the digestive system

- Intestinal cells (which coat the inside of the digestive tract) are responsible for the breakdown and absorption of nutrients from food in the small intestine.

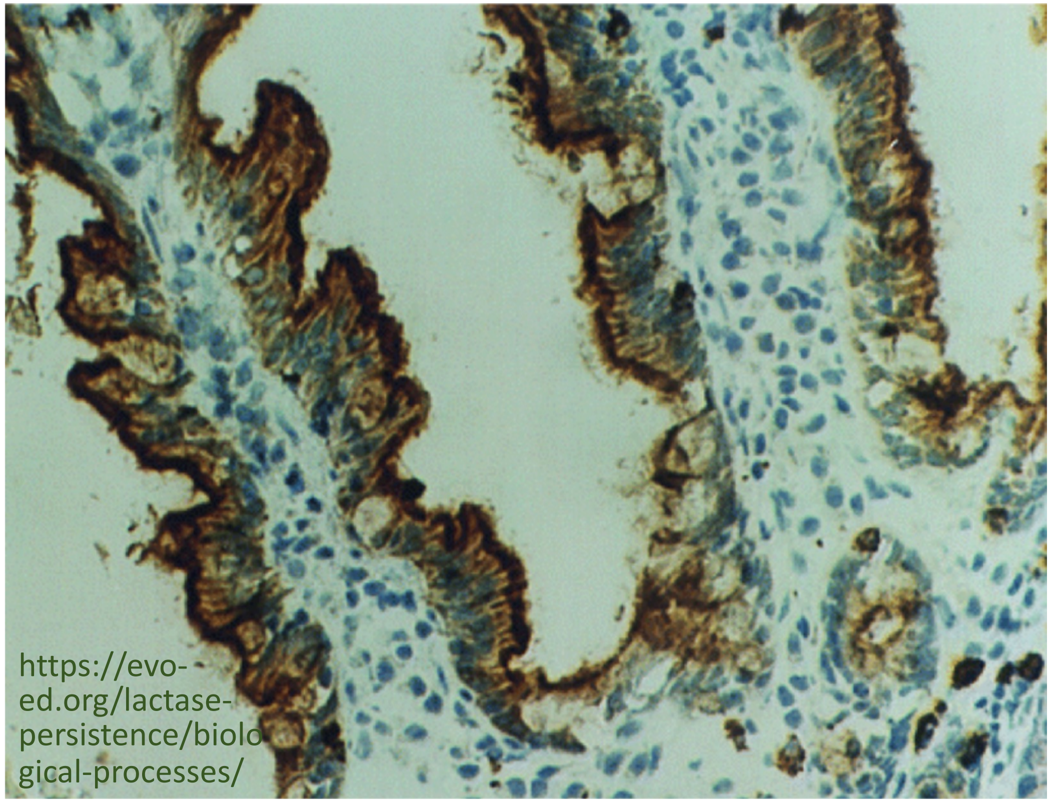


First food: mother's milk

- The intestinal cells of all infant mammals are characterized by high concentrations of the enzyme *lactase* during infancy, when milk is the main source of nutrition.

Where is lactase detected?

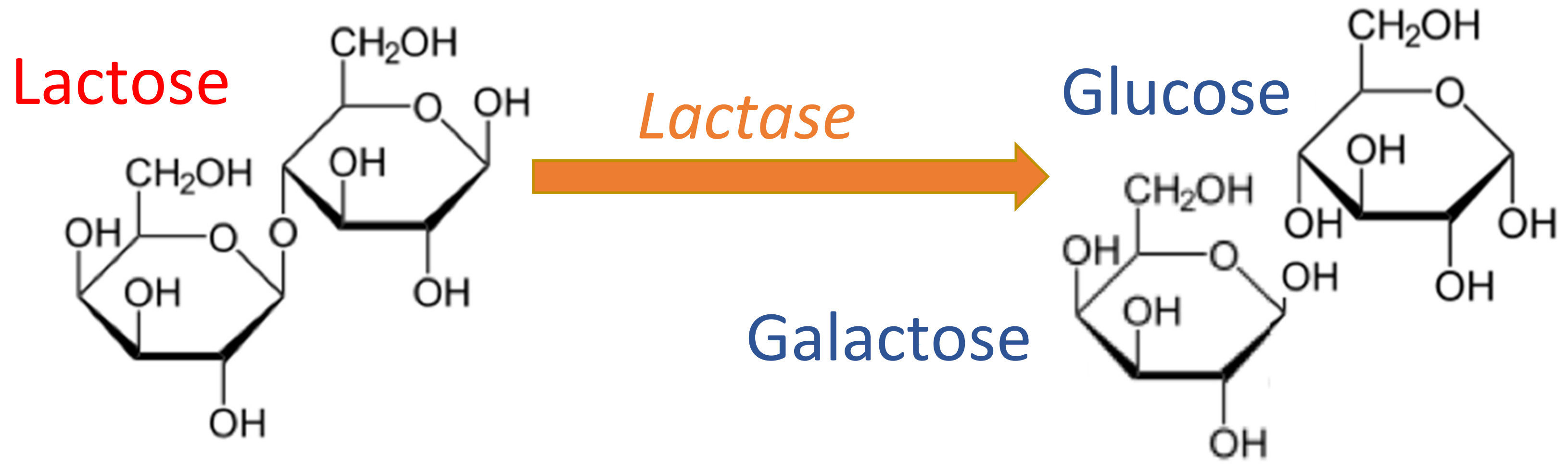
- In the protrusions below, we can observe the so-called “brush border”, the inner side of the small intestine, facing its content. It is there that lactase is detected.



