## COFALEC

CONFEDERATION OF EUROPEAN YEAST PRODUCERS

# THE WORLD OF YEAST

Beyond traditional uses, new innovative applications of yeast represent an opportunity for a more sustainable world.

9 Boulevard Malesherbes, 75008 Paris – contact@cofalec.com COFALEC has created this document to provide an overview of the countless uses of yeast in today's world.

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## WHAT YOU NEED TO KNOW ABOUT YEAST



# One cubic centimeter of fresh yeast contains up to 10 billion living yeast cells !

Each of these cells is a microscopic fungus.

This unicellular eukaryotic micro-organism, is like our own human cells, equipped with a nucleus and organelles well bounded by membranes, which allows for very complex metabolism.

Yeasts have a particular metabolism that enables them to live both aerobically (in the presence of oxygen) and anaerobically (without oxygen).

#### IN AEROBIC MODE, yeast find optimal conditions for rapid multiplication.

This mode is used on a large scale in a yeast production factory. In this industrial process, the energy required for yeast growth and multiplication is mainly provided by a substrate rich in sugar: molasses or low green syrup, co-products of sugar production from sugar beet (in Europe) or sugar cane.

## IN ANAEROBIC MODE, when yeast is deprived of oxygen, it turns to the surrounding matter to trigger the famous fermentation process.

They transform the rich-sugar substrate into alcohol, flavor compounds and carbon dioxide, making it possible to produce bread, wine, beer and other alcohols.



## YEAST, A MICRO-ORGANISM WITH MULTIPLE APPLICATIONS

## BAKERY

1



- Leavening agent
- Flavour development
- Fermentation optimisation
- Nutritional enhancement

## 2 ALCOHOLIC BEVERAGES



- Alcoholic fermentation
- Aroma and flavour profiles
- Fermentation optimisation

# **3** NUTRITION & HUMAN HEALTH



- Probiotic effect & immune system support
- High nutritional value (amino-acid, vitamins and minerals)

### **5** SUSTAINABLE AGRICULTURE



- Biofertilizers and biofungicides
- Biocontrol and biostimulation agents
- Plant health enhancement

## **7** POLLUTION CONTROL & WASTE TREATMENT



- Bioremediation
- Wastewater treatment
- Organic pollutant degradation
- Heavy metal bioaccumulation

### **4** ALTERNATIVE PROTEIN SOURCES

- High-quallity protein profile
- Plant-based foods nutritional enhancement
- Protein extraction for dairy alternatives

## **6** BIOENERGY



- Bioethanol
- Many other prospects such as biodiesel and biogas

### 8 PRECISION FERMENTATION

- Biological factory
- Tailored fermentation process
- Customised flavour
- Pharmaeuticals and food ingredients production





## BAKERY

Bakers' yeast is both the oldest and best-known yeast.



## **ROLE OF YEAST**

**Raising dough:** when fermenting, yeast produces carbon dioxide. This gas expands the gluten proteins in the flour and causes the dough to rise.

**Revealing flavours and taste:** fermentation generates volatile aromatic compounds. When baked, they give breads from all over the world their subtle and complex flavour profile.

#### Nutrition and health:

- Long fermentation improves the B-vitamin content of bread, particularly folate, an important vitamin for pregnant women. Folate levels in bread are 2.5 times higher when baker's yeast is used rather than baking powder. Folate (vitamin B9) plays a vital role in essential cell metabolism, such as the synthesis of nucleic and amino acids. This plays an important role against a number of diseases such as cardiovascular diseases, Alzheimer's and certain forms of cancer.
- Yeast promotes the bio-accessibility of minerals present in cereal flours (i.e., zinc, iron and magnesium).
- Yeast fermentation improves the nutritional properties and digestibility of cereals by breaking down certain constituents (i.e., proteins, fibres, sugars).
- Several compounds generated by fermentation, such as certain peptides extracted from flour, have a calming effect on the nervous system.



## A SHORT HISTORY

#### Fermentation has been key to food preservation for at least 15,000 years

- The first traces of bread made from fermented cereals date back 14,400 years to present-day Jordan, i.e. over 5,000 years before the invention of agriculture.
- As early as 3000 BC, the Egyptians and Babylonians were using fermentation to make leavened bread, although they were unable to explain this mystery.
- However, it was not until the work of Louis Pasteur in 1860 that yeast was identified as the microorganism responsible for alcoholic fermentation.
- The first industrial yeast plants appeared in the 18th century in northern Europe.
- The continuous addition method introduced in Germany in 1915 marked the birth of the modern yeast industry, synchronising the addition of the sugars on which yeast is fed. This avoids any excess sugar in the must, and thus the undesired formation of alcohol.
- Modern yeast production is still based on these same principles. Since then, it has been largely perfected thanks to a better knowledge of raw materials, yeast biology and process automation.



### **KEY DATA ON THE EUROPEAN BAKING MARKET**

- In 2010, the European bread market represented around 32 million tonnes in the 27 EU countries (source: Commission study).
- In the European Union as a whole, the market is divided equally between small-scale and industrial bakers, although there are major differences from one country to another.
- The production of baker's yeast remains the dominant activity of the yeast industry in the EU, although its relative weight is declining in favour of new applications.





### FORMAT OF YEAST USED

Currently, only strains of *Saccharomyces cerevisiae* are used in baking. Baker's yeast is available in a wide range of concentration levels (from 5% to 85% water) to meet the current needs of artisanal and industrial bakers.



#### LIQUID YEAST

Suitable for small-scale and industrial use, with a positive cold shelf-life in positive refrigeration.



#### YEAST In the form of packaged or crumbled blocks

with a positive cold shelf-life. Practical and economical, it is widely used.



DRY YEAST

Can be stored at room temperature but must be rehydrated before use. It is popular in regions where temperatures and humidity are high.



#### INSTANT DRY YEAST

It does not need to be rehydrated before being added to flour. It can therefore be used as easily as compressed yeast.



#### DRY YEAST WITH REDUCING POWER

In granulated form, it is used by pizza makers because it produces less gas during fermentation and improves dough elasticity which facilitates the shaping of the pizza.



