VII
Microorganisms in Industry

18 Industrial and Food Microbiology

Many aspects of our everyday lives are influenced in some way by microorganisms. In previous chapters we have noted how they can cause infectious diseases as well as providing a means of curing them, and the vital role they play in the environment. In addition, they are responsible for the production of much of what we eat and drink, synthesise industrially useful chemicals and can even extract precious metals from the earth (Table 18.1). In this chapter we shall look at some of the ways in which the activities of microorganisms have been harnessed for the benefit of humans, and developed on an industrial scale. The first applications of biotechnology many thousands of years ago were in the production of food and drink, so it is here that we shall begin our survey.

18.1 Microorganisms and food

To the general public, the association of microorganisms and food conjures up negative images of rotten fruit or food poisoning. On further reflection, some people may recall that yeast is involved in bread and beer production, but how many realise that microorganisms play a part in the manufacture of soy sauce, pepperoni and even chocolate? In the following we shall look at the contribution of microorganisms to the contents of our shopping baskets before considering one of the negative associations referred to above, the microbial spoilage of food.

The production of foodstuffs as a result of microbial fermentation reactions predates recorded history. The accidental discovery that such foods were less susceptible to spoilage than fresh foods must have made them an attractive proposition to people in those far-off days. Of course, until relatively recent

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Table 18.1 Some applications of microorganisms

Food products
Alcoholic drinks
Dairy products
Bread
Vinegar
Pickled foods
Mushrooms
Single-cell protein
Products from microorganisms
Enzymes
Amino acids
Vitamins
Antibiotics
Vaccines
Citric acid
Mining industries
Metal extraction
Desulphurisation of coal
Alternative fuels
Ethanol
Methane
Hydrogen
Agriculture
Microbial pesticides
Environment
Bioremediation
Sewage treatment/water purification
Insect control
Biosensors

times, nothing was known of the part played by microorganisms, so the production of beer, cheese and vinegar would not have entailed the carefully controlled processes in use today. Indeed, it was only with the development of isolation techniques towards the end of the nineteenth century (recall Chapter 1), that it became possible to use pure cultures of microorganisms in food production for the first time. The fermentation of foodstuffs, hitherto an art, became a science.

18.1.1 Alcoholic fermentations

There is evidence that alcoholic drinks including beer and wine were being produced thousands of years before the Christian era, making them among

the earliest known examples of the exploitation of microorganisms by humans. Ethanol results from the fermentation process because the conversion of sugar to carbon dioxide and water is incomplete:

$$C_6H_{12}O_6 \rightarrow 2CH_3CH_2OH + 2CO_2$$

Wine can be made from almost any fruit juice with a high sugar content. The vast majority of commercially produced wines, however, derive from the fermentation of the sugar present in grapes (Figure 18.1). Such fermentation reactions may be initiated by yeasts found naturally on the grape skin; however, the results of such fermentations are erratic and may be unpalatable. In commercial winemaking the *must* (juice) resulting from the crushed grapes is treated with sulphur dioxide to kill the natural microflora, and then inoculated with the yeast *Saccharomyces cerevisiae*, variety *ellipsoideus*. Specially

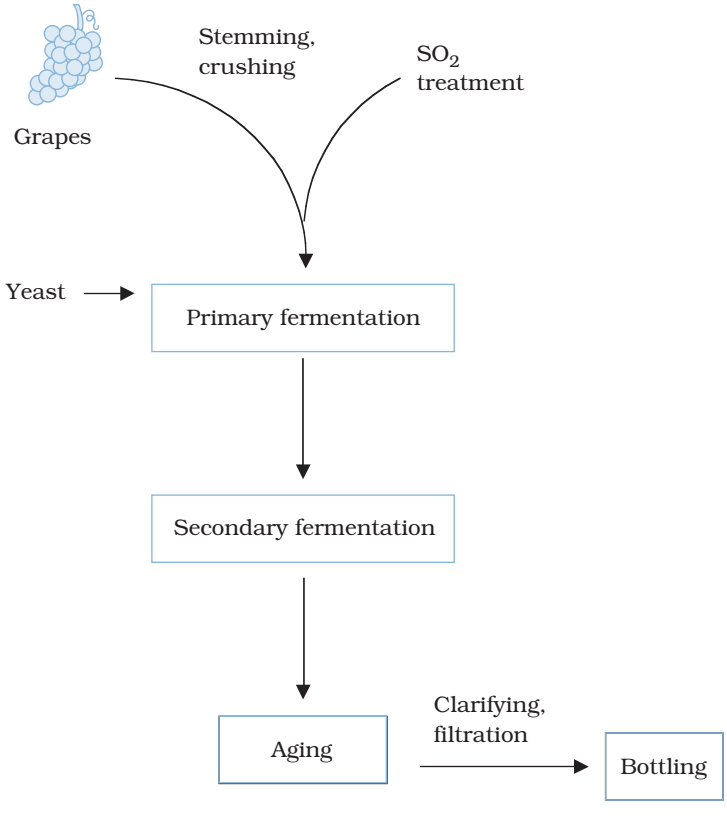

Figure 18.1 Wine production. The figure outlines the essential steps involved in the production of red wine. White wine production differs in certain details, but shares the main steps of crushing the grapes, fermenting their sugar content into alcohol and aging to allow flavour development.