Lactase is labeled brown.

Lactase "unlocks" an energy source

- Lactose is a disaccharide found in milk.
- Lactase breaks down lactose into two monosaccharides, glucose and galactose.
- These simple sugars can be absorbed by small intestine cells and used as an energy source.



Lactase regulation

- Nearly all known mammals -including **65%** of humans- show a decrease in lactase biosynthesis in the years after weaning.

- The regulation of lactase biosynthesis after weaning is the main factor that distinguishes individuals with **lactose tolerance** from those with **lactose intolerance**.

Lactase regulation

The decrease in lactose synthesis after weaning is probably a matter of cellular-level energy conservation:

- ❑ Energy is needed to produce any enzyme, including lactase, the enzyme required for digestion of milk.
- ❑ Mammals do not usually consume milk after weaning.
- ❑ Without milk consumption, the energy expended to produce lactase would mean a waste of energy on the cellular level.

Therefore, over time, the energy-saving option was selected: reducing lactase production after weaning.

Lactase is not produced in adults

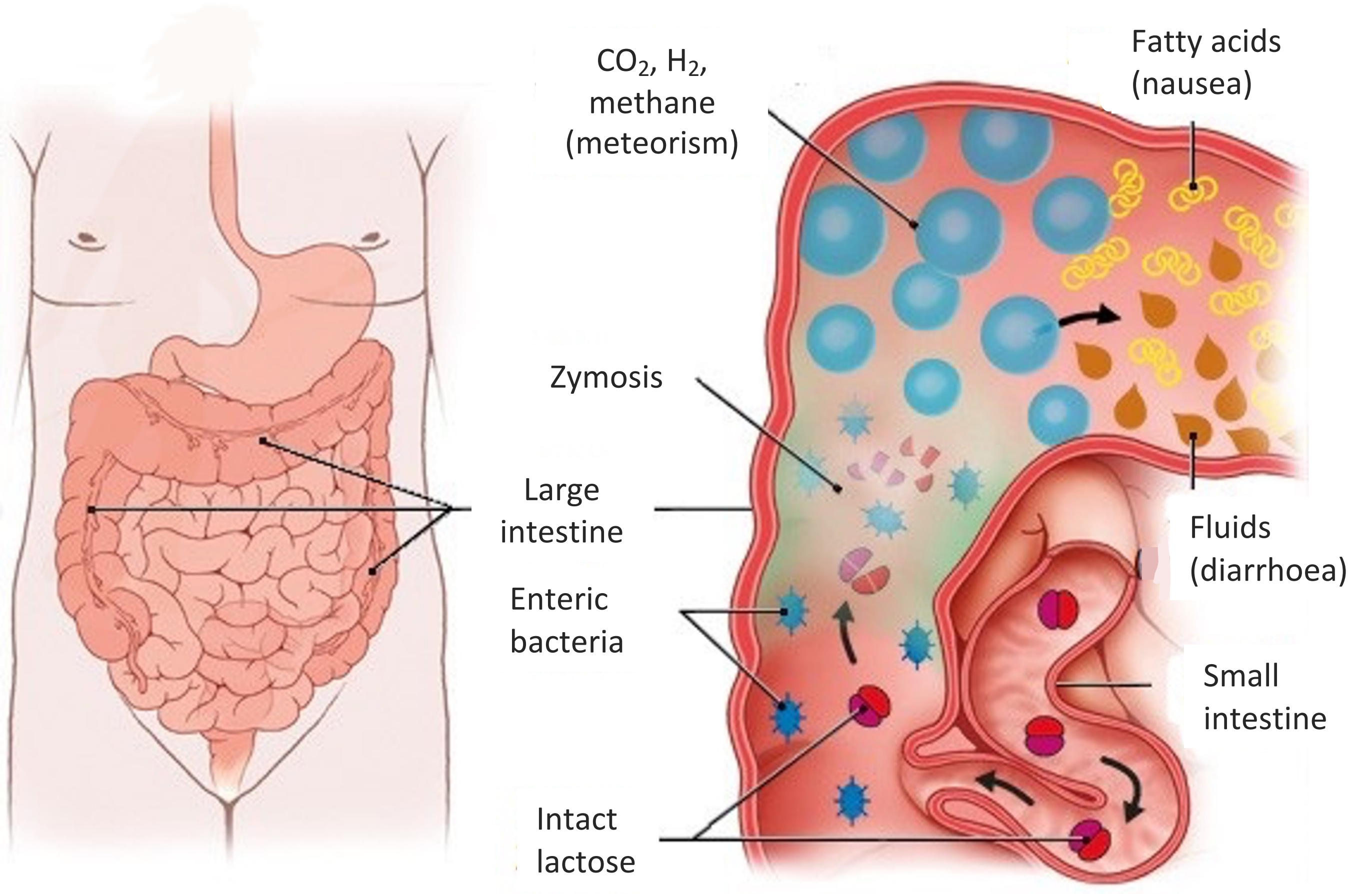
What will happen to humans who no longer produce lactase if they continue to consume milk in adulthood?

Lactase is not produced in adults

If undigested lactose passes into the large intestine, the individuals will show symptoms of lactose intolerance:

1. Increased concentration of saccharides in the large intestine creates an osmotic gradient, during which water enters the intestine. This causes cramps and diarrhea.

2. Large intestine bacteria absorb lactose as food, creating gaseous byproducts like methane, carbon dioxide and hydrogen. This leads to accumulation of gases and meteorism.