## **PROSPECTS AND INNOVATIONS**

#### Research into baker's yeast remains very dynamic.

#### **Recent innovations since 2000**

During the bread-making process, baker's yeast is exposed to numerous environmental stresses such as air-drying, freezing-thawing and high sucrose concentrations. Three types of yeast have been developed to optimise fermentation under these different stress conditions:

- Osmo-tolerant yeasts (resist osmotic pressure in sweeter doughs).
- Acid-resistant yeasts (resist the stress of antifungal agents in long-life breads).
- Yeasts for frozen doughs (used for long fermentation and in fermentation tanks).

#### Innovations to contribute to human health

Research has also led to the development of various natural `clean label' yeast-based solutions to improve the nutritional quality of bread.

- Deactivated salt-reducing yeasts make it possible to reduce the salt content of breads by 20 to 40% without altering the taste or texture.
- New yeast strains improve the bioavailability of vitamins and minerals in breads.

#### Future innovations to adapt to new uses

- Pursue the development of organic yeast production.
- Develop moisture tolerant yeast, that are ideal to ensure stability of yeast present in bread mixes, and allow an all-in-one bread mix.











## **ALCOHOLIC BEVERAGES**

Yeast is an invisible but essential partner of all alcoholic beverages, including wines, beers, and spirits.



## **ROLE OF YEAST**

#### **CONVERTING SUGARS INTO ALCOHOL**

Ubiquitous in nature, yeast has been used for centuries by humans in fermentation processes, particularly in winemaking and brewing.

In the absence of oxygen (anaerobic), yeast acts as a catalyst in a reaction transforming sugars from fruit (grapes, for wine production) or cereals (malt, for beer production) into alcohol (ethanol). This reaction releases carbon dioxide, flavours (volatile aromatic compounds) and energy.

Fermentation should not be confused with distillation, a complementary process which, by heating in an alembic, concentrates the alcoholic content of the previously fermented liquid.

#### EXPRESSING THE AROMATIC POTENTIAL OF ALCOHOLIC BEVERAGES

#### Wine yeasts, unveiling the wine sensory

There are over 300 selected wine yeasts strains available to winemakers, *Saccharomyces cerevisiae* being the main specie. Each strain can reveal different aromatic compounds during fermentation: - **Varietal aromas**, naturally present in grapes in the form of odourless "aromatic precursors". Revealed by the action of yeast, they are characterized by aromas such as citrus, floral and fruity notes.

- Aromas derived from the fermentation process, such as certain fruity aromas, are associated to the yeast metabolism in parallel of the fermentation reaction. The aromatic profile of each wine combines these aromas to give it a complex and fascinating style! Moreover, yeast can also have an impact on sensations of volume in the mouth, freshness, or more generally on the taste profile.

#### Two main types of brewer's yeast

Both types of fermentation are crucial for creating the large range of beer styles enjoyed worldwide:

• Ale fermentation with top-fermenting yeast that rise to the surface of the tank towards the end of fermentation (*Saccharomyces cerevisiae*).

They operate at warmer temperatures, between 15-20°C. This fermentation is relatively quick, lasting from a few days to a couple of weeks, and produces beers with robust, fruity, and complex flavors. These beers have a higher alcoholic content and a more fruity, estery and malty character.

• Lager fermentation with bottom-fermenting yeast that settle to the bottom of the tank towards the end of fermentation (*Saccharomyces uvarum* and *pastorianus* or *carlsbergiensis*).

They ferment at cooler temperatures, usually between 7-15°C. This process is slower, often taking up to several weeks, and results in cleaner, crispier, and more mellow flavours. These beers have a lower alcoholic and higher CO<sub>2</sub> content, making them more sparkling, potentially exhibiting hints of sulphur.



## FORMAT OF YEAST USED

Alcoholic fermentation is performed by either liquid yeast (mainly beer and spirits) or active dry yeast (mainly wine). Active dry yeast is one format of yeast used since the 1970's for alcoholic fermentation:
Active dry yeast enables sugars to be fermented efficiently, either after rehydration or directly in the must or wort, in the case of certain yeast strains.

• Active dry yeast offers excellent stability and longer shelf life. It gives winemakers and brewers greater flexibility and control over the fermentation process.





## **A SHORT HISTORY**

5400 BC: Large-scale wine production in the northern Zagros mountains (current-day Iran).
3140 BC: Estimated date of the first wine yeast residues found in ancient Egyptian wine jars.
1860: Louis Pasteur establishes a link between microscopic yeast cells and the fermentation process.
1888: Emil Hansen perfects Pasteur's method for isolating pure yeast cultures.

**1890:** Hermann Müller-Thurgau creates the concept of must inoculation with selected yeast cultures. **1965:** First active dry yeast industrially produced in California.

### **KEY DATA**

#### European wine market

• Europe is the largest wine-producing and consuming area in the world.

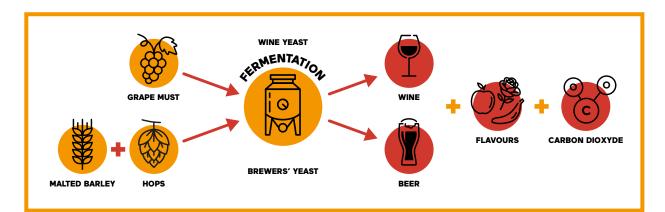
- The European wine market was valued at around US\$155 billion in 2022.
- The top wine-producing countries in Europe include Italy, France, and Spain. They account for about half of the world's wine production.

#### European beer market

- Europe is one of the largest beer markets in the world, valued at approximately US\$ 144 billion in 2022, with steady growth projected in the coming years.
- The top beer-producing countries in Europe include Germany, the United Kingdom, Poland, and Spain. Germany is the largest producer, brewing over 90 million hectolitres annually.