developed strains are used, which produce a higher percentage of alcohol (ethanol) than naturally occurring yeasts. Fermentation proceeds for a few days at a temperature of 22–27°C for red wines (lower for whites), after which the wine is separated from the skins by pressing. This is followed by *ageing* in oak barrels, a process that may last several months, and during which the flavour develops. *Malolactic* fermentation is a bacterial secondary fermentation carried out on certain types of wine. Malic acid, which has a sharp taste, is converted to the milder lactic acid, imparting smoothness to the wine. A secondary product of malolactic fermentation is diacetyl, which imparts a 'buttery' flavour to the wine.

Only black grapes (including the skins) are used in the production of red wines; white wines may use white grapes or black ones with the skins removed. When all or most of the sugar has been converted to alcohol, dry wines result; when some sugar remains, we get a sweeter wine. Most wines have an alcohol content of around 10–12%. For sparkling wines, an additional fermentation is carried out in the bottle to generate the bubbles of CO_2 characteristic of such wines.

Spirits such as brandy and rum result from the products of a fermentation process being concentrated by *distillation*. This gives a much higher alcohol content than that of wines.

Beer is produced by the fermentation of barley grain. The procedure varies according to the type of beer, but follows a series of clearly defined steps (Figure 18.2). Grain, unlike grapes, contains no sugar to serve as a substrate for the yeast, so before fermentation can begin it is soaked in water and allowed to germinate. This stimulates the production of the enzymes necessary for the conversion of starch to maltose (*'malting'*). An additional source of starch may be introduced during the next stage, *mashing*, in which the grains are ground up in warm water and further digestion takes place. The liquid phase, or *wort*, is drained off and boiled; this has the effect of inactivating the enzymes, precipitating proteins and killing off any microorganisms. It is at this point that *hops* are added. They impart flavour and colour to the finished product and also possess antimicrobial properties, thereby helping to prevent contamination. In the next stage, the wort is filtered and transferred to the fermentation vessel where yeast is introduced.

Two species of yeast are commonly used in the brewing process, both belonging to the genus *Saccharomyces*. *S. cerevisiae* is mainly used in the production of darker beers such as traditional English ales and stouts, whereas *S. carlsbergensis* (no prizes for guessing where this one was developed!) gives lighter coloured, less cloudy, lager-type beers. Cells of *S. cerevisiae* are carried to the surface of the fermentation by carbon dioxide bubbles (top fermenters), while *S. carlsbergensis* cells form a sediment at the bottom (bottom fermenters).

Fermentation takes about a week to complete, at a temperature appropriate for each type of yeast (*S. carlsbergensis* prefers somewhat lower

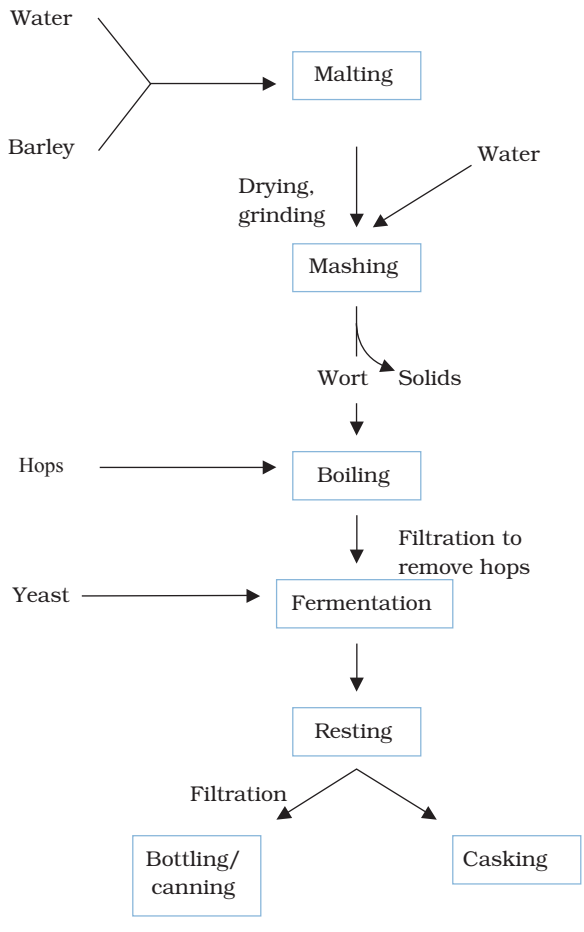

Figure 18.2 Beer production. The early stages serve to convert the carbohydrate present in the grain into a form that can be fermented by the yeast.

temperatures than *S. cerevisiae*). Following fermentation, the beer is allowed to age or 'rest' for some months in the cold. Beers destined for canning or bottling are filtered to remove remaining microorganisms. 'Spent' yeast may be dried and used as an animal food supplement.

