### Fermentation Biotechnology

Fermentation is a [metabolic](https://en.wikipedia.org/wiki/Metabolism) process that converts [**sugar**](https://en.wikipedia.org/wiki/Sugar) **to acids**, **gases** or [**alcohol**](https://en.wikipedia.org/wiki/Alcohol). It occurs in [yeast](https://en.wikipedia.org/wiki/Yeast) and [bacteria,](https://en.wikipedia.org/wiki/Bacteria) and also in oxygen-starved muscle cells, as in the case of [lactic acid](https://en.wikipedia.org/wiki/Lactic_acid_fermentation) [fermentation.](https://en.wikipedia.org/wiki/Lactic_acid_fermentation) Fermentation is also used more broadly to refer to the **bulk growth** of [microorganisms](https://en.wikipedia.org/wiki/Microorganism) on a [growth medium](https://en.wikipedia.org/wiki/Growth_medium), often with the goal of producing a specific **chemical product** like enzyme, vaccines, antibiotics, food product/additive etc. French microbiologist [**Louis Pasteur**](https://en.wikipedia.org/wiki/Louis_Pasteur) is often remembered for his insights into fermentation and its microbial causes. The science of fermentation is known as [**zymology**](https://en.wikipedia.org/wiki/Zymology).

Fermentation takes place in the lack of oxygen (when the [electron transport chain](https://en.wikipedia.org/wiki/Electron_transport_chain) is unusable) and becomes the cell’s primary means of [ATP](https://en.wikipedia.org/wiki/Adenosine_triphosphate) (energy) production. It turns [NADH](https://en.wikipedia.org/wiki/NADH) and [pyruvate](https://en.wikipedia.org/wiki/Pyruvate) produced in the [glycolysis](https://en.wikipedia.org/wiki/Glycolysis) step into [NAD+](https://en.wikipedia.org/wiki/Nicotinamide_adenine_dinucleotide) and various small molecules depending on the type of fermentation. In the presence of O2, NADH and pyruvate are used to generate ATP in [respiration.](https://en.wikipedia.org/wiki/Cellular_respiration) This is called [oxidative phosphorylation](https://en.wikipedia.org/wiki/Oxidative_phosphorylation), and it generates much more ATP than glycolysis alone. For that reason, cells generally benefit from avoiding fermentation when oxygen is available, the exception being [obligate anaerobes](https://en.wikipedia.org/wiki/Obligate_anaerobe) which cannot tolerate oxygen.

The first step, glycolysis, is common to all fermentation pathways:

C6H12O6 + 2 NAD+ + 2 ADP + 2 Pi → 2 CH3COCOO− + 2 NADH + 2 ATP + 2 H2O + 2H+

[Pyruvate](https://en.wikipedia.org/wiki/Pyruvate) is CH3COCOO−. Pi is [inorganic phosphate](https://en.wikipedia.org/wiki/Inorganic_phosphate). Two [ADP](https://en.wikipedia.org/wiki/Adenosine_diphosphate) molecules and two Pi are converted to two [ATP](https://en.wikipedia.org/wiki/Adenosine_triphosphate) and two water molecules via [substrate-level phosphorylation.](https://en.wikipedia.org/wiki/Substrate-level_phosphorylation) Two molecules of [NAD+](https://en.wikipedia.org/wiki/Nicotinamide_adenine_dinucleotide) are also reduced to [NADH.](https://en.wikipedia.org/wiki/Nicotinamide_adenine_dinucleotide) In oxidative phosphorylation the energy for ATP formation is derived from an [electrochemical proton gradient](https://en.wikipedia.org/wiki/Electrochemical_gradient) generated across the [inner](https://en.wikipedia.org/wiki/Inner_mitochondrial_membrane) [mitochondrial membrane](https://en.wikipedia.org/wiki/Inner_mitochondrial_membrane) (or, in the case of bacteria, the [plasma membrane](https://en.wikipedia.org/wiki/Cell_membrane)) via the electron transport chain. Glycolysis has substrate-level phosphorylation (ATP generated directly at the point of reaction).

Humans have used fermentation to produce food and beverages since the [Neolithic age](https://en.wikipedia.org/wiki/Neolithic_age). For example, fermentation is used for preservation in a process that produces [lactic acid](https://en.wikipedia.org/wiki/Lactic_acid) as found in such sour [foods](https://en.wikipedia.org/wiki/Food) as [pickled cucumbers,](https://en.wikipedia.org/wiki/Pickled_cucumber) [kimchi](https://en.wikipedia.org/wiki/Kimchi) and [yogurt,](https://en.wikipedia.org/wiki/Yogurt) as well as for producing alcoholic

beverages such as [wine](https://en.wikipedia.org/wiki/Wine) and [beer.](https://en.wikipedia.org/wiki/Beer) Fermentation can even occur within the stomachs of animals, such as humans.

### Definitions of Fermentation

To many people, fermentation simply means the production of alcohol: grains and fruits are fermented to produce beer and wine. If a food soured, one might say it was 'off' or fermented. Here are some definitions of fermentation. They range from informal, general usage to more scientific definitions.

1. Preservation methods for food via [microorganisms](https://en.wikipedia.org/wiki/Microorganisms) (general use).
2. Any process that produces alcoholic beverages or acidic dairy products (general use).
3. Any large-scale microbial process occurring with or without air (common definition used in industry).
4. Any energy-releasing metabolic process that takes place only under anaerobic conditions (becoming more scientific).
5. Any metabolic process that releases energy from a sugar or other organic molecules, does not require oxygen or an electron transport system, and uses an organic molecule as the final electron acceptor (most scientific).

### Examples of Fermentation

Fermentation does not necessarily have to be carried out in an [anaerobic](https://en.wiktionary.org/wiki/anaerobic) environment. For example, even in the presence of abundant oxygen, [yeast](https://en.wikipedia.org/wiki/Yeast) cells greatly prefer fermentation to [aerobic respiration,](https://en.wikipedia.org/wiki/Aerobic_respiration) as long as [sugars](https://en.wikipedia.org/wiki/Sugar#Chemistry) are readily available for consumption (a phenomenon known as the [Crabtree effect](https://en.wikipedia.org/wiki/Crabtree_effect)). The antibiotic activity of [hops](https://en.wikipedia.org/wiki/Hops) also inhibits aerobic metabolism in yeast. Fermentation react [NADH](https://en.wikipedia.org/wiki/NADH) with an [endogenous](https://en.wikipedia.org/wiki/Endogeny), [organic](https://en.wikipedia.org/wiki/Organic_compound) [electron acceptor](https://en.wikipedia.org/wiki/Electron_acceptor). Usually this is pyruvate formed from the sugar during the glycolysis step. During fermentation, [pyruvate](https://en.wikipedia.org/wiki/Pyruvate) is metabolized to various compounds through several processes:

1. [**Ethanol fermentation**](https://en.wikipedia.org/wiki/Ethanol_fermentation), aka alcoholic fermentation, is the production of [ethanol](https://en.wikipedia.org/wiki/Ethanol) and [carbon dioxide](https://en.wikipedia.org/wiki/Carbon_dioxide)
2. [**Lactic acid fermentation**](https://en.wikipedia.org/wiki/Lactic_acid_fermentation) refers to two means of producing [lactic acid](https://en.wikipedia.org/wiki/Lactic_acid):
   * **Homolactic fermentation** is the production of lactic acid exclusively
   * **Heterolactic fermentation** is the production of lactic acid as well as other acids and alcohols.