## **PROSPECTS AND INNOVATIONS**

- **Research into new natural yeast strains,** including non-*Saccharomyces* species, to diversify wine and beer sensory styles and provide certain technological advantages.
- **Reducing chemical inputs**: yeast can reduce the use of chemical inputs in the winemaking process, as they prevent the development of faults or spoilage microbes.
- **Non-GMO yeast improvement:** for the needs of GMO-averse consumers, to develop new yeast strains that offer improved performance and fermentation capabilities (e.g. higher alcohol tolerance, faster fermentation rates, increased resistance to stress conditions).
- Fermentation efficiency and optimal product quality: better real-time monitoring and control of fermentation parameters, via advanced sensors and automation technologies.
- **Sustainability and environmental impact:** develop new, less carbon-and energy-intensive yeast strains capable of efficiently fermenting alternative substrates, such as agricultural waste or non-traditional raw materials, and contribute to circular economy practices.
- **Bioinformatics and data analysis:** used to better understand yeast genetics, metabolism and fermentation dynamics, and better target strain development and process optimization.
- Artificial intelligence: to predict fermentation outcomes and optimize yeast performance.
- **Hybrid yeasts:** to combine the properties of different yeast strains to create hybrid yeasts, offering brewers and winemakers new options for product differentiation.







8 HUMAN HEALTH



## **NUTRITION AND HUMAN HEALTH**

Yeast is a powerful ally to support the everyday health of humans. As such, it is an essential tool in the service of a global health thanks to its beneficial role on microbiota.

## ROLE OF YEAST

#### → S. boulardii

#### 1. Improvement

of the intestinal barrier: strengthens the intestinal barrier, with an antagonism effect against pathogens<sup>(1)</sup>.

#### 2. Reduction

of gut inflammation: thanks to its anti-inflammatory properties, it helps reduce inflammation in the intestine, such as Crohn's disease and ulcerative colitis.

#### 3. Immune support:

about 70% of the immune system resides in the intestine. *S. boulardii* helps balance the gut microbiota, thereby strengthening the immune response. It can also positively influence the production of immunoglobulins.

#### 4. Prevention

of intestinal disorders:

used in more than 90 countries to prevent and treat various gastrointestinal disorders such as antibiotic-associated diarrhoea and traveller's diarrhoea.

#### → S. cerevisiae

**1. Beneficial Health Effects on digestive comfort:** intestinal pain, bloating and regulation of transit as well as protective properties for vaginal flora.

#### 2. Nutritional Yeast: dried and inactivated

*S. cerevisiae* is rich in macronutrients, providing essential amino acids.

*S. cerevisiae* is particularly rich in B-vitamins, such as B1, B2, B3, B5, B6, B7, B9, and B12 when fortified.

#### 3. Vitamin and Mineral Yeast:

can be added to any food as flakes or powder, enriched, fortified (with added vitamins and minerals) or unfortified.

*S. cerevisiae* produces vitamin D2 when exposed to UV light, making it a safe, vegan source of vitamin D. During fermentation, it can be supplemented with vitamins (such as B-vitamins) and/or minerals (selenium, iron, and zinc).

#### 4. Other Yeast Cell Components

• Beta-1,3-/1,6-glucan is a complex polysaccharide found in the cell walls of *S. cerevisiae.* It strengthens the immune system<sup>(2)</sup> and may support cancer treatments<sup>(3)</sup>.

• Mannan-oligosaccharides (MOS) are short chains of mannose that are known to confer health benefits when ingested. These prebiotics with bio-active properties support microbiome balance.

- Glutathione plays a crucial role in the cellular response to oxidative stress thanks to its ability to eliminate free radicals. It can also stimulate other defence processes.
- Choline (formerly classified as B-vitamin) plays an essential role for liver, muscle, brain function, lipid metabolism, cellular membrane composition, and prenatal health.

(1) Pais P, Almeida V, Yılmaz M, Teixeira MC. Saccharomyces boulardii: What Makes It Tick as Successful Probiotic? J Fungi (Basel). 2020 (2) Stier, H., Ebbeskotte, V. & Gruenwald, J. Immune-modulatory effects of dietary Yeast Beta-1,3/1,6-D-glucan. Nutr J 13, 38 (2014).

(3) Vlassopoulou et al. Effects of fungal beta-glucans on health – a systematic review of randomized controlled trials Fonction alimentaire. , 2021, 12 , 3366-3380



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## **KEY DATA**

Yeast has a very high nutritional value ← Source: DIN specification "Characteristics of Fresh and Dry Baker's Yeast", DIN Spec 91473

## **SPECIES OF YEAST USED**

| NUTRITIONAL ASPECTS PER 100G |                                 |  |
|------------------------------|---------------------------------|--|
| Nutrient                     | Amount per 100g<br>(Dry Matter) |  |
| Energy                       | 1610 kJ                         |  |
| Fat                          | 6% ± 2%                         |  |
| Carbohydrates                | 15% ± 9%                        |  |
| Fiber                        | 28% ± 5%                        |  |
| Protein                      | 50% ± 9%                        |  |
| Potassium                    | 0.6% - 2.5%                     |  |
| Sodium                       | < 1%                            |  |
| Calcium                      | 0.02% - 0.15%                   |  |
| Magnesium                    | 0.03% - 0.25%                   |  |
| Zinc                         | >0.005% (>50 ppm)               |  |
| Iron                         | 0.001% - 0.1%                   |  |
| Vitamin B1                   | 2 – 15 mg/100g                  |  |
| Vitamin B2                   | 6 – 8 mg/100g                   |  |
| Vitamin B6                   | 2 – 6 mg/100g                   |  |
| Folic Acid                   | 2 – 4 mg/100g                   |  |
| Niacin (Vitamin PP)          | 10 - 60 mg/100g                 |  |
| Biotin                       | 0.05 - 0.25 mg/100g             |  |

#### Two species of yeast are mostly used for applications in nutrition and human health:

- **S. boulardii:** As early as 1953, the probiotic yeast, *S. boulardii*, was marketed as a medicine to prevent acute diarrhoea. Since then, this microorganism has been considered in more than 100 clinical studies analysing its benefits for gastrointestinal health, leading to good digestive and immune health and therefore, general well-being.
- **S. cerevisiae:** commonly known as baker's yeast, it can be consumed in its whole form as nutritional inactivated yeast or vitamin and mineral fortified or enriched inactivated yeast. Components of the *S. cerevisiae* cell wall and cytosol offer a variety of proven health benefits<sup>(4)</sup>. Recent research has shown that certain strains are also qualified as a probiotic microorganism.