Beers typically have an alcohol content of around 4%. Small amounts of other secondary products such as amyl alcohol and acetic acid are also produced, and contribute to the beer's flavour. 'Light', or low-carbohydrate beers are produced by reducing the levels of complex carbohydrates. The yeasts do not possess the enzymes necessary to cope with these branched molecules, so a supplement of debranching enzymes may be added to aid their breakdown.

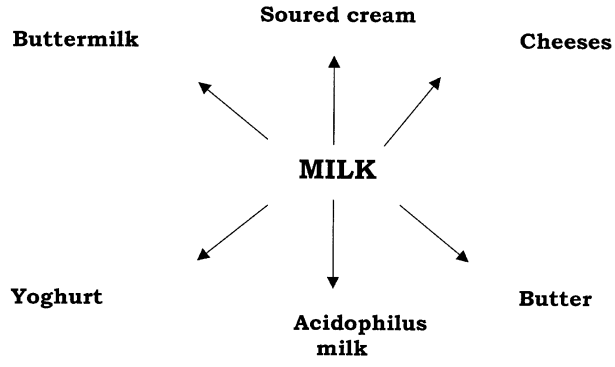

Figure 18.3 Fermented dairy products. Fermentation is initiated by the inoculation of a starter culture of lactic acid bacteria to convert lactose to lactic acid. Heterolactic fermenters such as *Leuconostoc* are added when aromatic flavouring compounds such as diacetyl are required.

18.1.2 Dairy products

Milk can be fermented to produce a variety of products, including butter, yoghurt and cheese (Figure 18.3). In each case, acid produced by the action of lactic acid bacteria causes coagulation or *curdling* of the milk proteins; in cheesemaking, the addition of rennin results in the separation of the semi-solid *curd* from the liquid *whey*. The subsequent steps in the process depend on the specific type of cheese (Table 18.2). Following separation, the curd of most cheeses is pressed and shaped, removing excess liquid and firming the texture. During the *ripening* process, salt is often added, and flavour develops due to continuing microbial action on the protein and fat components of the cheese. The length of the ripening period varies from a month to more than a year according to type, with the harder cheeses requiring longer periods. In some cases a fresh inoculation of microorganisms is made at this point, such as the addition of *Penicillium* spores to Camembert and Brie. For Emmental,

Table 18.2 Types of cheese. Cheeses are classified according to their texture. Unripened cheeses are those that have not undergone the aging or ripening process, during which additional flavours develop

Soft	Semi-soft	Semi-hard	Hard
Unripened: Mozzarella Cottage	Roquefort Stilton	Cheddar Emmental	Parmesan Pecorino
Ripened: Camembert Brie			

Propionibacterium freudenreichii is added, leading to the generation of carbon dioxide bubbles that form the holes characteristic of this type of cheese.

Yoghurt is another milk derivative. Thickened milk is exposed to the action of two types of bacteria, *Streptococcus salivarius* ssp. *thermophilus* and *Lactobacillus delbrueckii* ssp. *bulgaricus*, both of which ferment lactose present in milk into lactic acid. In addition, the latter contributes aromatics responsible for imparting flavour to the yoghurt.

Other dairy products such as soured cream and buttermilk are also produced by means of the fermentative properties of species of streptococci and lactobacilli.

18.1.3 Bread

The biological agent responsible for bread production is yeast. In fact, baker's yeast and brewer's yeast are just different strains of the same species, *Saccharomyces cerevisiae*. In breadmaking, aerobic rather than anaerobic conditions are favoured, so sugar present in the dough is converted all the way to carbon dioxide rather than to alcohol. It is CO_2 production that causes the bread to rise. Any small amount of ethanol that may be produced is evaporated during the baking process.

Many other popular foodstuffs are the result of microbial fermentation processes (see Table 18.3). These include vinegar, soy sauce and sauerkraut. *Silage* is animal fodder made from the fermentation of grass and other plant material by the action of lactic acid bacteria.