Sugars are the most common [substrate](https://en.wikipedia.org/wiki/Substrate_%28biochemistry%29) of fermentation, and typical examples of fermentation products are [ethanol,](https://en.wikipedia.org/wiki/Ethanol) [lactic acid,](https://en.wikipedia.org/wiki/Lactic_acid) [carbon dioxide](https://en.wikipedia.org/wiki/Carbon_dioxide), and [hydrogen gas (H2).](https://en.wikipedia.org/wiki/Hydrogen) However, more exotic compounds can be produced by fermentation, such as [butyric acid](https://en.wikipedia.org/wiki/Butyric_acid) and [acetone](https://en.wikipedia.org/wiki/Acetone). [Yeast](https://en.wikipedia.org/wiki/Yeast) carries out [fermentation](https://en.wikipedia.org/wiki/Fermentation_%28food%29) in the production of [ethanol](https://en.wikipedia.org/wiki/Ethanol) in [beers](https://en.wikipedia.org/wiki/Beer), [wines](https://en.wikipedia.org/wiki/Wine), and other alcoholic drinks, along with the production of large quantities of [carbon dioxide.](https://en.wikipedia.org/wiki/Carbon_dioxide) Fermentation occurs in [mammalian](https://en.wikipedia.org/wiki/Mammal) [muscle](https://en.wikipedia.org/wiki/Muscle) during periods of intense exercise where oxygen supply becomes limited, resulting in the creation of [lactic acid.](https://en.wikipedia.org/wiki/Lactic_acid)

#### Ethanol fermentation

The [chemical equation](https://en.wikipedia.org/wiki/Chemical_equation) below shows the alcoholic fermentation of [glucose,](https://en.wikipedia.org/wiki/Glucose) whose [chemical](https://en.wikipedia.org/wiki/Chemical_formula) [formula](https://en.wikipedia.org/wiki/Chemical_formula) is C6H12O6. One glucose molecule is converted into two [ethanol](https://en.wikipedia.org/wiki/Ethanol) molecules and two [carbon dioxide](https://en.wikipedia.org/wiki/Carbon_dioxide) molecules:

C6H12O6 → 2 C2H5OH + 2 CO2

C2H5OH is the [chemical formula](https://en.wikipedia.org/wiki/Chemical_formula) for [ethanol](https://en.wikipedia.org/wiki/Ethanol).

Before fermentation takes place, one [glucose](https://en.wikipedia.org/wiki/Glucose) molecule is broken down into two [pyruvate](https://en.wikipedia.org/wiki/Pyruvate) molecules. This is known as [glycolysis.](https://en.wikipedia.org/wiki/Glycolysis)

#### Lactic acid fermentation

**Homolactic fermentation** (producing only lactic acid) is the simplest type of fermentation. The pyruvate from glycolysis undergoes a simple [redox](https://en.wikipedia.org/wiki/Redox) reaction, forming [lactic acid.](https://en.wikipedia.org/wiki/Lactic_acid) It is unique because it is one of the only respiration processes to not produce a gas as a byproduct. Overall, one molecule of glucose (or any six-carbon sugar) is converted to two molecules of lactic acid: C6H12O6→2CH3CHOHCOOH

It occurs in the muscles of animals when they need energy faster than the [blood](https://en.wikipedia.org/wiki/Blood) can supply oxygen. It also occurs in some kinds of [bacteria](https://en.wikipedia.org/wiki/Bacterium) (such as [*Lactobacilli*](https://en.wikipedia.org/wiki/Lactobacilli)) and some [fungi](https://en.wikipedia.org/wiki/Fungi). It is this type of bacteria that converts [lactose](https://en.wikipedia.org/wiki/Lactose) into lactic acid in [yogurt](https://en.wikipedia.org/wiki/Yogurt), giving it its sour taste. These lactic acid bacteria can carry out either homolactic fermentation, where the end-product is mostly lactic acid, or

**Heterolactic fermentation**, where some lactate is further metabolized and results in ethanol and carbon dioxide (via the [phosphoketolase](https://en.wikipedia.org/wiki/Phosphoketolase) pathway), acetate, or other metabolic products, e.g.: C6H12O6→CH3CHOHCOOH+C2H5OH+CO2

If lactose is fermented (as in yogurts and cheeses), it is first converted into glucose and galactose (both six-carbon sugars with the same atomic formula): C12H22O11 + H2O → 2 C6H12O6 Heterolactic fermentation is in a sense intermediate between lactic acid fermentation, and other types, e.g. alcoholic fermentation. The reasons to go further and convert lactic acid into anything else are:

* The acidity of lactic acid impedes biological processes; this can be beneficial to the fermenting organism as it drives out competitors who are unadapted to the acidity; as a result the food will have a longer shelf-life (part of the reason foods are purposely fermented in the first place); however, beyond a certain point, the acidity starts affecting the organism that produces it.
* The high concentration of lactic acid (the final product of fermentation) drives the equilibrium backwards ([Le Chatelier's principle](https://en.wikipedia.org/wiki/Le_Chatelier%27s_principle)), decreasing the rate at which fermentation can occur, and slowing down growth
* Ethanol, that lactic acid can be easily converted to, is volatile and will readily escape, allowing the reaction to proceed easily. [CO2](https://en.wikipedia.org/wiki/Carbon_dioxide) is also produced, however it's only weakly acidic, and even more volatile than ethanol.
* Acetic acid (another conversion product) is acidic, and not as volatile as ethanol; however, in the presence of limited oxygen, its creation from lactic acid releases a lot of additional energy. It is a lighter molecule than lactic acid, that forms fewer hydrogen bonds with its surroundings (due to having fewer groups that can form such bonds), and thus more volatile and will also allow the reaction to move forward more quickly.
* If [propionic acid,](https://en.wikipedia.org/wiki/Propionic_acid) [butyric acid](https://en.wikipedia.org/wiki/Butyric_acid) and longer monocarboxylic acids are produced (see [mixed](https://en.wikipedia.org/wiki/Mixed_acid_fermentation) [acid fermentation](https://en.wikipedia.org/wiki/Mixed_acid_fermentation)), the amount of acidity produced per glucose consumed will decrease, as with ethanol, allowing faster growth.

### Aerobic respiration

In [aerobic respiration,](https://en.wikipedia.org/wiki/Aerobic_respiration) the pyruvate produced by glycolysis is oxidized completely, generating additional ATP and NADH in the [citric acid cycle](https://en.wikipedia.org/wiki/Citric_acid_cycle) and by [oxidative phosphorylation.](https://en.wikipedia.org/wiki/Oxidative_phosphorylation) However, this can occur only in the presence of oxygen. Oxygen is toxic to organisms that are [obligate](https://en.wikipedia.org/wiki/Obligate_anaerobe) [anaerobes,](https://en.wikipedia.org/wiki/Obligate_anaerobe) and is not required by [facultative anaerobic organisms](https://en.wikipedia.org/wiki/Facultative_anaerobic_organism). In the absence of oxygen, one of the fermentation pathways occurs in order to regenerate [NAD+](https://en.wikipedia.org/wiki/NAD%2B); lactic acid fermentation is one of these pathways.

### Hydrogen gas production in fermentation

[Hydrogen](https://en.wikipedia.org/wiki/Hydrogen) gas is produced in many types of fermentation ([mixed acid fermentation](https://en.wikipedia.org/wiki/Mixed_acid_fermentation), [butyric acid](https://en.wikipedia.org/wiki/Butyric_acid) fermentation, [caproate](https://en.wikipedia.org/wiki/Caproate) fermentation, [butanol](https://en.wikipedia.org/wiki/Butanol) fermentation, [glyoxylate](https://en.wikipedia.org/wiki/Glyoxylate) fermentation), as a way to regenerate NAD+ from NADH. [Electrons](https://en.wikipedia.org/wiki/Electron) are transferred to [ferredoxin](https://en.wikipedia.org/wiki/Ferredoxin), which in turn is oxidized by [hydrogenase,](https://en.wikipedia.org/wiki/Hydrogenase) producing H2. Hydrogen gas is a [substrate](https://en.wikipedia.org/wiki/Substrate_%28biochemistry%29) for [methanogens](https://en.wikipedia.org/wiki/Methanogen) and [sulfate](https://en.wikipedia.org/wiki/Sulfate-reducing_bacteria) [reducers,](https://en.wikipedia.org/wiki/Sulfate-reducing_bacteria) which keep the concentration of hydrogen low and favor the production of such an energy-rich compound, but hydrogen gas at a fairly high concentration can nevertheless be formed, as in [flatus.](https://en.wikipedia.org/wiki/Flatus)

As an example of mixed acid fermentation, bacteria such as [*Clostridium pasteurianum*](https://en.wikipedia.org/wiki/Clostridium_pasteurianum) ferment glucose producing [butyrate](https://en.wikipedia.org/wiki/Butyric_acid), [acetate](https://en.wikipedia.org/wiki/Acetic_acid), [carbon dioxide](https://en.wikipedia.org/wiki/Carbon_dioxide) and hydrogen gas: The reaction leading to acetate is:

C6H12O6 + 4 H2O → 2 CH3COO− + 2 HCO3− + 4 H+ + 4 H2

Glucose could theoretically be converted into just CO2 and H2, but the global reaction releases little energy.

