Fermentation's great promise

Much of our food, including sauerkraut, chocolate and yoghurt, is the product of fermentation: it has already been digested by bacteria, fungi or yeasts. But these micro-organisms can do a lot more than that. Researchers are working in specialized labs on new tempeh, chemicals and biofuel.

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PHOTO VERSE BEELDWAREN

'The possibilities fermentation offers are endless,' says Professor Eddy Smid of the Wageningen Laboratory for Food Microbiology. He works with fermentation to develop new, healthy foods with a unique taste, better nutritional value or a longer shelf life. Micro-organisms are key to this process. The principle is not new: people have been making use of bacteria, fungi and yeasts to process foods for thousands of years (see inset). But scientists are still finding new applications.
Smid, for example, is always looking for the best match between various micro-organisms. 'So we came up with Tinder for Microbes. That is a resource for predicting which micro-organisms combine well together, using the microbe's profile, such as its preferred substrate feed and the substances the organism forms.' >

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Fermenting lupin to make tempeh and then making propionic acid bacteria form vitamin B12 produces a sustainable meat substitute.

> **‘Lupin tempeh is rich in protein *and* vitamin B12’**

A well-known match is the one between lactic acid and propionic acid bacteria, which combine to make the famous Emmenthaler cheese. The lactic acid formed by lactic acid bacteria is the perfect diet for the propionic acid bacteria, which in turn form flavour and aroma components that give the cheese a sweet flavour. ‘Those propionic acid bacteria also produce vitamins, including vitamin B12,’ says Smid. ‘We’ve been using that characteristic to develop a new tempeh with this essential vitamin. Normally, only animal products contain vitamin B12.’

Tempeh, a meat substitute made of fermented soya beans, is made by the *Rhizopus* fungus, which produces lactic acid. Researchers from Smid’s team added propionic acid bacteria, which enriched the tempeh with vitamin B12. This new meat substitute is of interest to vegans, who don’t eat animal products and therefore don’t get any B12.

LUPIN TEMPEH

Because the production of soya often goes hand in hand with large-scale deforestation, Smid sought a sustainable alternative to soya beans. He found one a few years ago in the form of lupin, a European crop with a nutritional value comparable to that of soya. He could ferment lupin to get lupin tempeh using the same fungus that is used with soya, after which fermentation with propionic acid bacteria formed vitamin B12. Smid: ‘That gave us a sustainable product that was rich in protein and vitamin B12.’

One of the interested companies, and one which also participates in Smid’s research, is the Dutch ‘Vegetarian Butcher’. ‘Fermented products may well be even more interesting as meat substitutes than the plant-based ingredients currently being used,’ says Niko Koffeman, co-founder of the company. ‘Fermentation leads to natural, unprocessed products. If they also contain a natural source of vitamin B12, we’ve got hold of something unique.’ Using regionally grown lupin makes the product even more sustainable. Koffeman hopes to have the new product on the supermarket shelves within a few years.

Filip Oosterlinck, product manager at De Hobbit, a Belgian producer of natural, organic meat substitutes, is collaborating in Smid’s research too. The vast majority of the conventional meat substitutes produced by the company, among them tempeh and natto, are fermented and inspired by Far Eastern cuisine. ‘Those fermentation processes are thousands of years old. They provide complete, safe and natural foods without additives,' says Oosterlinck. 'Given fermentation's potential for so many new products, I expect we are going to be marketing more and more fermented meat substitutes based on new ingredients such as chickpeas or black beans.'

MAKING CHEMICALS

Applications of fermentation outside the food sector are relatively new. Jeroen Hugenholtz, a researcher at Wageningen Food and Biobased Research, focuses on making chemicals out of waste streams. He studied how processes can be steered and optimized through the choice of microorganisms, ingredients and fermentation conditions, such as temperature and acidity (pH). 'We want our research on the efficient fermentation of waste streams to give companies new opportunities and a greater understanding of cost-effective chemical production,' says Hugenholtz. 'By using waste streams such as organic waste, it is theoretically possible to cut costs by at least 50 per cent because the raw material is cheap.' One example of successful conversion of a waste stream into chemicals is the production of bioplastic from organic waste through a process in which various bacteria produce fatty acids under anaerobic conditions. Subsequently the *Pseudomonas* bacterium converts the fatty acids into polyhydroxyalkanoate (PHA), one of the building blocks for bioplastic.

Different species of microbe produce different materials from organic waste: lactic acid bacteria ferment sugars into lactic acid, the basis for poly-lactic acid (PLA), another type of bioplastic. 'We monitor and steer these kinds of conversions by making use of one or two bacteria and then manipulating the pH and the oxygen supply,' explains Hugenholtz.

A big advantage of microbes, in Hugenholtz's view, is their capacity to convert contaminated waste streams, which is not possible using chemical methods, for which the raw materials need to be very pure. Fermentation is also more sustainable and cleaner: no solvents, acids, leaching or high temperatures are required, as they are for chemical processes. But microorganisms alone will not suffice, thinks Hugenholtz, who sees fermentation and traditional chemical methods as complementary. 'It is often efficient to finish off >

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PHOTO GUY ACKERMANS

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FERMENTATION FOR MORE THAN JUST FOOD

Every day we eat products on which fungi, yeasts and bacteria have left their mark. Sauerkraut, beer, wine, yoghurt and cheese are obvious fermented foodstuffs, but so are coffee, cocoa and Parma ham. In all these products, bacteria, yeasts and fungi have converted substances into aromas or antibacterial components such as bacteriocins, which counteract specific pathogens. Manufacturers of raw-milk cheeses add bacteriocin-forming microbes to their products during fermentation, to halt the growth of pathogenic Listeria bacteria. Fermentation is important outside the food industry too. Yeasts and bacteria can produce antibiotics, insulin and other medicines, as well as chemicals and fuels. This becomes especially interesting if it is done using waste streams.