## **A SHORT HISTORY**

- **Saccharomyces cerevisiae var boulardii (S. boulardii)** was discovered by French microbiologist Henri Boulard in 1923 in Southeast Asia. During a cholera epidemic, he noticed that people drinking a mixture of lychee and mangosteen peels did not develop diarrhoea. He isolated a yeast strain from these peels, named *Saccharomyces boulardii*, marking the birth of the first probiotic yeast.
- **Saccharomyces cerevisiae (S. cerevisiae)**: extensively used in bakeries and breweries, recent reports also highlight its health benefits, including anti-infective properties, antioxidant activities, and other probiotic-related effects.

## **PROSPECTS AND INNOVATIONS**

#### Yeast Microbial Consortia

- **S. boulardii:** Research continues to explore new applications for *S. boulardii.* Beyond the intestine, clinical studies have shown its effectiveness in the case of skin imperfections.
- **S. cerevisiae:** Research on *S. cerevisiae* cell wall components include potential bowel transit benefits from mannan-oligosaccharides, possible synergy of yeast beta-1,3/1,6-glucan with respiratory vaccines, and iron yeast benefits for female athletes<sup>(5)</sup>.

(4) Lesage, G., and H. Bussey. "Cell Wall Assembly in Saccharomyces Cerevisiae." Microbiology and Molecular Biology Reviews, vol. 70, no. 2, 1 June 2006.

(5) Gemilang Lara Utama, et al. "Potential Application of Yeast Cell Wall Biopolymers as Probiotic Encapsulants." Polymers, vol. 15, no. 16, 20 Aug. 2023.











## ALTERNATIVE PROTEIN SOURCES

Yeast is a highly promising solution to address the escalating global demand for proteins, to mitigate the environmental impact of animal protein production and to reduce Europe's heavy dependence on imported problematic plant protein for animal feed (e.g. soya).

## **ROLE OF YEAST**

#### As a direct natural protein source

Proteins are composed of amino acids, there are 20 types of them, 9 of which are essential and must come from our diet. Animal proteins (meat, eggs, dairy) provide these essential amino acids, unlike most plant proteins. This is why vegans often need dietary supplements. However, yeast is an exceptional source of all essential amino acids:

• Protein Content: Yeast is 45% protein by dry weight, and 50% of these are essential amino acids.

• Digestibility: Comparable to egg proteins, yeast proteins are highly digestible.

• Nutritional Value: In addition to its protein content, yeast is rich in fibre, vitamins, and minerals.

Yeast can be consumed as a powder, condiment, or food industry ingredient, rounding off the taste of soups, sauces, stock cubes, snacks, and ready meals.

#### PROCESS:

Yeast extract and yeast-based ingredients process

#### As a fermenting agent

Reducing animal proteins in favour of plant-based proteins is key for sustainability and a healthy diet, yet taste and food habits hinder consumer acceptance. Yeast-based fermentation can optimise nutritional and sensory qualities of plantbased foods:

• Flavour Improvement: Certain yeast strains eliminate unwanted tastes (e.g., hexanal's herbaceous flavour) and improve texture.

• Microbial Synergy: Yeast can ferment proteinrich plants (e.g., legumes) in combination with bacteria, improving taste and digestibility.

• Innovative Products: Fermented plant-based products, such as dairy-free cheese substitutes from almond, walnut, pea, and soy purées, present vast innovation opportunities.

#### PROCESS:

Living yeast production technology



## KEY DATA

**A high Protein Content:** Yeast benefits from a high protein content, ranging from 45–65% on a dry weight basis vs. 8–14% for plant sources like grains. Yeast protein is highly digestible with a Protein Digestibility Corrected Amino Acid Score (PDCAAS) of 1, the highest possible score indicating an excellent absorption by the human body.





## **SPECIES OF YEAST USED**

*Saccharomyces cerevisiae:* The most widely used for its high protein content and ease of cultivation. <u>Applications:</u> dietary supplements, meat substitutes (e.g. veggie burgers and sausages).

Saccharomyces boulardii: Used for its probiotic properties.

<u>Applications:</u> probiotic supplements for gut health, fortified foods for improved digestion.

*Cyberlindnera jadinii:* Valued for its high protein content and ability to grow on various substrates. <u>Applications:</u> meat substitutes, flavouring purposes for soups, sauces.

*Kluyveromyces lactis:* Commonly used in dairy-related fermentation processes, this yeast is also investigated for its protein production capabilities and nutritional profile.

<u>Applications:</u> lactase enzyme supplements for lactose-intolerant people, fermented dairy products (e.g. cheese and yoghurt substitutes).

*Pichia pastoris:* Known for its high protein expression and ability to perform post-translational modifications, making it suitable for producing complex proteins.

<u>Applications:</u> recombinant proteins for pharmaceuticals (e.g. insulin), bioengineered food ingredients (e.g. baking and brewing enzymes).

*Yarrowia lipolytica:* Utilized for its high lipid content and potential for producing single-cell proteins, making it a candidate for both protein and oil production.

Applications: single-cell protein products (animal feeds), omega-3 fatty acid supplements.



## A SHORT HISTORY

**1950s-1960s:** Early research highlighted yeast's rapid growth and high protein content. In 1963, FAO/WHO compiled data on its amino-acid content and the biological value of its proteins.

**1970s-1980s:** Industrial-scale production based on optimized strains and improvements in process efficiency characteristics. Regulatory bodies like the FDA (1977) and the European Union approved specific yeast strains *(Saccharomyces cerevisiae)* for use as a protein supplement in food.

**1990s-2000s:** Genetic engineering enhanced yeast strains for higher yield and better nutrition, broadening its benefits beyond protein content.

**2010s-Present:** Focus on sustainable methods and integration of yeast-based proteins into mainstream foods to address food security and environmental issues.



## **PROSPECTS AND INNOVATIONS**

Yeast proteins offer exciting prospects in the alternative protein market:

**Precision Fermentation:** It goes further than traditional fermentation and involves the production of very specific ingredients through the intervention of carefully selected and enhanced micro-organisms, such as yeast. Enzymes, flavors, proteins, vitamins, natural pigments and much more can now be produced this way, with a high level of purity. At the end of the fermentation process, these molecules are separated from the yeast that produced them. This process which has been used for years to produce medicines such as insulin, can now be used to produce key food products such as dairy proteins using less water, land and energy.