### Methane gas production in fermentation

[Acetic acid](https://en.wikipedia.org/wiki/Acetic_acid) can also undergo a [dismutation](https://en.wikipedia.org/wiki/Dismutation) reaction to produce [methane](https://en.wikipedia.org/wiki/Methane) and [carbon dioxide](https://en.wikipedia.org/wiki/Carbon_dioxide): CH3COO− + H+ → CH4 + CO2 ΔG° = -36 kJ/reaction

This [disproportionation](https://en.wikipedia.org/wiki/Disproportionation) reaction is catalysed by [methanogen](https://en.wikipedia.org/wiki/Methanogen) [archaea](https://en.wikipedia.org/wiki/Archaea) in their fermentative metabolism. One electron is transferred from the [carbonyl](https://en.wikipedia.org/wiki/Carbonyl) function ([e− donor](https://en.wikipedia.org/wiki/Electron_donor)) of the [carboxylic](https://en.wikipedia.org/wiki/Carboxylic)

group to the [methyl](https://en.wikipedia.org/wiki/Methyl) group ([e− acceptor](https://en.wikipedia.org/wiki/Electron_acceptor)) of acetic acid to respectively produce CO2 and methane gas.

### History of Fermentation

The use of [fermentation](https://en.wikipedia.org/wiki/Fermentation_in_food_processing), particularly for [beverages,](https://en.wikipedia.org/wiki/Alcoholic_beverage) has existed since the [Neolithic](https://en.wikipedia.org/wiki/Neolithic) and has been documented dating from 7000–6600 BCE in [Jiahu](https://en.wikipedia.org/wiki/Jiahu), [China](https://en.wikipedia.org/wiki/Neolithic_China), 6000 BCE in Georgia, 3150 BCE in [ancient Egypt,](https://en.wikipedia.org/wiki/Ancient_Egypt) 3000 BCE in [Babylon,](https://en.wikipedia.org/wiki/Babylon) 2000 BCE in pre-Hispanic Mexico, and 1500 BC in [Sudan](https://en.wikipedia.org/wiki/Sudan). Fermented foods have a religious significance in [Judaism](https://en.wikipedia.org/wiki/Chametz) and [Christianity](https://en.wikipedia.org/wiki/Christianity_and_alcohol). The [Baltic](https://en.wikipedia.org/wiki/Baltic_mythology) [god](https://en.wikipedia.org/wiki/Baltic_mythology) Rugutis was worshiped as the agent of fermentation.

[**Louis Pasteur**](https://en.wikipedia.org/wiki/Louis_Pasteur) (1822–1895), during the 1850s and 1860s, showed that fermentation is initiated by living organisms in a series of investigations. In 1857, Pasteur showed that lactic acid fermentation is caused by living organisms. In 1860, he demonstrated that bacteria cause [souring](https://en.wikipedia.org/wiki/Souring) in milk, a process formerly thought to be merely a chemical change, and his work in identifying the role of microorganisms in food spoilage led to the process of [pasteurization](https://en.wikipedia.org/wiki/Pasteurization). In 1877, working to improve the French [brewing industry,](https://en.wikipedia.org/wiki/Brewing_industry) Pasteur published his famous paper on fermentation, "*Etudes sur la Bière*", which was translated into English in 1879 as "Studies on fermentation". He defined fermentation (incorrectly) as "Life without air", but correctly showed that specific types of microorganisms cause specific types of fermentations and specific end- products.

Although showing fermentation to be the result of the action of living microorganisms was a breakthrough, it did not explain the basic nature of the fermentation process, or prove that it is caused by the microorganisms that appear to be always present. Many scientists, including Pasteur, had unsuccessfully attempted to extract the fermentation [enzyme](https://en.wikipedia.org/wiki/Enzyme) from [yeast](https://en.wikipedia.org/wiki/Yeast)[.[26]](https://en.wikipedia.org/wiki/Fermentation#cite_note-Modern_History_Sourcebook_1895-26) Success came in 1897 when the German chemist [**Eduard Buechner**](https://en.wikipedia.org/wiki/Eduard_Buechner) ground up yeast, extracted a juice from them, then found to his amazement that this "dead" liquid would ferment a sugar solution, forming carbon dioxide and alcohol much like living yeasts. Buechner's results are considered to mark the birth of biochemistry. The "unorganized ferments" behaved just like the organized ones. From that time on, the term enzyme came to be applied to all ferments. It was then understood that fermentation is caused by enzymes that are produced by microorganisms. In 1907, Buechner won the [Nobel Prize in chemistry](https://en.wikipedia.org/wiki/Nobel_Prize_in_chemistry) for his work.

Advances in microbiology and fermentation technology have continued steadily up until the present. For example, in the late 1970s, it was discovered that microorganisms could be [mutated](https://en.wikipedia.org/wiki/Mutation) with physical and chemical treatments to be higher-yielding, faster-growing, tolerant of less oxygen, and able to use a more concentrated medium. Strain [selection](https://en.wikipedia.org/wiki/Selection_%28biology%29) and [hybridization](https://en.wikipedia.org/wiki/Hybrid_%28biology%29) developed as well, affecting most modern [food fermentations](https://en.wikipedia.org/wiki/Fermentation_%28food%29). Other approach to advancing the fermentation industry has been done by companies such as [BioTork,](https://en.wikipedia.org/wiki/BioTork) a [biotechnology](https://en.wikipedia.org/wiki/Biotechnology) company that naturally [evolves](https://en.wikipedia.org/wiki/Evolves) microorganisms to improve fermentation processes. This approach differs from the more popular [genetic modification](https://en.wikipedia.org/wiki/Genetic_modification), which has become the current industry standard.

### Industrial fermentation

Industrial fermentation is **the intentional use of** [**fermentation**](https://en.wikipedia.org/wiki/Fermentation_%28biochemistry%29) **by** [**microorganisms**](https://en.wikipedia.org/wiki/Microorganism)such as [bacteria](https://en.wikipedia.org/wiki/Bacteria) and [fungi](https://en.wikipedia.org/wiki/Fungi) to make products useful to humans. Fermented products have applications as [**food**](https://en.wikipedia.org/wiki/Food) as well as in **general** [**industry**](https://en.wikipedia.org/wiki/Industry). Some commodity chemicals, such as [acetic acid,](https://en.wikipedia.org/wiki/Acetic_acid) [citric acid,](https://en.wikipedia.org/wiki/Citric_acid) and [ethanol](https://en.wikipedia.org/wiki/Ethanol) are made by fermentation. The rate of fermentation depends on the concentration of microorganisms, cells, cellular components, and enzymes as well as temperature, pH and for [aerobic](https://en.wikipedia.org/wiki/Aerobic_respiration) fermentation oxygen. Product recovery frequently involves the concentration of the dilute solution. Nearly all commercially produced enzymes, such as [lipase,](https://en.wikipedia.org/wiki/Lipase) [invertase](https://en.wikipedia.org/wiki/Invertase) and [rennet,](https://en.wikipedia.org/wiki/Rennet) are made by fermentation with [genetically modified microbes](https://en.wikipedia.org/wiki/Genetic_engineering). In some cases, production of biomass itself is the objective, as in the case of [baker's yeast](https://en.wikipedia.org/wiki/Baker%27s_yeast) and lactic acid bacteria [starter](https://en.wikipedia.org/wiki/Starter_cultures) [cultures](https://en.wikipedia.org/wiki/Starter_cultures) for cheese making. In general, fermentations can be divided into four types:

* Production of biomass (viable cellular material)
* Production of extracellular metabolites (chemical compounds)
* Production of intracellular components (enzymes and other proteins)
* Transformation of substrate (in which the transformed substrate is itself the product)

These types are not necessarily disjoint from each other, but provide a framework for understanding the differences in approach. The organisms used may be bacteria, yeasts, molds, animal cells, or plant cells. Special considerations are required for the specific organisms used in the fermentation, such as the dissolved [oxygen](https://en.wikipedia.org/wiki/Oxygen) level, nutrient levels, and [temperature](https://en.wikipedia.org/wiki/Temperature).