Continuous production of lactase

On the other hand, **35%** of humans continues to produce lactase after weaning, and are therefore capable of consuming milk and other dairy products in adulthood.

What happens on molecular level?

- Retaining of lactase production, the opposite of lactose intolerance, is the result of an evolutionarily preserved *mutation* in the regulatory mechanisms for the production of lactase mRNA.

- This mutation, which is connected to the continuation of lactase production into adulthood, consists of a change in a single nucleotide (SNP, single nucleotide polymorphism).

Lactase biosynthesis

The human lactase gene (LCT), composed of 55 thousand base pairs, is part of the second chromosome, with 17 exons.

```
MELSWHVVFI ALLSFSCWGS DWESDRNFIS TAGPLTNDLL HNLSGLLGDQ SSNFVAGDKD MYVCHQPLPT FLPEYFSSLH
ASQITHYKVF LSWAQLLPAG STQNPDEKTV QCYRRLLKAL KTARLQPMVI LHHQTLPAST LRRTEAFADL FADYATFAFH
SFGDLVGIWF TFSDLEEVIK ELPHQESRAS QLQTLSDAHR KAYEIYHESY AFQGGKLSVV LRAEDIPELL LEPPISALAQ
DTVDFLSLDL SYECQNEASL RQKLSKLQTI EPKVKVFIFN LKLPDCPSTM KNPASLLFSL FEAINKDQVL TIGFDINEFL
SCSSSSKKSM SCSLTGSLAL QPDQQQDHET TDSSPASAYQ RIWEAFANQS RAERDAFLQD TFPEGFLWGA STGAFNVEGG
WAEGGRGVSI WDPRRPLNTT EGQATLEVAS DSYHKVASDV ALLCGLRAQV YKFSISWSRI FPMGHGSSPS LPGVAYYNKL
IDRLQDAGIE PMATLFHWDL PQALQDHGGW QNESVVDAFL DYAAFCFSTF GDRVKLWVTF HEPWVMSYAG YGTGQHPPGI
SDPGVASFKV AHLVLKAHAR TWHHYNSHHR PQQQGHVGIV LNSDWAEPLS PERPEDLRAS ERFLHFMLGW FAHPVFVDGD
YPATLRTQIQ QMNRQCSHPV AQLPEFTEAE KQLLKGSADF LGLSHYTSRL ISNAPQNTCI PSYDTIGGFS QHVNHVWPQT
SSSWIRVVPW GIRRLLQFVS LEYTRGKVPI YLAGNGMPIG ESENLFDDSL RVDYFNQYIN EVLKAIKEDS VDVRSYIARS
LIDGFEGPSG YSQRFGLHHV NFSDSSKSRT PRKSAYFFTS IIEKNGFLTK GAKRLLPPNT VNLPSKVRAF TFPSEVPSKA
KVVWEKFSSQ PKFERDLFYH GTFRDDFLWG VSSSAYQIEG AWDADGKGPS IWDNFTHTPG SNVKDNATGD IACDSYHQLD
ADLNMLRALK VKAYRFSISW SRIFPTGRNS SINSHGVDYY NRLINGLVAS NIFPMVTLFH WDLPQALQDI GGWENPALID
LFDSYADFCF QTFGDRVKFW MTFNEPMYLA WLGYGSGEFP PGVKDPGWAP YRIAHAVIKA HARVYHTYDE KYRQEQKGVI
SLSLSTHWAE PKSPGVPRDV EAADRMLQFS LGWFAHPIFR NGDYPDTMKW KVGNRSELQH LATSRLPSFT EEEKRFIRAT
ADVFCLNTYY SRIVQHKTPR LNPPSYEDDQ EMAEEEDPSW PSTAMNRAAP WGTRRLLNWI KEEYGDIPIY ITENGVGLTN