**Hybrid Foods:** Development of yeast-based protein blends with plant or animal proteins to improve the texture, flavour, and nutritional profiles of meat alternatives.

**Metabolic Engineering:** Optimisation of yeast metabolic pathways for higher protein yield from non-traditional, sustainable feedstocks like agricultural waste.

**Functional Foods:** Incorporation of yeast proteins into fortified foods with added vitamins, minerals, or probiotics for improved well-being.

**Sustainable Production Systems:** Implementation of closed-loop, zero-waste systems to minimize environmental impact and promote circular economy principles in yeast protein production.







## SUSTAINABLE AGRICULTURE

Yeast is a powerful ally for a more sustainable agriculture, by reducing dependence on chemical products and providing an alternative to the use of antibiotics in livestock.

## **ROLE OF YEAST**

#### **Biostimulation and Bionutrition**

New ranges of products based on yeasts and other microorganisms have been developed to improve plant growth conditions and increase agronomic performance, with three types of actions:

- Stimulation of plants' biological resistance to abiotic stresses (drought, heat...).
- Improving the bioavailability of soil nutrients and thus their uptake by plants.
- Improvement of the rhizosphere or "plant microbiota". This part of the soil close to plant roots is very rich in microorganisms and biological substances and is in continuous interaction with the plants. New yeast-based products enhance soil fertility. They can be applied via irrigation water in field crops and high value crops (vines, market gardening, horticulture, etc.) in both conventional and organic farming.

#### **Biofungicides (Biocontrol):**

Yeasts protect plant health by serving as excellent natural bio-fungicides, managing the balance of pest populations rather than eradicating them. Thanks to their ability to grow quickly in a wide range of environments, they act preventively by spatial and nutrient competition against the occurrence of fungal diseases such as botrytis, powdery mildew and downy mildew. These natural agents are safe for humans and the

### A SHORT HISTORY

environment. Some specific yeast derivatives are elicitors as they stimulate plant defence and prime the plant against fungal disease. Some yeast components can be recognised by special receptors located on plant cell membranes. These receptors trigger intracellular defence pathways. This response results in enhanced synthesis of metabolites which reduce damage and increase resistance to pests and diseases.

#### **Probiotic for livestock:**

Antibiotic resistance, linked to the excessive use of these drugs in livestock, is a serious threat to animal and human health. Yeasts can be used as a probiotic to boost animals' immune systems and gut health, thereby enabling a significant reduction in the use of antibiotics. They act on the digestibility of feed and the reduction of animal stress while optimising growth potential. They are used as feed additives to protect the microbial communities in the digestive tracts of monogastric and ruminant animals to regulate ruminal pH and reduce the risk of acidosis.

#### **Organic fertilizer:**

At the end of the industrial yeast production process, molasses and sugar syrup substrates used to grow yeast are rich in valuable nutrients, that are valorised by yeast producers in the form of feed for livestock or potassium-rich biofertilizers (vinasses).

According to Scopus publications\*, the use of yeast in agriculture originated in 1880. However, for decades, the yeast flora and their diversity have been highly neglected for research purposes regarding agricultural use.

A strong development of scientific research on the uses of yeast for sustainable agriculture emerged in the 2000s, and its business applications have increasingly accelerated since the 2010s.





### **KEY DATA IN AGRICULTURE**

#### Use of yeast in agriculture:

According to the Scopus study<sup>\*</sup>, the use of yeast in agriculture and biological science has exponentially increased since the year 2000, as recorded by increasing interest in research papers on the use of yeast for more sustainable agriculture (over 3000 papers in 2018).

#### Biocontrol<sup>\*</sup>:

- The biocontrol market grew by 40% in value between 2015 and 2019 when, at the same time, conventional products decreased by 19%.
- The share of biocontrol in the crop protection market increased from 3.3 to 5.6% between 2015 and 2019.
- Yeast as a biocontrol agent is used in viticulture, arboriculture, and marginally in vegetable and field crops.

#### Biostimulant\*:

- The biostimulants market grew by +25% in value between 2015 and 2019 when, at the same time, fertilization products decreased by 6%.
- Yeast as a biostimulant is used in field crops, viticulture, arboriculture and vegetable crops.

### **SPECIES OF YEAST USED**

- A wide range of yeast strains are used for more sustainable agricultural purposes: from *Ascomycetes* (e.g., *Saccharomyces, Candida*) to *Basidiomycetes* (e.g., *Filobasidiella, Rhodotorula*).
- In the animal feed industry, *Saccharomyces cerevisiae* is the most commonly used yeast species.
- There are 23 genera of yeasts reported to have plant growth-promoting capabilities and the dominant ones are *Candida spp., Rhodotorula spp., Cryptococcus spp.,* and *Saccharomyces sp.*

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#### **PROSPECTS AND INNOVATIONS**

#### Yeast Microbial Consortia

The application of microbial consortia is a new approach in synthetic biology. Synthetic yeast consortia, simple or complex synthetic mixed cultures, have been used to produce various metabolites. Cooperation between the members of a consortium and cross-feeding can be applied to create stable microbial communication. These consortia can consume a variety of substrates, perform more complex functions, produce metabolites in high titer, rate, and yield (TRY), and show higher stability during industrial fermentations. Given the novelty of this approach, few yeasts are used to build these consortia, including *Saccharomyces cerevisiae, Pichia pastoris, and Yarrowia lipolytica*.

Yeast consortium has important plant growth promotion, biocontrol and pesticide remediation properties. Therefore, these consortia can be used as potential inoculum for boosting agricultural productivity and detoxification of pesticides from soils. These are cost-effective, environment-friendly and socially acceptable, as they reduce chemical and pesticide application.

\* Data from a Kynetec study, a consultancy firm specialising in data, analytics and insights from the agricultural sector.











Yeast is a powerful ally to explore innovative applications and sustainable solutions for bioenergy and in particular biofuels.