### General overview of Industrial fermentation

In most industrial fermentations, the organisms are submerged in a liquid medium; in others, such as the fermentation of [cocoa beans,](https://en.wikipedia.org/wiki/Cocoa_bean) [coffee](https://en.wikipedia.org/wiki/Coffee) cherries, and [miso](https://en.wikipedia.org/wiki/Miso), fermentation takes place on the moist surface of the medium. There are also industrial considerations related to the fermentation process. For instance, to avoid biological process contamination, the fermentation medium, air, and equipment are sterilized. Foam control can be achieved by either mechanical foam destruction or chemical anti-foaming agents. Several other factors must be measured and controlled such as pressure, temperature, agitator shaft power, and viscosity. An important element for industrial fermentations is scale up. This is the conversion of a laboratory procedure to an industrial process. It is well established in the field of industrial microbiology that what works well at the laboratory scale may work poorly or not at all when first attempted at large

scale. It is generally not possible to take fermentation conditions that have worked in the laboratory and blindly apply them to industrial-scale equipment. Although many parameters have been tested for use as scale up criteria, there is no general formula because of the variation in fermentation processes. The most important methods are the maintenance of constant power consumption per unit of broth and the maintenance of constant volumetric transfer rate.

#### Phases of microbial growth

When a particular organism is introduced into a selected growth medium, the medium is inoculated with the particular organism. Growth of the inoculum does not occur immediately, but takes a little while. This is the period of adaptation, called the lag phase. Following the lag phase, the rate of growth of the organism steadily increases, for a certain period—this period is the log or exponential phase. After a certain time of exponential phase, the rate of growth slows down, due to the continuously falling concentrations of nutrients and/or continuously increasing (accumulating) concentrations of toxic substances. This phase, where the increase of the rate of growth is checked, is the deceleration phase. After the deceleration phase, growth ceases and the culture enters a stationary phase or a steady state. The biomass remains constant, except when certain accumulated chemicals in the culture lyse the cells (chemolysis). Unless other micro- organisms contaminate the culture, the chemical constitution remains unchanged. If all of the nutrients in the medium are consumed, or if the concentration of toxins is too great, the cells may become [scenescent](https://en.wikipedia.org/wiki/Senescence) and begin to die off. The total amount of biomass may not decrease, but the number of viable organisms will decrease.

#### Fermentation Medium

The microbes used for fermentation grow in (or on) specially designed [growth medium](https://en.wikipedia.org/wiki/Growth_medium) which supplies the nutrients required by the organisms. A variety of media exists, but invariably contains a carbon source, a nitrogen source, water, salts, and micronutrients. In the production of wine, the medium is grape must. In the production of bio-ethanol, the medium may consist mostly of whatever inexpensive carbon source is available.

Carbon sources are typically sugars or other carbohydrates, although in the case of substrate transformations (such as the production of vinegar) the carbon source may be an alcohol or

something else altogether. For large scale fermentations, such as those used for the production of ethanol, inexpensive sources of carbohydrates, such as [molasses,](https://en.wikipedia.org/wiki/Molasses) [corn steep liquor](https://en.wikipedia.org/wiki/Corn_steep_liquor), sugar cane juice, or sugar beet juice are used to minimize costs. More sensitive fermentations may instead use purified [glucose,](https://en.wikipedia.org/wiki/Glucose) [sucrose](https://en.wikipedia.org/wiki/Sucrose), [glycerol](https://en.wikipedia.org/wiki/Glycerol) or other sugars, which reduces variation and helps ensure the purity of the final product. Organisms meant to produce enzymes such as [beta galactosidase,](https://en.wikipedia.org/wiki/Beta_galactosidase) [invertase](https://en.wikipedia.org/wiki/Invertase) or other amylases may be fed starch to select for organisms that express the enzymes in large quantity.

[Fixed nitrogen](https://en.wikipedia.org/wiki/Nitrogen_fixation) sources are required for most organisms to synthesize [proteins](https://en.wikipedia.org/wiki/Proteins), [nucleic acids](https://en.wikipedia.org/wiki/Nucleic_acid) and other cellular components. Depending on the enzyme capabilities of the organism, nitrogen may be provided as bulk protein, such as soy meal; as pre-digested polypeptides, such as [peptone](https://en.wikipedia.org/wiki/Peptone) or [tryptone](https://en.wikipedia.org/wiki/Tryptone); or as ammonia or nitrate salts. Cost is also an important factor in the choice of a nitrogen source. Phosphorus is needed for production of [phospholipids](https://en.wikipedia.org/wiki/Phospholipids) in cellular membranes and for the production of [nucleic acids.](https://en.wikipedia.org/wiki/Nucleic_acid) The amount of phosphate which must be added depends upon the composition of the broth and the needs of the organism, as well as the objective of the fermentation. For instance, some cultures will not produce secondary metabolites in the presence of phosphate.

Growth factors and trace nutrients are included in the fermentation broth for organisms incapable of producing all of the vitamins they require. [Yeast extract](https://en.wikipedia.org/wiki/Yeast_extract) is a common source of micronutrients and vitamins for fermentation media. Inorganic nutrients, including [trace elements](https://en.wikipedia.org/wiki/Trace_element) such as iron, zinc, copper, manganese, molybdenum and cobalt are typically present in unrefined carbon and nitrogen sources, but may have to be added when purified carbon and nitrogen sources are used. Fermentations which produce large amounts of gas (or which require the addition of gas) will tend to form a layer of foam, since fermentation broth typically contains a variety of foam- reinforcing proteins, peptides or starches. To prevent this foam from occurring or accumulating, [antifoaming agents](https://en.wikipedia.org/wiki/Antifoaming_agent) may be added. Mineral buffering salts, such as carbonates and phosphates, may be used to stabilize pH near optimum. When metal ions are present in high concentrations, use of a [chelating agent](https://en.wikipedia.org/wiki/Chelator) may be necessary.

### Production of biomass

[Microbial](https://en.wikipedia.org/wiki/Microbial) [cells](https://en.wikipedia.org/wiki/Cell_%28biology%29) or [biomass](https://en.wikipedia.org/wiki/Biomass) is sometimes the intended product of fermentation. Examples include [single cell protein,](https://en.wikipedia.org/wiki/Single_cell_protein) [baker’s yeast,](https://en.wikipedia.org/wiki/Bakers_yeast) [*Lactobacillus*](https://en.wikipedia.org/wiki/Lactobacillus)*,* [*E. coli*](https://en.wikipedia.org/wiki/E._coli)*,* and others. In the case of single-cell protein, [algae](https://en.wikipedia.org/wiki/Algae) are grown in large open ponds which allow photosynthesis to occur. If the biomass is to be used for inoculation of other fermentations, care must be taken to prevent [mutations](https://en.wikipedia.org/wiki/Mutation) from occurring.