PNTEDTDRIF YHKTYINEAL KAYRLDGIDL RGYVAWSLMD NFEWLNGYTV KFGLYHVDFN NTNRPRTARA SARYYTEVIT
NNGMPLARED EFLYGRFPEG FIWSAASAAY QIEGAWRADG KGLSIWDTFS HTPLRVENDA IGDVACDSYH KIAEDLVTLQ
NLGVSHYRFS ISWSRILPDG TTRYINEAGL NYYVRLIDTL LAASIQPQVT IYHWDLPQTL QDVGGWENET IVQRFKEYAD
VLFQRLGDKV KFWITLNEPF VIAYQGYGYG TAAPGVSNRP GTAPYIVGHN LIKAHAEAWH LYNDVYRASQ GGVISITISS
DWAEPRDPSN QEDVEAARRY VQFMGGWFAH PIFKNGDYNE VMKTRIRDRS LAAGLNKSRL PEFTESEKRR INGTYDFFGF
NHYTTVLAYN LNYATAISSF DADRGVASIA DRSWPDSGSF WLKMTPFGFR RILNWLKEEY NDPPIYVTEN GVSQREETDL
NDTARIYYLR TYINEALKAV QDKVDLRGYT VWSAMDNFEW ATGFSERFGL HFVNYSDPSL PRIPKASAKF YASVVRCNGF
PDPATGPHAC LHQPDAGPTI SPVRQEEVQF LGLMLGTTEA QTALYVLFSL VLLGVCGLAF LSYKYCKRSK QGKTQRSQQE
LSPVSSF
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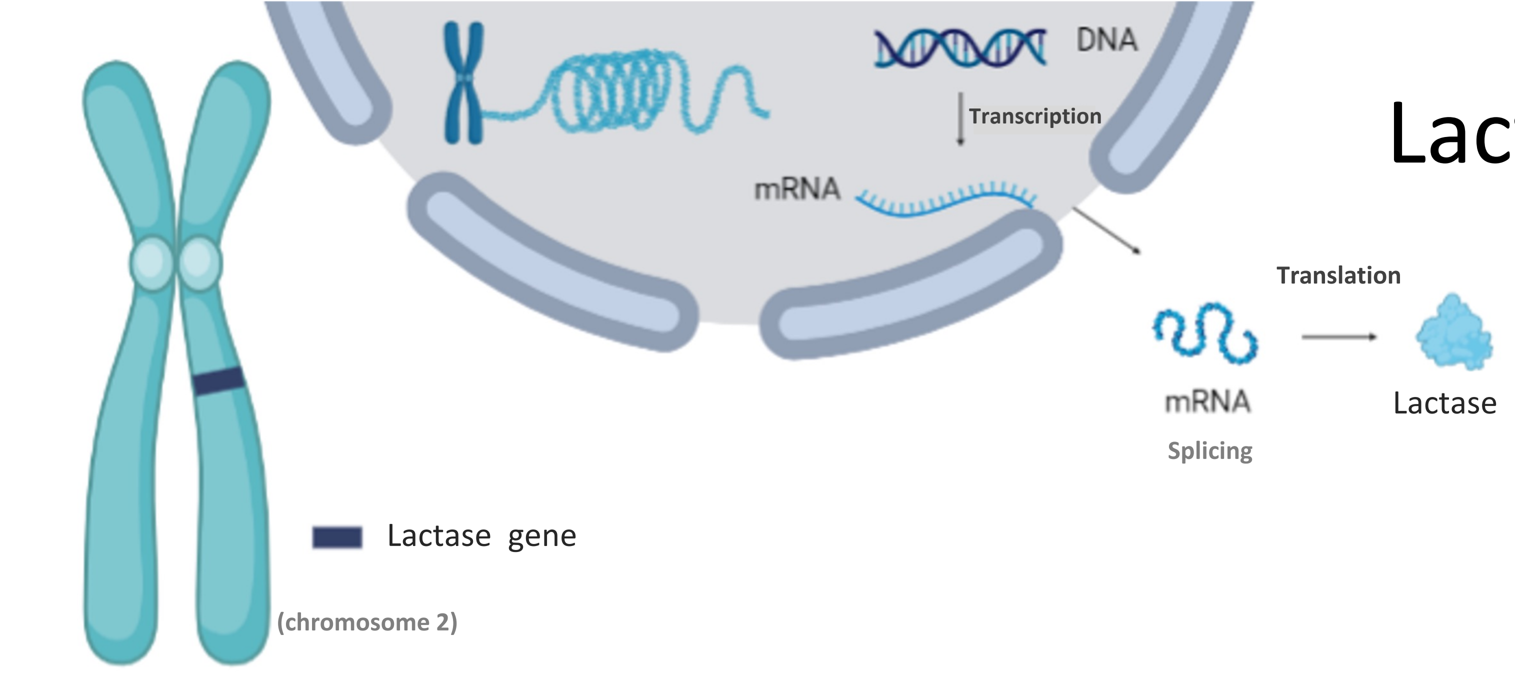
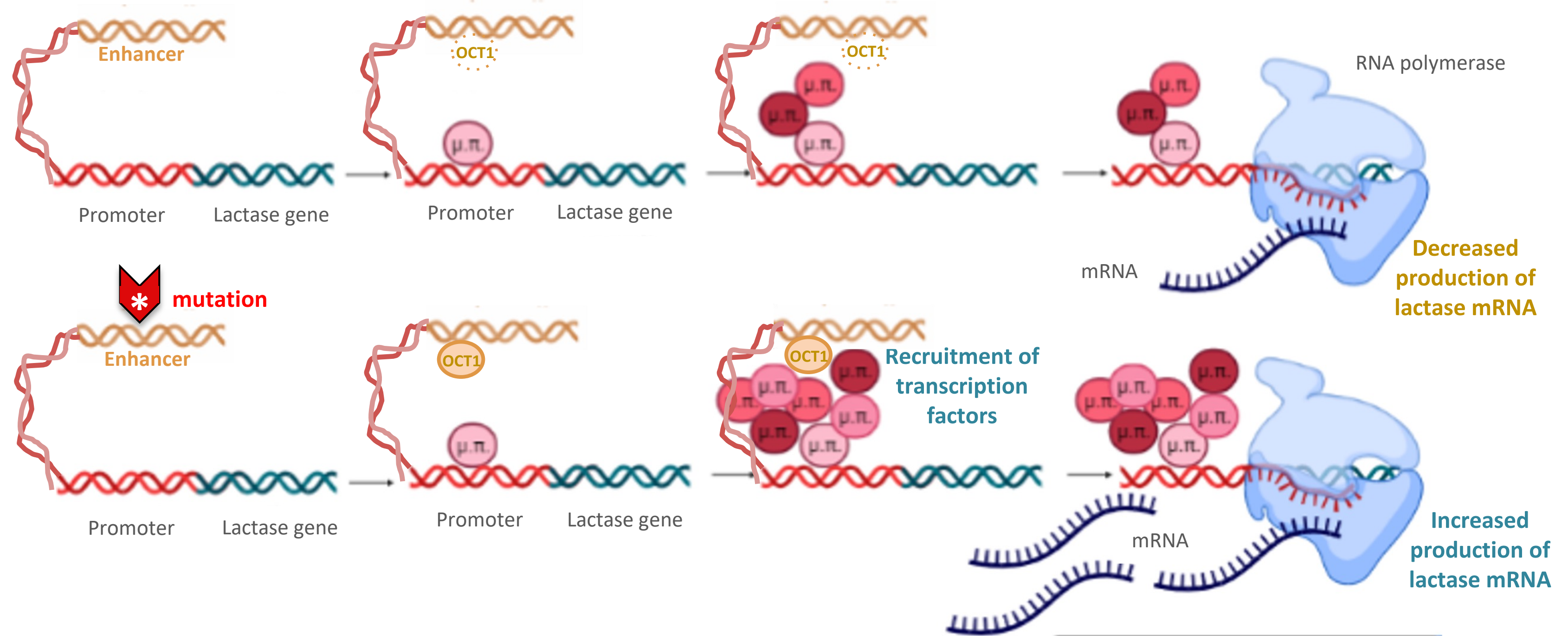
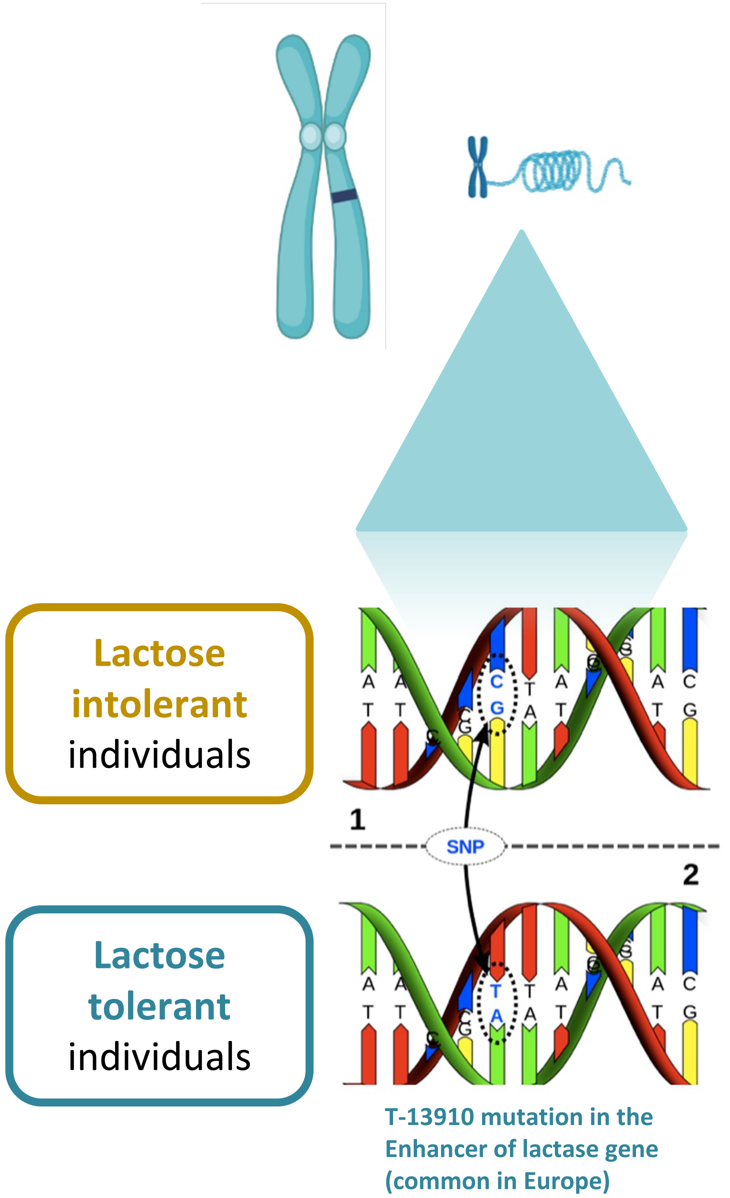
Lactase biosynthesis







Lactose intolerant adults

1 After weaning, there is a decrease in effectiveness of the transcription factors controlling the lactase gene.

2 This decreased effectiveness of the transcription factors leads to reduced transcription of the lactase gene.

3 This results in **lower lactase levels** in intestinal cells and **inability to digest lactase** in milk.

Lactose tolerant adults

- * A **mutation** at an Enhancer site upstream of the lactase gene increases binding of an Activator called Oct1, which participates in the regulation of lactase gene transcription.
- 1 The Oct1 activator attracts more general transcription factors to the lactase gene throughout adult life.
- 2 This increased attraction of transcription factors prevents the decreased transcription of the lactase gene that would normally happen, and thus lactase production continues into adulthood.
- 3 This results in **steady lactose levels** in intestinal cells, and a **retained ability to digest lactose** in milk throughout adulthood.

Anthropology and Biogeography of lactose tolerance

https://evo-ed.org/lactase-persistence/biological-processes/

The Neolithic Revolution

The neolithic revolution describes a time period about 12.000-6.000 years ago, during which humans throughout the world began the transition from a hunter-gatherer lifestyle to a farmer-herder lifestyle.