Biofuels are renewable energy sources derived from biological material, known as biomass. Used as alternatives to fossil fuels, they can help reduce greenhouse gas emissions and improve the EU's security of fuel supply. At a time of growing food security concerns, integrated food and energy systems (IFES) could be a smart way of ensuring the multi-purpose use of the feedstocks concerned (food, feed and energy), **provided they do not compete with food and feed production**.

## **ROLE OF YEAST**

Yeast, in particular *Saccharomyces cerevisiae*, is essential for bioethanol production, as it ferments sugars and converts them into ethanol.

#### **FIRST-GENERATION BIOETHANOL:**

They use crops such as cereals (mainly corn and wheat), sugar beet and sugar cane, whose simple sugars are transformed into ethanol by fermentation. First-generation biofuels are widely used in the transport sector, in various blending ratios (e.g. E10, E85) or even as a single, clean energy source (E100).

#### **SECOND-GENERATION BIOETHANOL:**

They use lignocellulosic biomass (e.g., agricultural residues, wood chips, and grasses). Their production involves pretreatment and enzymatic hydrolysis to break down cellulose and hemicellulose into sugars that will further be fermented by yeast. Second-generation bioethanol are more sustainable as they use non-food biomass. However, their development is slowed by higher costs in the production process.



## FORMAT OF YEAST USED

At the beginning, yeasts were used in a dry format. However, recently plants have started to use a liquid form of yeast that have advantages because they are already hydrated. Some have been stabilised for longer shelf life.



### A SHORT HISTORY

The development of biofuel started in the late 20th century. In the EU, biofuel production and consumption truly soared from the 2010s:

- **1970s Oil Crises:** The search for renewable energy sources intensified, leading to the use of yeast for bioethanol production from crops like corn and sugarcane.
- Late 20th century: Advances in genetic engineering began to enhance yeast's efficiency and expand its applications in biofuel production. However, nowadays, most yeast used for bioethanol production in the European union are still non-GMOs.
- Early 21st century: Yeasts were engineered to produce biodiesel, biogas, and advanced biofuels, including jet fuel, from various biomass sources.



## **KEY DATA**

- Ethanol reduces greenhouse gas emissions by 70-90% (excluding land-use changes) due to its sulphur-free composition and higher-octane rating than petrol.
- Biomass accounted for 59% of renewable energy consumption in the EU in 2021, with the industry sector consuming 21.1 million tonnes of oil equivalent (Mtoe) of biomass.

## **PROSPECTS AND INNOVATIONS**

With the ambition to decarbonize all transport in the EU as shown by the recently adopted Fit for 55 Package, demand for sustainable alternatives to fossil fuels is expected to grow dramatically by 2030. Optimizing yeast-based bioproduction processes is therefore essential. Here are the main prospects:

**1.** Improve the bioethanol production process and the efficiency of fermentation during which the yeast converts the substrate into bioethanol.

**2. Biodiesel production:** this promising research has not yet led to industrialization. Some yeasts can accumulate high levels of lipids (oils) which can be converted to biodiesel by transesterification. Oleaginous yeasts, such as *Yarrowia lipolytica*, *Cryptococcus curvatus* and *Rhodotorula glutinis*, can use a variety of raw materials, including waste, agricultural residues and industrial by-products.

**3. Biogas Production (through Anaerobic Digestion):** Yeast can improve the efficiency and stability of microbial communities in anaerobic digesters, where organic matter is broken down by microorganisms to produce biogas (a mixture of methane and CO<sub>2</sub>).

**4. Biohydrogen Production (through Dark Fermentation):** Yeasts, in combination with bacteria, can be used in dark fermentation processes to produce hydrogen gas from organic substrates. Yeast is used to break down complex substrates, making them available for hydrogen-producing bacteria.

**5. Butanol Production(through A-B Fermentation):** Genetically engineered yeasts can be used to produce butanol, with higher energy content and lower volatility than ethanol. Yeasts can be engineered to produce butanol directly from sugars.

**6. Alcohol to jet advanced fuels:** Yeasts can be engineered to produce alcohols, which are then catalytically upgraded into jet fuel by processes like alcohol-to-jet (ATJ) conversion. The alcohols undergo dehydration, oligomerization, and hydrogenation to form high-density hydrocarbons suitable for aviation fuel. Ethanol can hence be used as a platform molecule to produce sustainable aviation fuels (SAF).

**7. Production of 3rd generation bioethanol from algal biomass:** A promising process that does not compete with agricultural land, and also captures CO<sub>2</sub> through photosynthesis. Its cost remains high due to the complex structure of algae sugars, requiring pre-treatment and the use of specialized yeasts.

**8. 4th generation bioethanol:** Its principle is to use genetically modified yeasts and bacteria to produce ethanol directly from CO<sub>2</sub> or plastic waste! This technique, which is still under development, would therefore make it possible to produce a biofuel while capturing CO<sub>2</sub> during the production process.



SUGAR BEET SUGAR CAN WHEAT CORN



NON-EDIBLE BIOMASS WOOD STRAW GRASS WASTE



ALGAL BIOMASS MACROALGAE MICROALGAE



BREAKTHROUGH PYROLYSIS SOLAR-TO-FUEL ENGINEERED ALGAE GASIFICATION









## POLLUTION CONTROL AND WASTE TREATMENT

Yeast offers versatile and environmentally friendly technologies, paving the way for a cleaner future.



## **ROLE OF YEAST**

The use of yeast in the areas of pollution control and waste treatment is largely attributed to its metabolic capabilities, resilience in various environmental conditions, its diversity, and its ability to be genetically modified for enhanced performance.

#### **BIOREMEDIATION AND POLLUTANT DEGRADATION**

- Due to their ability to metabolize a wide range of organic pollutants, yeasts (particularly *Saccharomyces cerevisiae*) can degrade hydrocarbons, phenols, and other toxic organic compounds through their enzymatic pathways. For instance, *Candida tropicalis* has shown efficacy in degrading phenol, with removal efficiencies reaching up to 98% under optimal conditions.
- Additionally, yeasts can be used to remediate heavy metals through biosorption, a process where metal ions are sequestered by the cell wall components. For instance, *Saccharomyces cerevisiae* can absorb significant amounts of cadmium and lead from aqueous solutions, for heavy metal bioremediation and potential recycling of some valuable heavy metals.