### Production of extracellular metabolites

Microbial [metabolites](https://en.wikipedia.org/wiki/Metabolite) can be divided into two groups: those produced during the growth phase of the organism, called **primary metabolites** and those produced during the stationary phase, called **secondary metabolites**. Some examples of primary metabolites are [ethanol,](https://en.wikipedia.org/wiki/Ethanol) [citric acid,](https://en.wikipedia.org/wiki/Citric_acid) [glutamic acid,](https://en.wikipedia.org/wiki/Glutamic_acid) [lysine,](https://en.wikipedia.org/wiki/Lysine) [vitamins](https://en.wikipedia.org/wiki/Vitamin) and [polysaccharides](https://en.wikipedia.org/wiki/Polysaccharide). Some examples of secondary metabolites are [penicillin](https://en.wikipedia.org/wiki/Penicillin), [cyclosporin A,](https://en.wikipedia.org/wiki/Cyclosporin_A) [gibberellin,](https://en.wikipedia.org/wiki/Gibberellin) and [lovastatin](https://en.wikipedia.org/wiki/Lovastatin).

#### Primary metabolites

[Primary metabolites](https://en.wikipedia.org/wiki/Primary_metabolite) are compounds made during the ordinary metabolism of the organism during the growth phase. A common example is ethanol or lactic acid, produced during [glycolysis.](https://en.wikipedia.org/wiki/Glycolysis) Citric acid is produced by some strains of [*Aspergillus niger*](https://en.wikipedia.org/wiki/Aspergillus_niger) as part of the [citric acid](https://en.wikipedia.org/wiki/Citric_acid_cycle) [cycle](https://en.wikipedia.org/wiki/Citric_acid_cycle) to acidify their environment and prevent competitors from taking over. Glutamate is produced by some [*Micrococcus*](https://en.wikipedia.org/wiki/Micrococcus)species, and some [*Corynebacterium*](https://en.wikipedia.org/wiki/Corynebacterium)species produce lysine, threonine, tryptophan and other amino acids. All of these compounds are produced during the normal "business" of the cell and released into the environment. There is therefore no need to rupture the cells for product recovery.

#### Secondary metabolites

[Secondary metabolites](https://en.wikipedia.org/wiki/Secondary_metabolite) are compounds made in the stationary phase; penicillin, for instance, prevents the growth of bacteria which could compete with [*Penicillium*](https://en.wikipedia.org/wiki/Penicillium)molds for resources. Some bacteria, such as [*Lactobacillus*](https://en.wikipedia.org/wiki/Lactobacillus) species, are able to produce [bacteriocins](https://en.wikipedia.org/wiki/Bacteriocin) which prevent the growth of bacterial competitors as well. These compounds are of obvious value to humans wishing to prevent the growth of bacteria, either as [antibiotics](https://en.wikipedia.org/wiki/Antibiotic) or as [antiseptics](https://en.wikipedia.org/wiki/Antiseptic) (such as

[gramicidin S](https://en.wikipedia.org/wiki/Gramicidin_S)). Fungicides, such as [griseofulvin](https://en.wikipedia.org/wiki/Griseofulvin) are also produced as secondary metabolites. Typically secondary metabolites are not produced in the presence of glucose or other carbon sources which would encourage growth, and like primary metabolites are released into the surrounding medium without rupture of the cell membrane.

### Production of intracellular components

Of primary interest among the intracellular components are microbial [enzymes](https://en.wikipedia.org/wiki/Enzyme): [catalase,](https://en.wikipedia.org/wiki/Catalase) [amylase,](https://en.wikipedia.org/wiki/Amylase) [protease,](https://en.wikipedia.org/wiki/Protease) [pectinase](https://en.wikipedia.org/wiki/Pectinase), [glucose isomerase](https://en.wikipedia.org/wiki/Glucose_isomerase), [cellulase](https://en.wikipedia.org/wiki/Cellulase), [hemicellulase](https://en.wikipedia.org/wiki/Hemicellulase), [lipase](https://en.wikipedia.org/wiki/Lipase), [lactase](https://en.wikipedia.org/wiki/Lactase), [streptokinase](https://en.wikipedia.org/wiki/Streptokinase) and many others. Recombinant proteins, such as [insulin](https://en.wikipedia.org/wiki/Insulin), [hepatitis B vaccine,](https://en.wikipedia.org/wiki/Hepatitis_B_vaccine) [interferon,](https://en.wikipedia.org/wiki/Interferon) [granulocyte colony-stimulating factor](https://en.wikipedia.org/wiki/Granulocyte_colony-stimulating_factor), [streptokinase](https://en.wikipedia.org/wiki/Streptokinase) and others are also made this way. The largest difference between this process and the others is that the cells must be ruptured (lysed) at the end of fermentation, and the environment must be manipulated to maximize the amount of the product. Furthermore, the product (typically a protein) must be separated from all of the other cellular proteins in the lysate to be purified.

### Transformation of substrate

Substrate transformation involves the transformation of a specific compound into another, such as in the case of [phenylacetylcarbinol,](https://en.wikipedia.org/wiki/Phenylacetylcarbinol) and [steroid](https://en.wikipedia.org/wiki/Steroid) [biotransformation](https://en.wikipedia.org/wiki/Biotransformation), or the transformation of a raw material into a finished product, in the case of food fermentations and sewage treatment.

#### Food fermentation

Ancient fermented food processes, such as making [bread](https://en.wikipedia.org/wiki/Bread), [wine,](https://en.wikipedia.org/wiki/Wine) [cheese,](https://en.wikipedia.org/wiki/Cheese) [curds](https://en.wikipedia.org/wiki/Curds), [idli](https://en.wikipedia.org/wiki/Idli), [dosa,](https://en.wikipedia.org/wiki/Dosa) etc., can be dated to more than [seven thousand years ago](https://en.wikipedia.org/wiki/5th_millennium_BC). They were developed long before man had any knowledge of the existence of the [microorganisms](https://en.wikipedia.org/wiki/Microorganism) involved. Some foods such as [Marmite](https://en.wikipedia.org/wiki/Marmite) are the byproduct of the fermentation process, in this case in the production of [beer.](https://en.wikipedia.org/wiki/Beer)

#### Ethanol fuel

Fermentation is the main source of ethanol in the production of [Ethanol fuel](https://en.wikipedia.org/wiki/Ethanol_fuel). Common crops such as sugar cane, potato, cassava and corn are fermented by yeast to produce ethanol which is further processed to become fuel.

#### Sewage treatment

In the process of sewage treatment, sewage is digested by enzymes secreted by bacteria. Solid organic matters are broken down into harmless, soluble substances and carbon dioxide. Liquids that result are disinfected to remove pathogens before being discharged into rivers or the sea or can be used as liquid fertilizers. Digested solids, known also as sludge, is dried and used as fertilizer. Gaseous byproducts such as methane can be utilized as [**biogas**](https://en.wikipedia.org/wiki/Biogas)to fuel electrical generators. One advantage of bacterial digestion is that it reduces the bulk and odor of sewage, thus reducing space needed for dumping. The main disadvantage of bacterial digestion in sewage disposal is that it is a very slow process.

#### Agricultural Feed

A wide variety of agroindustrial waste products can be fermented to use as food for animals, especially ruminants. Fungi have been employed to break down cellulosic wastes to increase protein content and improve *in vitro* digestibility.

## BIOASSAY

**Bioassay** (commonly used shorthand for **biological assay or assessment**), or **biological standardization** is a type of scientific [experiment](https://en.wikipedia.org/wiki/Experiment). A bioassay involves the use of live animal or plant (*in vivo*) or tissue or cell (*in vitro*) to determine the biological activity of a substance, such as a hormone or drug. Bioassays are typically conducted to measure the effects of a substance on a living [organism](https://en.wikipedia.org/wiki/Organism) and are essential in the development of new [drugs](https://en.wikipedia.org/wiki/Drugs) and in monitoring environmental [pollutants](https://en.wikipedia.org/wiki/Pollutant). Both are procedures by which the [potency](https://en.wikipedia.org/wiki/Potency_%28pharmacology%29) or the nature of a substance is estimated by studying its effects on [living matter.](https://en.wikipedia.org/wiki/Living_matter) A bioassay can also be used to determine the concentration of a particular constitution of a mixture that may cause harmful effects on organisms or the environment.