https://evo-ed.org/lactase-persistence/biological-processes/

The Neolithic Revolution

During this period, humans developed new ways of interacting with their environment, such as planting and harvesting tools, mills for grinding corn, and pottery for food storage.

Pastoralism and milking

One of the major innovations of the neolithic revolution was pastoralism and the the practice of milking livestock (such as goats, sheep, cows and camels). This practice was adopted by different civilizations in the period between 12.000 and 7.000 years ago.

https://evo-ed.org/lactase-persistence/biological-processes/

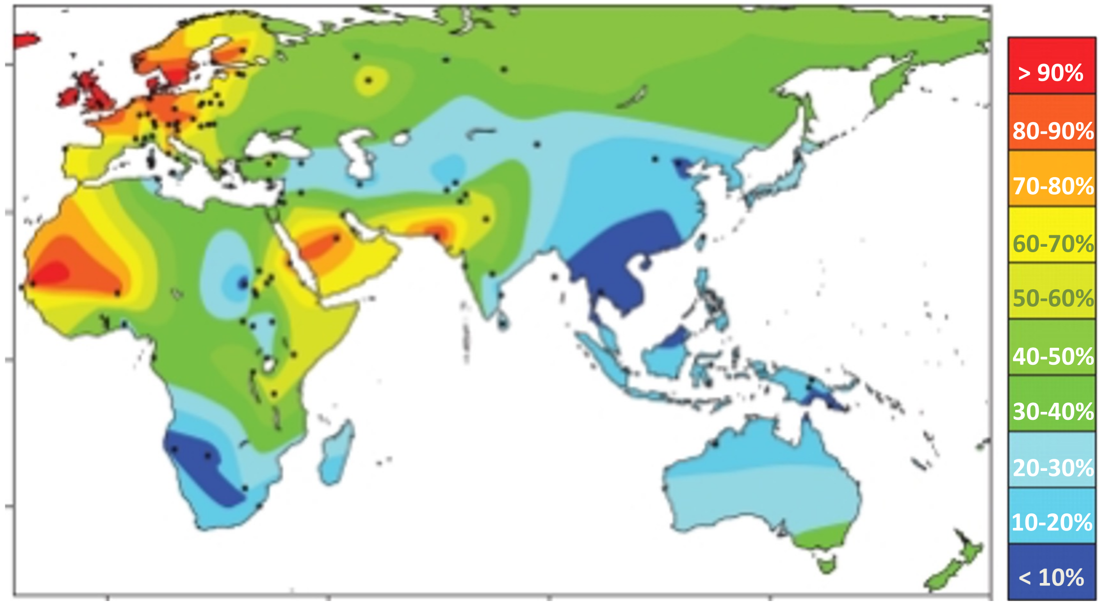
Milking livestock and milk consumption

- The Biocultural Coevolution theory suggests that livestock milking and lactase production coevolved.

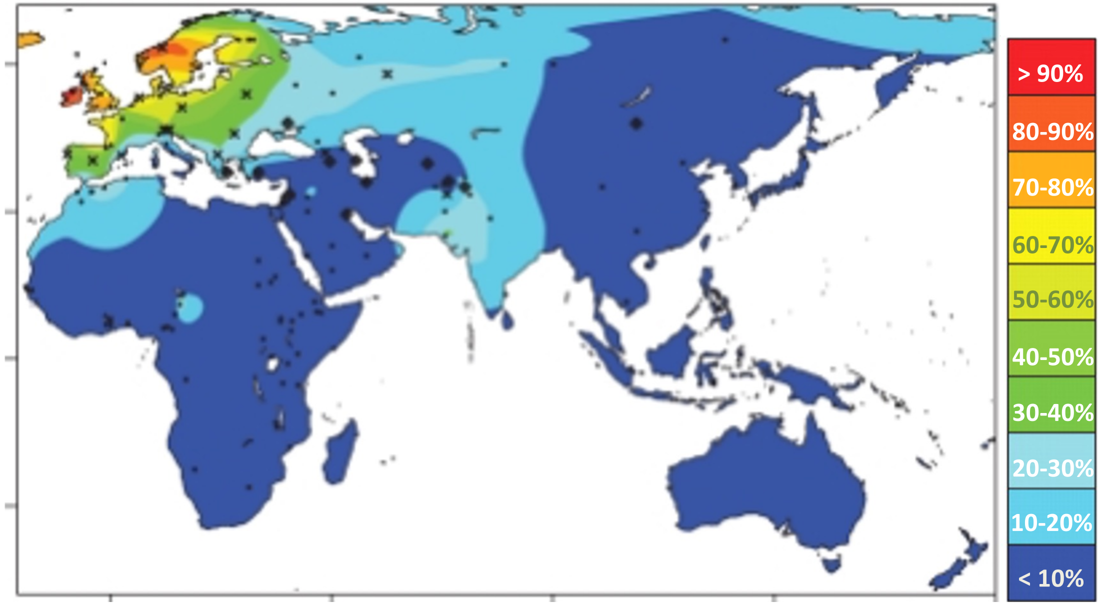
- This means that they arose around the same time, and their relationship is reciprocal: milk consumption led to the prevalence of lactose tolerance and, conversely, lactose tolerance enabled increased milk consumption.

How are human populations distributed today based on lactose tolerance?

Geographic distribution of lactose tolerance



Percentage of adults with lactose tolerance



Percentages of the T-13910 mutation upstream of the lactase gene

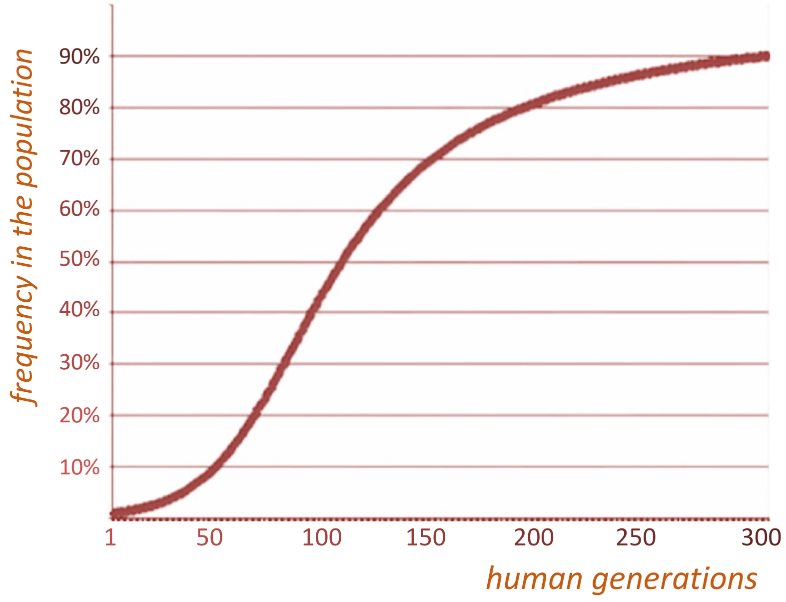
Figure source: Gerbault *et al.* (2011) *Phil. Trans. R. Soc. B* 366, 863-877.
Data sources: https://www.ucl.ac.uk/biosciences/gee/molecular-and-cultural-evolution-lab/global-lactase-persistence-association-database-glad; Ingram *et al.* (2022) Lactose malabsorption, in *Advanced Dairy Chemistry*, vol. 3, Lactose, water, salts and minor constituents, Springer, https://link.springer.com/book/10.1007/978-3-030-92585-7

*The T-13910 mutation (most common in Europe) enables **continuation of lactase production after weaning**. Other mutations (G-13915, G-13907, G-14009, C-14010) are connected to the same phenomenon in regions of Africa and Northwestern Asia (not shown here).*

What led to this geographic distribution?