#### WASTEWATER TREATMENT

- Yeasts can be integrated into treatments of industrial effluents. For example, *Saccharomyces cerevisiae* has been used in anaerobic digesters to improve the degradation of brewery wastewater, resulting in increased biogas production and reduced chemical oxygen demand (COD) levels.
- Moreover, the use of yeast in constructed wetlands and biofilters has been shown to effectively remove nitrogen and phosphorus from wastewater, thereby preventing its eutrophication.

#### GENETIC ENGINEERING FOR ENHANCED PERFORMANCE

- Thanks to genetic engineering, yeasts can express foreign genes that enhance their pollutant degradation capabilities. For instance, recombinant *Pichia pastoris* expressing organophosphorus hydrolase can degrade organophosphate pesticides, achieving degradation rates up to 95%.
- CRISPR-Cas9 technology has been employed to create yeast strains with enhanced tolerance to toxic environments and improved metabolic pathways for the efficient breakdown of pollutants.

#### ECONOMIC AND ENVIRONMENTAL BENEFITS

The use of yeast in pollution control and waste treatment offers several benefits:

- Yeasts are cost-effective due to their rapid growth rates.
- Most yeast species grow under anoxic conditions, with less oxygenation costs.
- In some cases, the yeast biomass generated from the waste treatment can be separated and used in fertilizers or even animal feeds, due to its high protein and vitamin content.
- These processes are more sustainable than conventional chemical treatments that are more costly and environmentally damaging with harmful byproducts.





## **SPECIES OF YEAST USED**

- Oxidative yeasts: Species like Candida tropicalis, Pichia anomala, and Hansenula polymorpha demonstrate robust abilities to degrade a wide range of pollutants, including:
- Hydrocarbons: Candida guilliermondii can remove up to 90% of total petroleum hydrocarbons.
- <u>Phenolics</u>: *Rhodotorula mucilaginosa* exhibits high efficiency in degrading phenolic compounds, a common class of industrial pollutants.
- **Mixed cultures:** Combining yeast with other microorganisms like bacteria or algae can create synergistic effects. For example, yeast-algae consortia can simultaneously remove organic matter and nutrients from wastewater.

## PROCESS

- Yeast treatment typically involves cultivating a chosen yeast strain in a bioreactor containing the polluted wastewater. These yeasts utilize the pollutants as a carbon and energy source for their growth. The process can be aerobic (requiring oxygen) or anaerobic (without oxygen), depending on the specific yeast and contaminant. The treated wastewater undergoes further processing to remove the yeast biomass before discharge.
- Yeast-based bioremediation typically involves the following steps.
- Inoculation: Yeast strains are introduced to the polluted medium (e.g., wastewater, soil).
- Nutrient addition: Nutrients necessary for yeast growth, such as nitrogen and phosphorus, are added to the medium.
- <u>Aeration</u>: Oxygen is supplied to facilitate aerobic metabolic processes.
- <u>Fermentation and degradation</u>: Yeast metabolizes pollutants through fermentation, breaking down complex organic compounds into simpler, less harmful substances.
- <u>Separation</u>: Treated water or soil is separated from yeast biomass, which can be further processed or used as biofuel.



#### A SHORT HISTORY

- In the 1970s, pioneering work by Yoshizawa in Japan demonstrated the effectiveness of yeast in wastewater treatment. Since then, these technologies have been increasingly used.
- In 2022, the Marc Regnier Prize was awarded to the Sublimus project for its sustainable methods of extracting rare and precious metals from urban wastewater, particularly activated sludge.

#### **KEY DATA**

- **Organic matter removal:** Yeast can achieve high removal rates of organic matter, as measured by Chemical Oxygen Demand (COD), with report of reductions exceeding 95% for specific wastewater.
- Heavy metal bioaccumulation: Certain yeast strains can accumulate heavy metals like chromium and lead, offering a bioremediation strategy for metal-contaminated environments.
- **Organic Pollutant Degradation:** Candida utilis has been shown to degrade up to 90% of phenol and other toxic organic compounds in industrial effluents.
- Wastewater Treatment Efficiency: Yeast-based treatments can reduce biochemical oxygen demand (BOD) and chemical oxygen demand (COD) by up to 85%, making the water safe for discharge or reuse.

### **PROSPECTS AND INNOVATIONS**

- **Engineered yeasts:** Researchers are exploring genetic modification of yeast strains to enhance their pollutant degradation capabilities and tolerate extreme conditions.
- **Biodiesel production:** Yeast can convert the **lipids present in wastewater** into biodiesel, offering a sustainable approach to waste treatment and fuel generation.
- **Bioaugmentation:** Developing microbial consortia, including yeast and bacteria, to target a broader range of contaminants.











## **PRECISION FERMENTATION**

Yeast plays a crucial role in the burgeoning field of precision fermentation, an old but revolutionary biological technique to produce valuable biomolecules with smaller environmental footprints.

## **ROLE OF YEAST**

In precision fermentation, **yeast acts as a biological factory** via the controlled cultivation of enhanced modified yeast cells to produce specific compounds of interest in the food & feed sectors such as protein, amino acid, fatty acid, flavor molecule, vitamin, pigments, enzymes or even in the pharmaceutical sector. At the end of the cultivation phase, the desired resulting compounds are filtered out, separating them from the yeast that produced them.

Several microorganisms can be used for precision fermentation including algae, bacteria or fungi but yeast offers several advantages for this purpose: :

- Rapid growth rate in comparison to other microorganisms.
- Efficient fermentation: Yeast readily consumes sugars as its primary energy source, efficiently converting them into desired products during fermentation.
- Well-characterized genetics and metabolism: Scientists have a deep understanding of yeast's metabolic pathways, enabling them to optimize production.
- **Predictable and scalable:** Yeast growth and fermentation processes are well-established, making it a predictable and reliable platform, enabling high-volume production of targeted molecules.
- Safe and generally regarded as safe (GRAS): Many yeast species have a long history of safe use in food and beverage production.
- **Cost-effectiveness:** With advancements in fermentation technology and strain optimization, the cost of producing biomolecules through yeast is becoming increasingly competitive.