### Use of bioassays

Bioassays are procedures that can determine the concentration or purity or biological activity of a substance such as vitamin, hormone or plant growth factor by measuring the effect on an organism, tissue, cells, enzyme or receptor. Bioassays may be [qualitative](https://en.wiktionary.org/wiki/qualitative) or [quantitative](https://en.wiktionary.org/wiki/quantitative). Qualitative bioassays are used for assessing the physical effects of a substance that may not be quantified, such as seeds fail to germinate or develop abnormally [deformity.](https://en.wikipedia.org/wiki/Deformity) An example of a qualitative bioassay includes [Arnold Adolph Berthold'](https://en.wikipedia.org/wiki/Arnold_Adolph_Berthold)s famous experiment on castrated chickens. This analysis found that by removing the testicles of a chicken, it would not develop into a rooster because the endocrine signals necessary for this process were not available. Quantitative bioassays involve estimation of the dose-response curve, how the response changes with increasing dose. That dose-response relation allows estimation of the dose or concentration of a substance associated with a specific biological response, such as the LC50 (concentration killing 50% of the exposed organisms). Quantitative bioassays are typically analyzed using the methods of [biostatistics](https://en.wikipedia.org/wiki/Biostatistics).

### Purpose of bioassays

1. Measurement of the [pharmacological](https://en.wikipedia.org/wiki/Pharmacology) activity of new or chemically undefined substances
2. Investigation of the function of [endogenous mediators](https://en.wikipedia.org/wiki/Endogenous_mediator)
3. Determination of the [side-effect profile,](https://en.wikipedia.org/wiki/Adverse_effect_%28medicine%29) including the degree of [drug toxicity](https://en.wikipedia.org/wiki/Drug_toxicity)
4. Measurement of the concentration of known substances (alternatives to the use of whole animals have made this use obsolete)
5. Assessing the amount of pollutants being released by a particular source, such as [wastewater](https://en.wikipedia.org/wiki/Wastewater) or [urban](https://en.wikipedia.org/wiki/Urban_area) [runoff.](https://en.wikipedia.org/wiki/Surface_runoff)
6. Determining the specificity of certain enzymes to certain substrates.

### Types of bioassays

Bioassays are of two types:

**Quantal:** A quantal assay involves an "all or none response".

**Graded:** Graded assays are based on the observation that there is a proportionate increase in the observed response following an increase in the concentration or dose. The parameters employed in such bioassays are based on the nature of the effect the substance is expected to produce. For example: contraction of smooth muscle preparation for assaying histamine or the study of blood pressure response in case of adrenaline.

A graded bioassay can be performed by employing any of the below-mentioned techniques. The choice of procedure depends on:

1. The precision of the assay required
2. The quantity of the sample substance available
3. The availability of the experimental animals.

### Bioassay Techniques

1. Matching Bioassay
2. Interpolation Method
3. Bracketing Method
4. Multiple Point Bioassay (i.e.-Three-point, Four-point and Six Point Bioassay)
5. Divided bioassay

**Matching Bioassay:** It is the simplest type of the bioassay. In this type of bioassay, response of the test substance taken first and the observed response is tried to match with the standard response. Several responses of the standard drug are recorded till a close matching point to that of the test substance is observed. A corresponding concentration is thus calculated. This assay is applied when the sample size is too small. Since the assay does not involve the recording of concentration response curve, the sensitivity of the preparation is not taken into consideration. Therefore, precision and reliability is not very good.

**Interpolation bioassay:** Bioassays are conducted by determining the amount of preparation of unknown potency required to produce a definite effect on suitable test animals or organs or tissue under standard conditions. This effect is compared with that of a standard. Thus the amount of the test substance required to produce the same biological effect as a given quantity the unit of a standard preparation is compared and the potency of the unknown is expressed as a % of that of the standard by employing a simple formula.

Many times, a reliable result cannot be obtained using this calculation. Therefore it may be necessary to adopt more precise methods of calculating potency based upon observations of relative, but not necessarily equal effects, likewise, statistical methods may also be employed. The data (obtained from either of assay techniques used) on which bioassay are based may be classified as quantal or graded response. Both these depend ultimately on plotting or making assumption concerning the form of DRC.

### Environmental bioassays

Environmental bioassays are generally a broad-range survey of [toxicity.](https://en.wikipedia.org/wiki/Toxicity) A toxicity identification evaluation is conducted to determine what the relevant [toxicants](https://en.wikipedia.org/wiki/Toxicant) are. Although bioassays are beneficial in determining the biological activity within an organism, they can often be time- consuming and laborious. Organism-specific factors may result in data that are not applicable to others in that species. For these reasons, other biological techniques are often employed, including [radio-immunoassays.](https://en.wikipedia.org/wiki/Radioimmunoassay)

## QUALITY CONTROL

Quality control, or QC for short, is a process by which entities review the quality of all factors involved in production. [ISO 9000](https://en.wikipedia.org/wiki/ISO_9000) defines quality control as "A part of quality management focused on fulfilling quality requirements".

This approach places an emphasis on three aspects:

1. Elements such as controls, job management, defined and well managed processes, performance and integrity criteria, and identification of records
2. Competence, such as knowledge, skills, experience, and qualifications
3. Soft elements, such as personnel, [integrity](https://en.wikipedia.org/wiki/Integrity), [confidence,](https://en.wikipedia.org/wiki/Confidence) [organizational culture](https://en.wikipedia.org/wiki/Organizational_culture), [motivation](https://en.wikipedia.org/wiki/Motivation), [team spirit](https://en.wikipedia.org/wiki/Team_spirit), and quality relationships.

Controls include product [inspection](https://en.wikipedia.org/wiki/Inspection), where every product is examined visually, and often using a [stereo microscope](https://en.wikipedia.org/wiki/Stereo_microscope) for fine detail before the product is sold into the external market. Inspectors will be provided with lists and descriptions of unacceptable [product defects](https://en.wikipedia.org/wiki/Product_defect) such as [cracks](https://en.wikipedia.org/wiki/Fracture) or surface [blemishes](https://en.wiktionary.org/wiki/blemish) for example. The quality of the outputs is at risk if any of these three aspects is deficient in any way.

Quality control emphasizes [testing](https://en.wikipedia.org/wiki/Test_method) of [products](https://en.wikipedia.org/wiki/Product_%28business%29) to uncover defects and reporting to management who make the decision to allow or deny product release, whereas [quality assurance](https://en.wikipedia.org/wiki/Quality_assurance) attempts to improve and stabilize production (and associated processes) to avoid, or at least minimize, issues which led to the defect(s) in the first place. For contract work, particularly work awarded by government agencies, quality control issues are among the top reasons for not renewing a contract.

## SHELF LIFE OF PRODUCTS

Shelf life is the length of time that a commodity may be stored without becoming unfit for use, consumption, or sale. In other words, it might refer to whether a commodity should no longer be on a pantry shelf (unfit for use), or just no longer on a supermarket shelf (unfit for sale, but not yet unfit for use). It applies to [cosmetics,](https://en.wikipedia.org/wiki/Cosmetics) [foods](https://en.wikipedia.org/wiki/Food) and [beverages,](https://en.wikipedia.org/wiki/Drink) [medical devices,](https://en.wikipedia.org/wiki/Medical_device) [medicines,](https://en.wikipedia.org/wiki/Medicine) [explosives](https://en.wikipedia.org/wiki/Explosives), [pharmaceutical drugs,](https://en.wikipedia.org/wiki/Pharmaceutical_drug) [chemicals](https://en.wikipedia.org/wiki/Chemical), [car tires,](https://en.wikipedia.org/wiki/Tire) [batteries](https://en.wikipedia.org/wiki/Battery_%28electricity%29), and many other [perishable](https://en.wikipedia.org/wiki/Decomposition) items. In some regions, an advisory ***best before*,** mandatory ***use by***, or ***freshness date*** is required on packaged perishable foods.