- Mutations allowing lactose tolerance developed in human populations after the neolithic revolution (10.000 years ago), with different areas of the Earth showing different development rates.

- Depending on the area, and considering population migrations, the archaeological record and the modern picture, the selective advantage from retaining expression of effective lactase in adults is calculated at a rate of 0.5% to >3%.



Theoretical curve of the increase in frequency of a mutation allowing lactose tolerance in a population, assuming a selective advantage of 5%. Frequency may rise from 1% to 90% of the population within 8.000 years (that is, 300 human generations).

Two different stories

- Lactose tolerance and pastoralism arose and spread independently in Europe and Africa (an example of convergent evolution).
- Convergent evolution is the independent evolution of similar traits along separate evolutionary paths.
- In other words, we have two different stories explaining how pastoralism and lactose tolerance emerged in Europe and Africa, respectively.

Sources: Itan *et al., PLoS Comput. Biol.* (2009); Ranciano *et al., Am J. Hum. Genet.* (2014).

History of lactose tolerance in Europe

- The first evidence of pastoralism were discovered in the Middle East: the bones of juvenile bovines, killed before their first birthday, prove that humans in the area had began taming and milking cattle.

https://evo-ed.org/lactase-persistence/biological-processes/

- The bovine remains found in the Middle East were roughly 10.500 years old.

Migration (a)

The first tame bovines in Greece and the Balkans (8.000 years ago) were more closely related to Middle Eastern tame bovines than to wild European bovines…

This observation shows that the pastoralists who migrated to Europe from the Middle East brought their cattle with them.

Migration (b)

Middle Eastern pastoralists, being skilled and specialized food producers, spread rapidly, prevailing over the local hunter-gatherers they encountered in Central and Northern Europe.

The diffusion of migrating pastoralists from the Middle East in Europe led to the spread of mutations (mainly T-13910) allowing lactose tolerance.