Table 18.3 Fermented food products

Product	Source	Fermentative microorganisms
Sauerkraut	Cabbage	*Leuconostoc brevis, Lactobacillus plantarum*
Olives	Olives	*Leuconostoc brevis, Lactobacillus plantarum, Lactobacillus brevis*
Pepperoni	Ground beef, pork	*Lactobacillus plantarum, Pediococcus pentosaceus*
Pickles	Cucumber	*Leuconostoc brevis, Lactobacillus plantarum, Pediococcus* spp.
Soy sauce	Soybean curd	*Aspergillus oryzae, Saccharomyces rouxii, Pediococcus soyae*
Tempeh*	Soybean	*Rhizopus oligosporus*
Kombucha	Tea	*Gluconobacter, Saccharomyces*, etc.
Sake	Rice	*Aspergillus oryzae, Saccharomyces cerevisiae*
Vinegar	Wine, cider	*Acetobacter, Gluconobacter*
Cocoa, chocolate	Cacao beans	*Saccharomyces cerevisiae, Candida rugosa, Acetobacter, Geotrichum*

*Tempe (or tempeh) is a solid fermented soya bean 'cake' that is widely consumed as a meat substitute in Indonesia. It forms an important part of the diet of many Indonesians.

18.1.4 Microorganisms as food

As we have seen in the previous sections, a number of microorganisms are involved in the production of food products. Others, however, *are* foodstuffs! Perhaps the most obvious of these are mushrooms, the stalked fruiting bodies of certain species of Basidiomycota (see Chapter 8), notably *Agaricus bisporus*. These are grown in the dark at favourable temperatures in order to stimulate the production of fruiting bodies. Another fungus, *Fusarium venenatum*, forms the basis of Quorn™, a processed mycoprotein that has been used as a meat substitute for some years in the UK. Whereas mushrooms are grown as agricultural products, Quorn™ must be produced under highly regulated sterile conditions. Other microbial food sources include certain algae (seaweed), which form an important part of the diet in some parts of the world, and bacteria and yeast grown in bulk as *single-cell protein* (SCP) for use as a protein-rich animal food supplement. The cyanobacterium *Spirulina* has been collected from dried-up ponds in parts of central Africa for use as a food supplement since time immemorial and is now available at health stores in the West.

18.1.5 The microbial spoilage of food

We have described in previous chapters the nutritional versatility of microorganisms and their role in the global recycling of carbon. Unfortunately for us, fresh foods such as meats, fruit and vegetables provide a rich source of nutrients, which a wide range of heterotrophic microorganisms find just as attractive as we do. Certain microbial types are associated with particular foodstuffs, depending on their chemical composition and physical factors such as pH and water content. Acidic foods such as fruits, for example, tend to favour the growth of fungi rather than bacteria.

Often, spoilage organisms come from the same source as the food – for example, soil on vegetables, or meat exposed to the contents of the animal's intestine following slaughter. Others are introduced as contaminants during transport, storage or preparation. Among the most commonly found spoilage organisms are a number of human pathogens, including *Pseudomonas, Salmonella, Campylobacter* and *Listeria*. Thus, although microbial spoilage may merely lead to foodstuffs being rendered unpalatable, in other cases it can also result in serious and even fatal illness ('food poisoning'). Whilst observable changes to foodstuffs are only likely after the microbial population has reached a considerable size, food poisoning can result from the presence of much smaller numbers of contaminants.

Some foodstuffs are more susceptible to spoilage than others: fresh items such as meat, fish, dairy produce, and fruit and vegetables are all highly perishable. Foods such as rice and flour, on the other hand, are much more resistant, because having no water content they do not provide suitable conditions

for microbial growth. Drying is one of a number of methods of food preservation, all designed to prevent growth of microorganisms by making conditions unfavourable. Other methods include heating/canning, drying, pickling, smoking and, in many countries, irradiation. Such methods all have to strike a balance between inhibiting microbial growth and not having an adverse effect on the appearance, texture and palatability of the foodstuffs.

18.2 Microorganisms in the production of biochemicals

Many products of microbial metabolism find an application in the food and other industries. These include amino acids, enzymes, steroids and antibiotics (Table 18.4). Microbial growth conditions are adjusted so that production of the metabolite in question takes place at an optimal rate. Often an unnaturally high rate of production is achieved by the use of a mutated or genetically engineered strain of microorganism, or by manipulating culture conditions to favour excess metabolite production.

The development of a microbial means of producing *acetone* was vital to the allied effort in World War I. Acetone was a crucial precursor in explosives manufacture, and the demands of war soon outstripped supply by traditional methods of production. The problem was solved when Chaim Weizmann isolated a strain of *Clostridium acetobutylicum* that could ferment molasses to acetone and butanol (another industrially useful product). Nowadays, acetone is made more cheaply from petrochemicals.

Microbially produced *amino acids* are used in the food industry, in medicine and as raw materials in the chemical industry. The one produced in the greatest quantities by far is *glutamic acid* (almost two million tons per year), with most of it ending up as the flavour enhancer monosodium glutamate. The amino acids *aspartic acid* and *phenylalanine* are components of the artificial sweetener aspartame and are also synthesised on a large scale.