# Background

Shelf life is the recommended maximum time for which products or fresh (harvested) produce can be stored, during which the defined quality of a specified proportion of the goods remains acceptable under expected (or specified) conditions of distribution, storage and display. Most expiration dates are used as guidelines based on normal and expected handling and exposure to temperature. Use prior to the expiration date does not guarantee the safety of a food or drug, and a product is not necessarily dangerous or ineffective after the expiration date.

According to the USDA, "canned foods are safe indefinitely as long as they are not exposed to freezing temperatures, or temperatures above 90 °F (32.2° C). If the cans look ok, they are safe to use. Discard cans that are dented, rusted, or swollen. High-acid canned foods (tomatoes, fruits) will keep their best quality for 12 to 18 months; low-acid canned foods (meats, vegetables) for 2 to 5 years 80 °F (27 °C).

"Sell by date" is a less ambiguous term for what is often referred to as an "expiration date". Most food is still edible after the expiration date. A product that has passed its shelf life might still be safe, but quality is no longer guaranteed. In most food stores, waste is minimized by using [stock](https://en.wikipedia.org/wiki/Stock_rotation) [rotation](https://en.wikipedia.org/wiki/Stock_rotation), which involves moving products with the earliest sell by date from the warehouse to the sales area, and then to the front of the shelf, so that most shoppers will pick them up first and thus they are likely to be sold before the end of their shelf life. This is important, as consumers enjoy fresher goods, and furthermore some stores can be fined for selling out of date products; most if not all would have to mark such products down as [wasted,](https://en.wikipedia.org/wiki/Food_waste) resulting in a financial loss.

Shelf life depends on the degradation mechanism of the specific product. Most can be influenced by several factors: exposure to [light,](https://en.wikipedia.org/wiki/Light) [heat,](https://en.wikipedia.org/wiki/Heat) moisture, transmission of [gas](https://en.wikipedia.org/wiki/Gas)es, mechanical [stress](https://en.wikipedia.org/wiki/Stress_%28physics%29) and contamination by things such as micro-organisms. Product quality is often mathematically modelled around a parameter (concentration of a chemical compound, a microbiological index, or moisture content).

For some foods, health issues are important in determining shelf life. Bacterial contaminants are ubiquitous, and foods left unused too long will often be contaminated by substantial amounts of bacterial colonies and become dangerous to eat, leading to [food poisoning.](https://en.wikipedia.org/wiki/Food_poisoning) However, shelf life alone is not an accurate indicator of how long the food can safely be stored. For example, pasteurized milk can remain fresh for five days after its sell-by date if it is refrigerated properly. In contrast, if milk already has harmful bacteria, the use-by dates become irrelevant.

The expiration date of pharmaceuticals specifies the date the manufacturer guarantees the full potency and safety of a drug. Most medications continue to be effective and safe for a time after the expiration date. A rare exception is a case of renal tubular acidosis purportedly caused by expired [tetracycline.](https://en.wikipedia.org/wiki/Tetracycline) A study conducted by the U.S. Food and Drug Administration covered over 100 drugs, prescription and over-the-counter. The study showed that about 90% of them were safe and effective as long as 15 years past their expiration dates. Joel Davis, a former FDA expiration-date compliance chief, said that with a handful of exceptions - notably nitroglycerin, insulin and some liquid antibiotics - most expired drugs are probably effective.

Shelf life is not significantly studied during drug development, and drug manufacturers have economic and liability incentives to specify shorter shelf lives so that consumers are encouraged to discard and repurchase products. One major exception is the [Shelf Life Extension Program](https://en.wikipedia.org/wiki/Shelf_Life_Extension_Program) (SLEP) of the [U.S. Department of Defense](https://en.wikipedia.org/wiki/U.S._Department_of_Defense) (DoD), which commissioned a major study of drug efficacy from the FDA starting in the mid-1980s. One criticism is that the U.S. [Food and Drug](https://en.wikipedia.org/wiki/Food_and_Drug_Administration) [Administration](https://en.wikipedia.org/wiki/Food_and_Drug_Administration) (FDA) refused to issue guidelines based on SLEP research for normal marketing of pharmaceuticals even though the FDA performed the study. The SLEP and FDA signed a memorandum that scientific data could not be shared with the public, public health departments, other government agencies, and drug manufacturers. State and local programs are not permitted to participate. The failure to share data has caused foreign governments to refuse donations of

expired medications. One exception occurred during the 2010 [Swine Flu Epidemic](https://en.wikipedia.org/wiki/Swine_flu_%28outbreak_in_the_US%29) when the FDA authorized expired [Tamiflu](https://en.wikipedia.org/wiki/Tamiflu) based on SLEP Data. The SLEP discovered that drugs such as [Cipro](https://en.wikipedia.org/wiki/Cipro) remained effective nine years after their shelf life, and, as a cost-saving measure, the US military routinely uses a wide range of SLEP tested products past their official shelf life if drugs have been stored properly.

[Preservatives](https://en.wikipedia.org/wiki/Preservative) and [antioxidants](https://en.wikipedia.org/wiki/Antioxidant) may be incorporated into some food and drug products to extend their shelf life. Some companies use [induction sealing](https://en.wikipedia.org/wiki/Induction_sealing) and [vacuum](https://en.wikipedia.org/wiki/Vacuum)/oxygen-barrier pouches to assist in the extension of the shelf life of their products where oxygen causes the loss.

The DoD Shelf-Life Program defines shelf-life as,

The total period of time beginning with the date of manufacture, date of cure (for elastomeric and rubber products only), date of assembly, or date of pack (subsistence only), and terminated by the date by which an item must be used (expiration date) or subjected to inspection, test, restoration, or disposal action; or after inspection/laboratory test/restorative action that an item may remain in the combined wholesale (including manufacture's) and retail storage systems and still be suitable for issue or use by the end user. Shelf-life is not to be confused with service-life (defined as, A general term used to quantify the average or standard life expectancy of an item or equipment while in use. When a shelf-life item is unpacked and introduced to mission requirements, installed into intended application, or merely left in storage, placed in pre- expended bins, or held as [bench stock,](https://en.wikipedia.org/w/index.php?title=Bench_stock&action=edit&redlink=1) shelf-life management stops and service life begins).

Shelf life is often specified in conjunction with a specific product, package, and distribution system. For example, an [MRE](https://en.wikipedia.org/wiki/MRE) [field ration](https://en.wikipedia.org/wiki/Field_ration) is designed to have a shelf life of three years at 80 °F (27 °C) and six months at 100 °F (38 °C).

### Temperature control

Nearly all chemical reactions can occur at normal temperatures (although different reactions proceed at different rates). However most reactions are accelerated by high temperatures and the degradation of foods and pharmaceuticals is no exception. The same applies to the breakdown of many chemical explosives into more unstable compounds. [Nitroglycerine](https://en.wikipedia.org/wiki/Nitroglycerin) is notorious. Old

[explosives](https://en.wikipedia.org/wiki/Explosive_material) are thus more dangerous (i.e. liable to be triggered to explode by very small disturbances, even trivial jiggling) than more recently manufactured explosives. [Rubber](https://en.wikipedia.org/wiki/Rubber) products also degrade as [sulphur](https://en.wikipedia.org/wiki/Sulfur) [bonds](https://en.wikipedia.org/wiki/Chemical_bond) induced during [vulcanization](https://en.wikipedia.org/wiki/Vulcanization) revert; this is why old [rubber bands](https://en.wikipedia.org/wiki/Rubber_band) and other rubber products soften and get crispy, and lose their elasticity as they age.

The usually quoted [rule of thumb](https://en.wikipedia.org/wiki/Rule_of_thumb) is that chemical reactions double their rate for each temperature increase of 10 °C (18 °F) because [activation energy](https://en.wikipedia.org/wiki/Activation_energy) barriers are more easily surmounted at higher temperatures. However, as with many rules of thumb, there are many [caveats](https://en.wiktionary.org/wiki/caveat) and exceptions. The rule works best for reactions with [activation energy](https://en.wikipedia.org/wiki/Activation_energy) values around 50 kJ/mole; many of these are important at the usual temperatures we encounter. It is often applied in shelf life estimation, sometimes wrongly. There is a widespread impression, for instance in industry, that "triple time" can be simulated in practice by increasing the temperature by 15 °C (27 °F), e.g., storing a product for one month at 35 °C (95 °F) simulates three months at 20 °C (68 °F). This is mathematically incorrect (if the rule was precisely accurate the required temperature increase would be about 15.8 °C (28.4 °F)), and in any case the rule is only a rough approximation and cannot always be relied on.