Waste streams can also be a source of products that are useful in foodstuffs. Through fermentation, a waste stream that contains relatively large numbers of proteins can provide the food industry with many useful amino acids and flavour components derived from them.

‘Companies are interested in sustainable alternatives to palm oil’

PHOTO GUY ACKERMANS

JACCO VAN HAVEREN
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the processing of fermented products made by fermentation using chemical processes. For example, we convert lactic acids formed by fermentation into polylactic acids, PLA, chemically.’
There are millions of species of bacteria and fungi which are capable of making countless food ingredients, chemicals or the building blocks for them out of waste streams. The yeast *Pichia pastoris* makes polypeptides – short protein chains of just a dozen or so amino acids – out of sugars. Some of these small proteins could replace certain ingredients in cosmetics. It will take a few years, however, before the first applications appear on the market. Jacco van Haveren, programme manager at Biobased Chemicals & Fuels in Wageningen: ‘The development could take years, but if it works well, it could go quickly.’
Clostridium is another example. This bacterium produces alcohols, aldehydes and fatty acids, substances that can be used to produce biofuels, solvents, cosmetics and pharmaceuticals. Some fatty acids are also suitable for use as preservatives. Both the food and the cosmetic industries currently use the preservative benzoate, a substance that has been linked to hyperactivity in children, and which sometimes gets converted into toxic benzene. The Wageningen study focuses on developing safe alternatives using biobased fatty acids.
‘One thing producers are very interested in, for both cosmetics and food products, is sustainable alternatives to palm oil,’ says Van Haveren. ‘Fatty acids formed by fermentation provide the basis for several oils that could replace palm oil.’

AVIATION FUEL

The research on applications for biofuel is very promising too. ‘The *Clostridium* bacterium makes a mix of acetone, ethanol and most of all, butanol out of potato peel,’ says Van Haveren. ‘Butanol is a building block for biofuel and the most interesting option, commercially. We are now trying to steer the process so that even more butanol is formed.’ If you use traditional chemistry to link three butanol molecules, you get dodecane, which has potential as aviation fuel. ‘Our goal is to make the production of aviation fuel out of waste streams consisting

REDUCING CLIMATE EMISSIONS IN THE CHEMICAL INDUSTRY

Fermentation is one of the technologies used to produce biobased chemicals. By producing these chemicals and using milder processes, the chemical industry is helping reduce its climate emissions. Wageningen makes a contribution as a knowledge partner.
Biobased alternatives are possible for all basic chemicals currently produced from oil. These might be substances with the same structure as petrochemicals (known as drop-in substances), or new chemicals with new and unique characteristics such as biodegradability or non-toxicity, which can be used in packaging or cosmetics. Wageningen Food & Biobased Research is doing research in collaboration with the industry on developing biobased chemicals which are produced sustainably and have improved characteristics. That is good for the environment as well as for the competitiveness of companies.
‘Biobased drop-in products will make a significant contribution to reducing CO_2 emissions,’ says Jacco van Haveren, programme manager for Biobased Chemicals and Fuels. But creating new substances can also resolve the undesirable aspects of currently available chemicals.

of potato peel and other wet biomass economically viable within 10 years,' states Van Haveren. 'But we do need financiers and funds for that, to invest in a factory.'
Misha Valk, head of Future Fuels at the Dutch company SkyNRG, a global leader in sustainable aviation fuel, sees an important role for fermentation of waste streams in making air travel more sustainable. 'At present, the commercial production of biokerosene from used vegetable oil or animal fats is possible,' he says. 'A new factory in Delfzijl will start producing 100,000 tons of sustainable kerosene a year from 2022. But there is nowhere near enough used oil and fats to meet the demand. So in the near future fermentation will definitely be an important technology for making kerosene out of waste streams commercially.'

BOTTLENECKS

Chemicals and materials obtained through fermentation currently cover only a fraction of the total demand. 'Only five to seven per cent of all chemicals are biobased,' states Van Haveren. 'And only five to ten per cent of those are obtained using fermentation.' One of the bottlenecks for large-scale production is that many processes are still in the development phase. Research on upscaling of processes developed in the laboratory is also expensive.
One positive exception is the production of bioplastic based on PLA (polylactic acid), almost all of which is obtained by fermenting sugars in agricultural products and waste streams. But in spite of the large-scale commercial production of 150,000 to 200,000 tons a year, PLA makes up only a tiny proportion of the 300 million tons of plastics produced globally every year. There is growth, though, says Van Haveren. As the supply of cheap oil slowly but surely runs out, biobased chemicals can compete better with petrochemicals. 'At the moment, PLA plastic is still a little more expensive than conventional plastic, but if we start producing it on an even larger scale, it will compete with oil-based plastics pricewise.'
Van Haveren sees concern about the climate, dwindling oil supplies and the toxicity of some traditional chemicals, such as the preservative benzoate, as an additional incentive to focus on manufacturing chemical building blocks, with a key role for fermentation. This will produce safer and more sustainable products, thanks to a tradition going back thousands of years of making micro-organisms work for us. ■

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PHOTO ALAMY

Clostridium bacteria make butanol out of potato peel. Linking three butanol molecules using traditional chemistry produces dodecane, which has potential as aviation fuel.

'Producing aviation fuel from biomass waste will be economically viable within 10 years'