Table 18.4 Commercially useful products of microbial metabolism

Product	Use
Amino acids:	
Glutamic acid	Flavour enhancer
Lysine	Animal feed additive
Aspartic acid + phenylalanine	Artificial sweetener (aspartame)
Citric acid	Antioxidant, flavour enhancer, emulsifier
Enzymes	Numerous – see Table 18.5
Antibiotics	Treatment of infectious diseases
Vitamins	Dietary supplements
Steroids	Anti-inflammatory drugs, oral contraceptives

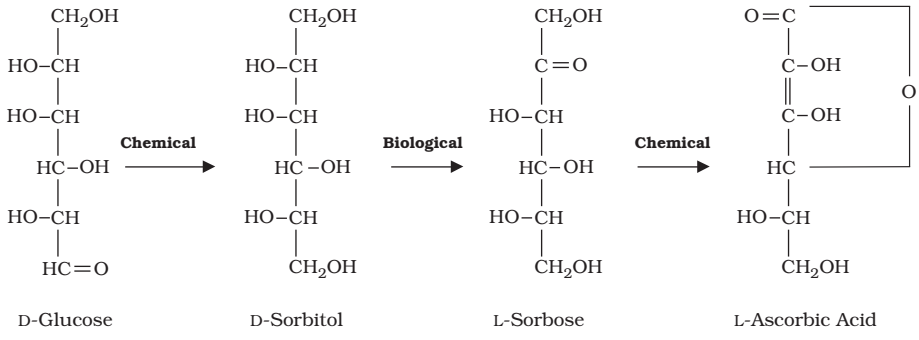

Figure 18.4 Ascorbic acid is produced by a combination of chemical and microbial reactions. Most steps in the synthesis of ascorbic acid are purely chemical, but the conversion of sorbitol to sorbose is carried out by the sorbitol dehydrogenase enzyme of *Acetobacter suboxydans*.

A number of organic acids are produced industrially by microbial means, most notably *citric acid*, which has a wide range of applications in the food and pharmaceutical industries. This is mostly produced as a secondary metabolite by the large-scale culture of the mould *Aspergillus niger*.

Certain microorganisms serve as a ready source of *vitamins*. In many cases these can be synthesised less expensively by chemical means; however, *riboflavin* (by the mould *Ashbya gossypii*) and *vitamin B_{12}* (by the bacteria *Propionibacterium shermanii* and *Pseudomonas denitrificans*) are produced by large-scale microbial fermentation. Microorganisms play a partial role in the production of *ascorbic acid* (*vitamin C*). Initially, glucose is reduced chemically to sorbitol, which is then oxidised by a strain of *Acetobacter suboxydans* to the hexose sorbose. Chemical modifications convert this to ascorbic acid (Figure 18.4).

Enzymes of fungal and bacterial origin have been utilised for many centuries in a variety of processes. It is now possible to isolate and purify the enzymes needed for a specific process, and the worldwide market is worth over a billion pounds. The most useful industrial enzymes include proteases, amylases, lipases and pectinases. Thanks to advances in protein engineering it is now becoming possible to 'design' completely new enzymes with specific properties for industrial applications. Some applications of enzymes are listed in Table 18.5, and two examples are briefly described below. Syrups and modified starches are used in a wide range of foodstuffs, including soft drinks, confectionery and ice cream, as well as having a wealth of other applications. Different enzymes or combinations of enzymes are used to produce the desired consistencies and physical properties. *High fructose corn syrup* (HFCS) is a sweetener used as a component of a multitude of processed foods since the 1970s. It is some 75% sweeter than sucrose and has several other advantages. HFCS is a mixture of fructose, dextrose (a form of glucose) and disaccharides, and is produced by the action of a series of three enzymes on

Table 18.5 Some industrial applications of microbially produced enzymes

Industry	Enzyme	Application
Food and drink	Rennet	Cheese manufacture
	Lipase	
	Pectinase	Fruit juice production
		Coffee bean extraction
	Amylase	Improved bread dough quality
		Haze removal in beer
	Amylase	Fructose syrup production
	Glucoamylase	
	Glucose isomerase	
Animal feed	Amylase	Improved digestibility
	Cellulase	
	Protease	
Detergent	Protease	Stain and grease removal
	Lipase	
	Amylase	
	Cellulase	Fabric softener
Paper	Cellulase	Pulp production
Textiles	Cellulase	'Stone-washed' jeans
Leather	Protease	Dehairing, softening, fat removal
	Lipase	
Molecular biology	*Taq* polymerase	Polymerase chain reaction

the starch (amylose and amylopectin) of corn (maize). *Alpha amylase* hydrolyses the internal α-1,4 glycosidic bonds of starch, but is not able to degrade ends of the chain. The resulting di- and oligosaccharides are broken down to the monomer glucose by the action of *glucoamylase*, then finally *glucose isomerase* converts some of the glucose to its isomer fructose.

Enzymes have been added to *cleaning products* such as washing powders, carpet shampoos and stain removers since the 1960s, and this remains one of the principal industrial applications of enzymes. *Proteases* are the most widely used enzymes in this context; working in combination with a surfactant, they hydrolyse protein-based stains such as blood, sweat and various foods. Greasy and oily stains present a different challenge, made all the more difficult by the move towards lower washing temperatures. The inclusion of *lipases* aids the removal of stains such as butter, salad dressing and lipstick, while *amylases* deal with starch-based stains such as cereal or custard. The food and detergent industries between them account for around 80% of all enzyme usage.