The same is true, up to a point, of the chemical reactions of living things. They are usually catalyzed by [enzymes](https://en.wikipedia.org/wiki/Enzyme) which change reaction rates, but with no variation in catalytic action, the rule of thumb is still mostly applicable. In the case of [bacteria](https://en.wikipedia.org/wiki/Bacterium) and [fungi](https://en.wikipedia.org/wiki/Fungi), the reactions needed to feed and reproduce speed up at higher temperatures, up to the point that the proteins and other compounds in their cells themselves begin to break down, or [denature,](https://en.wikipedia.org/wiki/Denaturation_%28biochemistry%29) so quickly that they cannot be replaced. This is why high temperatures kill bacteria and other micro-organisms: 'tissue' breakdown reactions reach such rates that they cannot be compensated for and the cell dies. On the other hand, 'elevated' temperatures short of these result in increased growth and reproduction; if the organism is harmful, perhaps to dangerous levels.

Just as temperature increases speed up reactions, temperature decreases reduce them. Therefore, to make explosives stable for longer periods, or to keep rubber bands springy, or to force bacteria to slow down their growth, they can be cooled. That is why shelf life is generally extended by temperature control: ([refrigeration](https://en.wikipedia.org/wiki/Refrigeration), [insulated shipping containers,](https://en.wikipedia.org/wiki/Insulated_shipping_container) controlled [cold chain](https://en.wikipedia.org/wiki/Cold_chain), etc.) and why some medicines and foods *must* be refrigerated. Since such storing of such goods is

temporal in nature and shelf life is dependent on the temperature controlled environment, they are also referred to as [cargo](https://en.wikipedia.org/wiki/Cargo) even when in special storage to emphasize the inherent time- temperature sensitivity matrix.

[Temperature data loggers](https://en.wikipedia.org/wiki/Temperature_data_logger) and [time temperature indicators](https://en.wikipedia.org/wiki/Time_temperature_indicator) can record the temperature history of a shipment to help estimate their remaining shelf life.

According to the [USDA,](https://en.wikipedia.org/wiki/USDA) "foods kept frozen continuously are safe indefinitely."

### Product Packaging

Barrier packaging can often help control or extend shelf life. When moisture content is a mechanism for product degradation, packaging with a low [moisture vapor transmission rate](https://en.wikipedia.org/wiki/Moisture_vapor_transmission_rate) and the use of [desiccants](https://en.wikipedia.org/wiki/Desiccant) help keep the moisture in the package within acceptable limits. When [oxidation](https://en.wikipedia.org/wiki/Oxidation) is the primary concern, packaging with a low [oxygen transmission rate](https://en.wikipedia.org/wiki/Oxygen_transmission_rate) and the use of [oxygen absorbers](https://en.wikipedia.org/wiki/Oxygen_absorber) can help extend the shelf life. [Produce](https://en.wikipedia.org/wiki/Produce) and other products with respiration often require packaging with controlled barrier properties. The use of a [modified atmosphere](https://en.wikipedia.org/wiki/Modified_atmosphere) in the package can extend the shelf life for some products. Some [active packaging](https://en.wikipedia.org/wiki/Active_packaging) is also available with antibacterial properties.

### Parallel names

***Best before*** or ***best by*** dates appear on a wide range of frozen, dried, tinned and other foods. These dates are only advisory and refer to the quality of the product, in contrast with *use by* dates, which indicate that the product may no longer be safe to consume after the specified date. Food kept after the *best before* date will not necessarily be harmful, but may begin to lose its optimum flavour and texture. Eggs are a special case, since they may contain [salmonella](https://en.wikipedia.org/wiki/Salmonella) which multiplies over time; they should therefore be eaten before the *best before* date, which is, in the USA, a maximum of 45 days after the eggs are packed.

Sometimes the packaging process involves using pre-printed labels, making it impractical to write the *best before* date in a clearly visible location. In this case, wording like *best before see bottom* or *best before see lid* might be printed on the label and the date marked in a different location as indicated. Generally, foods that have a *use by* date written on the packaging must not

be eaten after the specified date. This is because such foods usually go bad quickly and may be injurious to health if spoiled. It is also important to follow storage instructions carefully for these foods (for example, if they specify that the product must be [refrigerated](https://en.wikipedia.org/wiki/Refrigeration)).

Bathroom products and toiletries usually state a time in months from the date the product is opened, by which they should be used. This is often indicated by a [graphic of an open tub](https://en.wikipedia.org/wiki/Period-after-opening_symbol), with the number of months written inside (e.g., "12M" means use the product within 12 months of opening).[[19]](https://en.wikipedia.org/wiki/Shelf_life#cite_note-orus1-19) Similarly, some food products say "eat within X days of opening".

#### Open dating

***Open dating*** is the use of a date stamped on the package of a food product to help determine how long to display the product for sale. This benefits the consumer by ensuring that the product is of best quality when sold. An open date does not supersede a [use-by date,](https://en.wikipedia.org/wiki/Shelf_life#Use_by) if shown, which should still be followed.

#### Sell by / display until

These dates are intended to help keep track of the stock in stores. Food that has passed its *sell by* or *display until* date, but has not yet reached its use by / best before date will still be edible, assuming it has been stored correctly. It is common practice in large stores to throw away such food, as it makes the stock control process easier; another common practice is for wholesalers to repurchase the expired product and resell it to [discount stores](https://en.wikipedia.org/wiki/Discount_store) at much lower [clearance sale](https://en.wikipedia.org/wiki/Closeout_%28sale%29) prices. These practices reduce the risk of customers unknowingly buying food without looking at the date, only to find out the next day that they cannot use it. Tampering with the posted date is illegal in many countries.

Most stores will [rotate stock](https://en.wikipedia.org/wiki/Stock_rotation) by moving the products with the earliest dates to the front of shelving units, which encourages customers to buy them first and hopefully saves them from having to be either marked down or thrown away, both of which would result in financial loss.

# Beer

#### Freshness date

A *freshness date* is the date used in the American brewing industry to indicate either the date the beer was bottled or the date before which the beer should be consumed. Beer is perishable. It can be affected by light, air, or the action of bacteria. Although beer is not legally mandated in the United States to have a shelf life, freshness dates serve much the same purpose and are used as a marketing tool.

#### Beginnings of freshness dating

[General Brewing Company](https://en.wikipedia.org/wiki/Eugene_Selvage#Lucky_Lager_Brewing_Company) of San Francisco marketed their [Lucky Lager](https://en.wikipedia.org/wiki/Lucky_Lager) Beer as "Age Dated" as early as late 1935. They stamped a date on each can lid to indicate that the beer was brewed before that date. This was not to ensure that the beer was "fresh" but to ensure that it had been aged properly. So many breweries had rushed beer to market before it was ready when Prohibition ended, that customers were wary of getting "green" beer. The [Boston Beer Company,](https://en.wikipedia.org/wiki/Boston_Beer_Company) maker of [Samuel Adams](https://en.wikipedia.org/wiki/Samuel_Adams_%28beer%29), was among the first contemporary brewers to start adding freshness dates to their product line in 1985. For ten years there was a slow growth in brewers adding freshness dates to their beer. The practice rapidly grew in popularity after the [Anheuser-Busch](https://en.wikipedia.org/wiki/Anheuser-Busch) company's heavily marketed "Born-On dates" starting in 1996. Many other brewers have started adding freshness dates to their products, but there is no standard for what the date means. For some companies, the date on the bottle or can will be the date that the beer was bottled; others have the date by which the beer should be consumed.