History of lactose tolerance in Africa

In Africa, lactose tolerance evolved independently from the European origin. The mutations responsible for continuous lactose production are different.

Wild Type:	…AAGATAA**T**GTAG**C**CC**C**TG…	
Europe:	…AAGATAA**T**GTAG**T**CC**C**TG…	(T-13910)
Kenya:	…AAGATAA**G**GTAG**C**CC**C**TG…	(G-13915)
Sudan:	…AAGATAA**T**GTAG**C**CC**G**TG…	(G-13907)

Sources: Itan *et al., PLoS Comput. Biol.* (2009); Ranciano *et al., Am J. Hum. Genet.* (2014).

A gap of a few thousand years

- Anthropological studies date the emergence of pastoralism and the practice of livestock milking to **10,500-6,500 years ago**.
- However, genetic research shows that the trait of continuous lactase production, and thus lactose tolerance, only became widespread in Europe between **7,000-5,000 years ago**.
- This find means that, for several thousand years, many humans must have milked sheep, goats, cows, or camels without being able to absorb the milk they fed on.

How can we explain this paradox?

A theory explaining the paradox

It is possible that neolithic humans fermented milk to make cheese, a process that significantly reduced its lactose content, making it more digestible.

Pottery shards unearthed in Northern Europe likely served as sieves for the straining and fermentation of milk to make cheese.

Short history of cheese

Source: Benjamin Rabier (1926), **La vache qui rit** (La grande marque francaise)

- Many paleolithic and neolithic civilizations stored and transported food and water in animal skins and intestines.

- When milk was first introduced to the neolithic diet in the Middle East, it was probably stored in inflated cow stomachs, causing the curds to separate from the whey.

- Cheese is thought to have accompanied pastoralism ever since its cultural beginnings, and is still very popular among European cultures today.

Source: https://evo-ed.org/lactase-persistence/biological-processes/

Interpreting the spread of lactose tolerance

- The original theory for the prevalence of lactose tolerance included the processing of milk into forms that were **more digestible energy sources, like cheese**.