We have already seen in Chapter 17 that *antibiotics* are now produced on a huge scale worldwide. Figure 18.5 outlines the stages in the isolation, development and production of an antibiotic.

Isolating an antibiotic from a natural source is not all that difficult, but finding a new one that is therapeutically useful is another matter. Initially, the

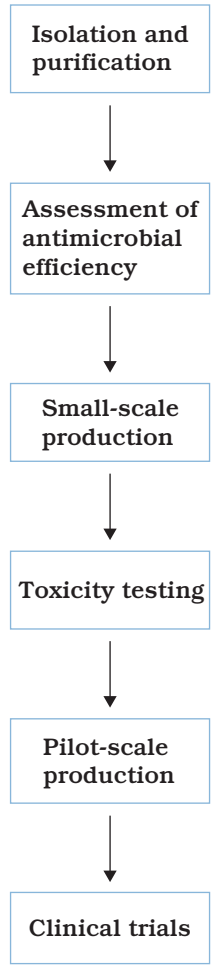

Figure 18.5 Stages in the isolation and development of an antibiotic. See the text for details.

antimicrobial properties of a new isolate are assessed by streaking it across an agar plate, then inoculating a range of bacteria at right angles (Figure 18.6). As the antibiotic diffuses through the agar, it will inhibit growth of any susceptible species. Isolates that are still deemed worth persisting with are then grown up in a laboratory scale fermenter; it is essential for commercial culture that the antibiotic-producing organism can be cultured in this way.

Before committing to large-scale production, exhaustive further tests must be carried out on two fronts: to ascertain the potency of the preparation and the breadth of its antimicrobial spectrum, and to carry out toxicity testing on animals to determine its *therapeutic index* (see Chapter 17). The final stages of development involve pilot-scale production, followed by clinical trials on human volunteers.

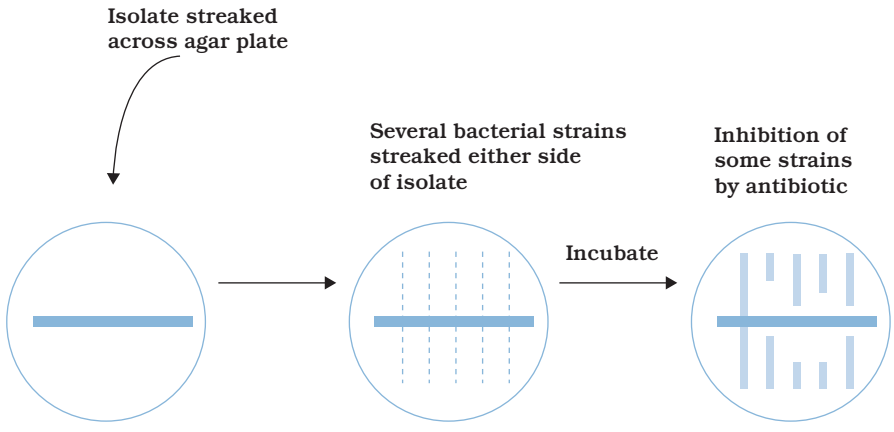

Figure 18.6 Assessing the antimicrobial properties of an antibiotic. Candidate antibiotic is streaked onto an agar plate along with several bacterial isolates. Following incubation, areas of clearing indicate inhibition of growth and thus susceptibility to the antibiotic.

When an antibiotic or any other fermentation product finally goes into production, it is cultured in huge stirred fermenters or *bioreactors*, which may be as large as 200 000 litres. A typical stirred fermenter has impellers for mixing the culture, an air line for aeration and microprocessor-controlled probes for the continuous monitoring and regulation of temperature, pH and oxygen content (Figure 18.7). Cultures with a high protein content may also have an antifoaming agent added. The process of *scale-up* is a complex operation, and not simply a matter of growing the microorganism in question in ever larger vessels. Factors such as temperature, pH and aeration must all be considered at the level of the individual cell if scale-up is to be successful. Fermenters are usually made from stainless steel, which can withstand heat sterilisation; the economic consequences of microbial contamination when working on such a large scale can be immense.