Depending on the desired end-product, precision fermentation can offer many benefits: elimination of the need for animal extraction (like for insulin or rennet), shift from a chemical synthesis to a biological synthesis. This bio-manufacturing process is more resource efficient, releases fewer greenhouse gas emissions and has less environmental impacts.

Yeast as a biological factory has a wide range of applications in precision fermentation:

- Food and drinks: Production of animal-free dairy proteins (casein and whey), meat substitutes, and specialty ingredients like flavors and sweeteners<sup>1</sup>.
- Specialty chemicals: Production of fragances, pigments, and enzymes for food, feed or textiles.
- Pharmaceuticals: Manufacture of recombinant insulin, vaccines, and therapeutic proteins.
- **Biofuels and biochemicals:** Development of sustainable biofuels and biochemicals to reduce reliance on fossil fuels.

<sup>1</sup> L.J.G. Hoppenreijs, A. Annibal, G.J.C. Vreeke, R.M. Boom, J.K. Keppler, Food proteins from yeast-based precision fermentation: Simple purification of recombinant B-lactoglobulin using polyphosphate, Food Research International, 2024, <u>https://doi.org/10.1016/j.foodres.2023.113801</u>





## **SPECIES OF YEAST USED**

- **Saccharomyces cerevisiae** (Baker's yeast), the heart of brewing and baking, is a popular choice due to its established industrial fermentation protocols and vast scientific knowledge base. <u>Applications:</u> Production of human insulin, essential amino acids, and flavors like vanillin.
- **Pichia pastoris (Komagataella pastoris)** is gaining traction due to its ability to produce complex proteins with proper folding and glycosylation patterns.

<u>Applications:</u> Production of biopharmaceuticals like interferon alpha (used to treat some cancers) and components of spider silk for material applications.

- Other emerging yeasts are being explored for their specific metabolic strengths, such as:
- Yarrowia lipolytica enables high lipid production.
- *Kluyveromyces lactis* has a particular tolerance to harsh fermentation conditions.
- Applications: To explore the production of alternative fats and biofuels.



## **A SHORT HISTORY**

Precision fermentation has been used for over 30 years. One of the earliest examples was the production of insulin in the 1970s using modified *S.Cerevisiae*, providing a solution for diabetic patients without having to extract insulin from cow or pig pancreases. The advent of genetic engineering techniques in the late 20th century paved the way for an increased use of precision fermentation with yeast.

Since then, the field has witnessed rapid advancements, with the production of a large range of highvalue products like essential amino acids, heme (the iron-containing molecule in red blood cells), enzymes, and even animal proteins such as milk proteins (caseins or whey).

## **KEY DATA**

- The European precision fermentation ingredients market could experience a market growth of 43,6% CAGR (Compound Annual Growth Rate) between 2023 and 2030<sup>2</sup>.
- Europe holds the largest market share of 40% of the precision fermentation market<sup>3</sup>:
  - By application, the dairy alternative segment contributed almost 60% of revenue share.
  - By micro-organism, yeast could represent the highest market share across 2023-2032.
- Precision fermentation significantly reduces the environmental footprint of food ingredient production. For example, yeast-based precision fermentation whey protein is more sustainable than **traditional milk-derived whey protein<sup>4</sup>**:
- Using precision fermentation process to produce 1 kg of whey protein reduces greenhouse gas emissions by 91%-97%, as compared to 1kg of conventional dairy protein<sup>5</sup>.
- Precision fermentation cuts blue water<sup>6</sup> consumption by 96%-99%<sup>5</sup>.
- Energy consumption is 30%-60% lower with precision fermentation<sup>5</sup>.
- Precision fermentation requires up to 99% less land compared to conventional livestock farming<sup>5</sup>.



### **PROSPECTS AND INNOVATIONS**

The future of precision fermentation with yeast is bright, with some exciting areas of development:

- Machine learning-aided strain design: Utilizing machine learning algorithms to accelerate the design and optimization of yeast strains for specific target molecules.
- **Consolidated bioprocessing:** Integrating multiple steps of the fermentation process for improved efficiency and reduced production costs.
- Novel yeast platforms: Exploring alternative yeast species with unique metabolic capabilities to expand the range of biomolecules that can be produced.
- Data-driven and Al-based biomanufacturing: Developing these cutting-edge technologies could reduce cost and time-to-market.

2. KBV Research (2023). Europe Precision Fermentation Ingredients Market Size, 2030. KBV Research.

Available at: <u>https://www.kbvresearch.com/europe-precision-fermentation-ingredients-market/</u> 3. Precedence Research (2023). Precision Fermentation Market Size, Trends, Growth, Report 2032.

Available at: https://www.precedenceresearch.com/precision-fermentation-market

4. FAO (UN Food and Agriculture Organization) (2013). A global life cycle assessment Greenhouse gas emissions from ruminant supply chains. Available at: <u>https://www.fao.org/4/i3461e/i3461e.pdf</u>

5. Science Media Centre. (2024). 'Precision fermentation' and the future of NZ food - Expert Reaction. Available at: <a href="https://www.sciencemediacentre.co.nz/2024/05/16/precision-fermentation-and-the-future-of-nz-food-expert-reaction/">https://www.sciencemediacentre.co.nz/2024/05/16/precision-fermentation-and-the-future-of-nz-food-expert-reaction/</a>.

6. "Blue" water is water that transfers rapidly into rivers, lakes and groundwater. Blue and green water. INRAE Silva Joint Research Unit. Available at: <u>https://appgeodb.nancy.inra.fr/biljou/en/fiche/eau-verte-eau-bleue</u>







# WHO WE ARE?

Founded in 1959, COFALEC is the European Confederation of Yeast Producers, with 47 members representing almost all the European industry, a world leader in its sector.









Many thanks to all our experts from COFALEC members, and in particular from AB MAURI, JASTBOLAGET, LALLEMAND and LESAFFRE, for their invaluable help in producing this brochure, which aims to reveal the wide variety of yeast applications.

We hope that this document will enable as many people as possible to discover the extraordinary powers of yeast in the service of a more sustainable world!

VINCENT SAINGIER, President of COFALEC

in

DIANE DORÉ, Secretary General



9 Boulevard Malesherbes, 75008 Paris contact@cofalec.com