- Later studies attempted to interpret the coevolution of milk consumption and lactose tolerance based on the **potential dietary advantages** offered by milk consumption.

Interpreting the spread of lactose tolerance

Prevailing Theories

- ❑ Calcium and Vitamin D absorption
- ❑ Increase of the IGF-1 factor (Insulin-like Growth Factor 1) in circulation
- ❑ Increased fertility

Systematic comparison of lactose tolerant and intolerant individuals showed that:

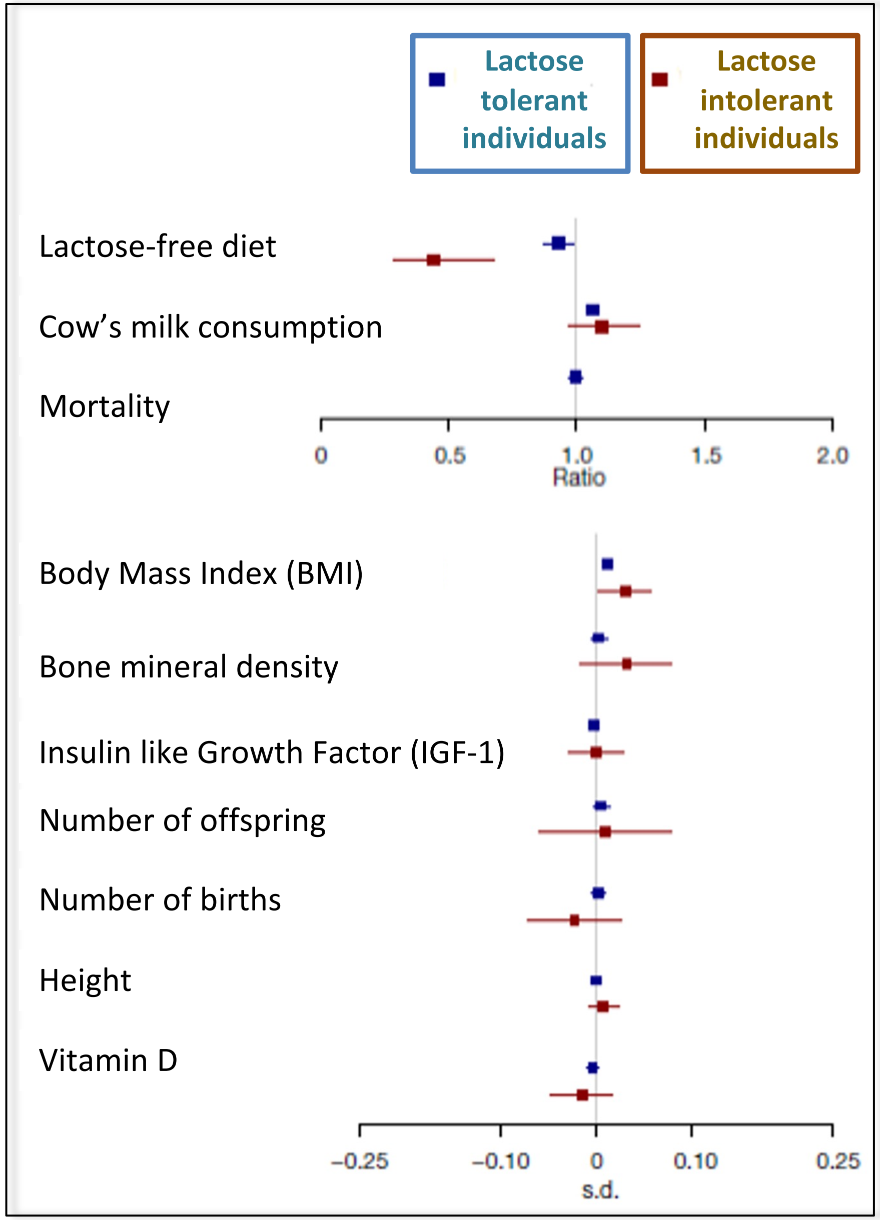
- ❑ No significant difference in vitamin D concentration or bone density
- ❑ No significant difference in Body Mass Index (BMI) or age of sexual maturity
- ❑ No significant impact on the number of births or offspring

Evershed *et al.* (2022) *Nature* 608, 336-345

CONCLUSION:

Coevolution of milk consumption and lactose tolerance and the spread of the lactose tolerance trait is not related to the dietary advantages that led to further milk consumption.

Effect of lactose tolerance alleles on different phenotypic traits:



Evershed *et al.* (2022) *Nature* 608, 336-345

Interpreting the spread of lactose tolerance

Alternative (more recent) interpretations:

Crisis Mechanism

- In times of hunger, the lack of food would turn people towards other sources of nutrition, such as milk consumption.
- The symptoms of lactose intolerance (e.g. diarrhea) would be more intense in malnutritioned individuals who had a hard time obtaining other food due to the shortages.
- Lactose intolerant individuals would not survive.
- Lactose tolerant individuals would have a survival advantage (positive selection of the lactose tolerance trait).

Chronic Mechanism

- Increased pathogen load, mostly zoonoses, connected to pastoralism and rising population density and mobility.
- Pathogen-induced mortality in lactose intolerant individuals that would consume dairy products, due to dehydration and intestinal disorders.
- Survival of lactose tolerant individuals, since they did not experience any symptoms from milk consumption (positive selection of the lactose tolerance trait).

Evershed *et al.* (2022) *Nature* 608, 336-345

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