18.3 Products derived from genetically engineered microorganisms

In Chapter 12 we saw how microorganisms can be genetically modified so that they produce commercially important proteins. This is done by incorporating the gene for the desired protein into an appropriate vector and inserting it into a host cell such as *E. coli* or *Saccharomyces cerevisiae*. The initial application of this technology was in the microbial production of medically important proteins such as insulin and epidermal growth factor (Table 18.6); however, bacteria and other microorganisms can be genetically modified to produce a range of other products such as pharmaceuticals, vaccines and modified enzymes. These include enzymes used in diagnostic

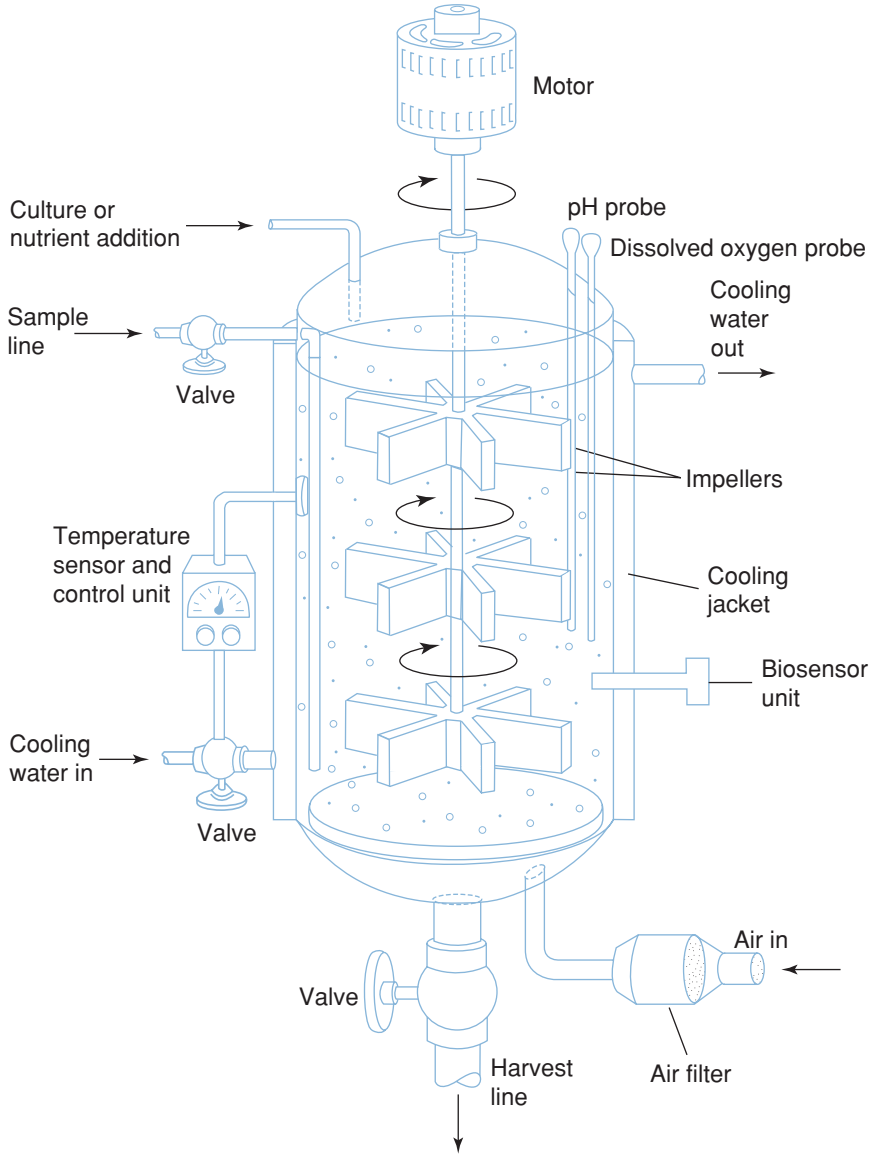

Figure 18.7 A continuous flow stirred tank reactor. Parameters such as pH and concentrations of specific metabolites are closely monitored to ensure the maintenance of optimum conditions. Outlets allow for the collection of samples during fermentation as well as the collection of cells and medium at the conclusion of the reaction. Additions and collections are carried out under aseptic conditions.

Table 18.6 Medically important proteins made by recombinant microorganisms

Protein	Application	Produced in
Insulin	Treatment of type 1 diabetes	*E. coli*
Human growth hormone	Treatment of pituitary dwarfism	*E. coli*
Hepatitis B vaccine	Vaccination of susceptible personnel, e.g. healthcare workers, drug users	*Saccharomyces cerevisiae*
Epidermal growth factor	Treatment of wounds, burns	*E. coli*
Acyltransferase	Used in synthesis of ovarian cancer drug taxol	*E. coli*
Endostatin	Antitumour agent	*Pichia pastoris* (yeast)
Tissue plasminogen activator (tPA)	'Clot-busting' drug	*E. coli*

and analytical applications, where a higher purity of preparation is required than, for example, the enzymes used in detergents. These are often derived originally from other microorganisms; for example, the thermostable DNA polymerase from *Thermus aquaticus* used in the polymerase chain reaction (PCR) is now commonly made by recombinant *E. coli* cells transformed with the *T. aquaticus* gene. Many of the more recent recombinant human proteins to be developed for therapeutic use have been too complex for expression in a microbial system (e.g. factor VIII), so it has been necessary to employ cultured mammalian cells.

18.4 Microorganisms in wastewater treatment and bioremediation

These applications of microbial processes in an environmental context are discussed in Chapter 14.

18.5 Microorganisms in the mining industry

An unexpected application for microorganisms is to be found in the mining industry. Acidophilic bacteria, including *Acidithiobacillus ferrooxidans*, are increasingly being used to extract valuable metals, notably copper, from low-grade ores that would not be worth working by conventional technologies. You may recall from Chapter 14 that *A. ferrooxidans* is the organism largely responsible for the phenomenon of acid mine drainage; by carrying out the same reactions in a different context, however, it can be put to a beneficial use. Tailings, that is, mineral waste with a low metal content, are gathered in huge tips and acidified water is sprinkled over them (Figure 18.8), stimulating the growth of indigenous bacteria. Bacterial oxidation results in soluble copper sulphate leaching from the tip and being collected for copper extraction. This bacterial action is known as direct bioleaching, but if you follow the

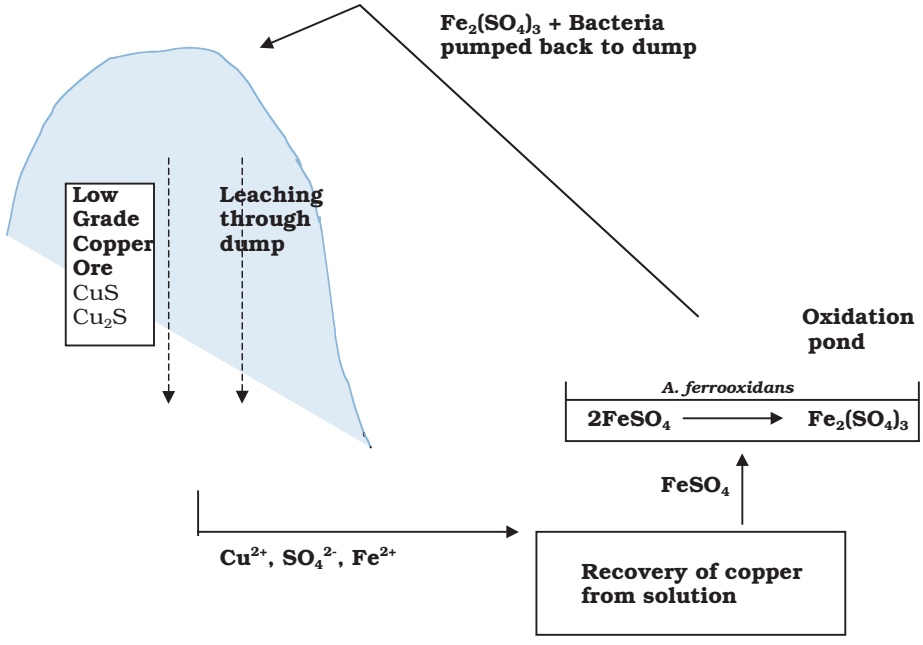

Figure 18.8 The bacterial extraction of copper. Solubilisation of copper sulphides occurs by a combination of direct (biological) and indirect (chemical) leaching. The ferric iron necessary for chemical oxidation is produced by bacterial oxidation of ferrous iron in the oxidation pond. Reproduced from Prescott, LM, et al. (2002) Microbiology, 5th edn, with permission from McGraw-Hill.

process in Figure 18.8, you will see that *A. ferrooxidans* has not finished yet! This remarkable organism can also oxidise iron from its ferrous to ferric form; the resulting ferric sulphate is a potent oxidising agent, which, when recycled to the tip, carries out indirect (chemical) bioleaching, and so the cycle continues. *A. ferrooxidans* has a number of other unusual features that enable it to survive in this hostile environment; it thrives in acidic conditions (pH <2.0), and has an unusually high tolerance of metal ions such as copper. Operations such as this must be carefully controlled to avoid serious adverse effects on the environment.

Bacteria are also involved in the extraction of other metals such as uranium and gold; the methodologies differ slightly, but still involve the conversion of an insoluble compound to a soluble one. It is only in the last couple of decades or so that the economic possibilities of *biohydrometallurgy* have been fully appreciated, and now a significant proportion of the world's copper and other metals is produced in this way. The method is inexpensive but rather slow; it may take years to extract the copper from a large tip. However, as high-grade copper-bearing ores become increasingly scarce, it seems likely to play an increasingly important role.

Sulphur-oxidising bacteria also have a role to play in the coalmining industry. Increased environmental awareness in many countries means that it is no longer acceptable to burn off the sulphur content of coal as sulphur dioxide; so an alternative must be found. One possibility is the *biodesulphurisation* of coal, using sulphur-oxidising bacteria to remove the sulphur before combustion. Whilst technically feasible, economic considerations mean that this has not yet been widely adopted.