

R-01-05

Project SAFE

Microbial features, events and processes in the Swedish final repository for low- and intermediate-level radioactive waste

Karsten Pedersen
Göteborg University

January 2001

Svensk Kärnbränslehantering AB

Swedish Nuclear Fuel
and Waste Management Co
Box 5864
SE-102 40 Stockholm Sweden
Tel 08-459 84 00
+46 8 459 84 00
Fax 08-661 57 19
+46 8 661 57 19



Project SAFE

Microbial features, events and processes in the Swedish final repository for low- and intermediate-level radioactive waste

Karsten Pedersen
Göteborg University

January 2001

Denna rapport har gjorts på uppdrag av SKB. Slutsatser och framförda åsikter i rapporten är författarnas egna och behöver nödvändigtvis inte sammanfalla med SKB:s.

Contents

	Page
Abstract	5
Summary	7
Sammanfattning	11
1 Microbes and microbial processes – general principles applicable to the SFR	13
1.1 The microbe – what is it?	13
1.2 The microbe – what can it do?	14
1.2.1 Microbial growth and activity	14
1.2.2 Biofilms	16
1.3 The microbe – why does it transfer electrons?	16
1.4 The microbe – what does it need?	17
1.4.1 Water	18
1.4.2 Temperature	18
1.4.3 Nutrients for growth	18
1.4.4 Energy and carbon	18
2 The prospect for microbial life in the SFR repository – environmental considerations	19
2.1 The repository environments	19
2.1.1 The waste types	19
2.1.2 Microbial environments in the SFR repository	24
2.2 Evaluation of transport processes in the repository, of importance for microbial activity	24
2.2.1 Diffusion in the repository	25
2.2.2 Advective flow in the repository	26
2.2.3 Mobility of microbes in the repository	26
2.2.4 Processes independent of transport	26
2.2.5 Transport-dependent processes	29
2.3 Repository content and possible microbial activities	32
3 Evaluation of the prospects of microbially induced processes in the repository	33
3.1 Bitumen degradation	33
3.2 Concrete degradation	34
3.3 Microbial corrosion of metal containers	35
3.4 Gas consumption	36
3.5 Cellulose degradation	37
3.6 Biogeochemical considerations	37
3.7 Formation of complexing agents	38
3.8 Clogging of preferential flow paths	39
3.9 Radionuclide dissolution and mobilisation	39
3.10 Methylation and alkylation of radionuclides	40
3.11 Gas formation	41
3.11.1 Gas formation in the repository	41

4	Microbial processes in the SFR interaction matrix	43
4.1	Environmental effects on the microbial state	43
4.2	Effects of the microbes on the state of the repository	44
5	References	47
	Appendix	51

Abstract

The waste disposed of in the Swedish final repository for low and intermediate radioactive waste (SFR) typically contains large amounts of organic substances. This waste thus constitutes a possible source of energy and nutrients for microorganisms. Microbes can degrade the waste to degradation products, which to a varying degree may create problems if the process is significant. The environment for microbial life in the SFR is, however, unique since it cannot be compared to any environment to which microbes have adapted naturally over millions of years. Most similar to the SFR are waste dumps and landfills. In those, microbes degrade the waste and form degradation products. The experience from such “analogues” and from research performed under repository-like conditions may provide useful clues about the microbial processes which may occur in the repository.

Microbes have the ability to degrade bitumen, used to solidify some wastes, but this degradation is very slow under anaerobic conditions. Bitumen degradation will, therefore, not influence the safety of the SFR. However, some microbes can produce acids that could influence concrete stability, particularly in the presence of oxygen. The future SFR environment is anaerobic, which suggests that acid production is a very unlikely problem. Sulphate-reducing bacteria (SRB) have the ability to produce sulphide, which may act as a corrosive on metals. Under specific conditions, with the local groundwater flow close to a metal surface and with dissolved organic material from the repository, pitting corrosion of metal canisters is a potential threat. This process appears to require conditions fairly atypical of the SFR, however. Large groups of microorganisms can use hydrogen as a source of energy, thereby contributing to the decrease of this gas mainly formed from water during the anaerobic corrosion of metals. Cellulose is an excellent substrate for many microorganisms and it will be the dominating carbon and energy source for microbially catalysed processes in the SFR. Degradation of cellulose will influence the chemical composition of the groundwater that flows through the SFR. Oxidised electron acceptors (e-acceptors) will be reduced. Many microorganisms can produce complexing agents that may have an accelerating effect on radionuclide migration. They may also influence radionuclide mobility by oxidation or reduction of the species, or by sorbing them into their cell envelope. The radionuclides will then be transported with the microbes instead of being sorbed on rock surfaces or other stationary, radionuclide-sorbing phases. Finally, as part of their metabolism, many microbes produce gases, especially carbon dioxide and methane. Under optimal conditions, large volumes of gas may be formed. Experiments performed in the UK under repository conditions showed that gas formation was initially fast, but that it decreased after the initial phase.

Summary

Waste disposed of in the Swedish final repository for low and intermediate radioactive waste (SFR) comes in a variety of forms and typically, it contains a significant microbially degradable component. Cellulose is an obvious such constituent. Ion exchange resins with organic matrices, various garbage and scrap materials and bitumen can also undergo microbial degradation, provided the conditions for microbial activity are favourable. Microbial degradation activities may cause unwanted effects in the SFR. The proper evaluation of the risk of these various events taking place requires a significant base in microbiology, including data from published experimental work. Textbook evaluation alone is not sufficient because of the unique environmental conditions the microbes encounter in the repository. In many respects, these repository environments are “new” to the microbes in the sense that they do not exist in nature and the microbes are therefore not fully adapted to deal with the various constraints of life that are created in the SFR. Still, specific parts of the SFR may, under specific conditions, favour microbial activity that may negatively influence SFR performance. The aim of this report is to evaluate whether there exist microbial processes that may threaten the integrity of the SFR.

The SFR environment offers many possible interactions between microbes and various other elements. Below follows a summary of possible interactions and how they relate to each other.

Microbial degradation of organic material conditioned in concrete may produce gas, with alkaline hydrolysis of cellulose being a possible first step, followed by fermentation of monomers and oligomers to gas. The gases formed may be carbon dioxide, methane, hydrogen, hydrogen sulphide or nitrogen. The process is restricted by alkaline conditions and hydrogen produced by corrosion. The types of waste and waste composition will influence microbial activity and the growth of microorganisms in the waste containers. The most favourable position for microbes, with respect to available energy, is inside the containers. However, restricted availability of electron acceptors (e-acceptors) and the build-up of toxic degradation products may limit the types and number of microbes that may proliferate inside containers. Bitumen can be degraded by microbes and is therefore a possible substrate for microbial activity. The growth of microbes would predominantly occur on bitumen surfaces inside steel canisters, locally generating large numbers of microbes. The backfill and the space in vaults may offer less stagnant and alkaline conditions for microbes, compared with inside the containers and inside the SILO. However, the amount of available energy will be much less here than in the waste. Significant microbial activity is only expected if waste, or degradation products from the waste (including alkaline degradation products), reaches these areas. The amount of organic carbon, nutrients and e-acceptors in the groundwater interacting with the waste influences the microbial activity. Of great importance is the availability of e-acceptors, such as oxygen (not expected), nitrate, ferric iron, manganese(IV), sulphate and carbon dioxide. The SFR is rich in nutrients and energy and these components will not be limiting for microbial activity *per se*. The magnitude, direction and distribution of groundwater flow in the different SFR components will influence the transport of microbes and, much more importantly, the transport of e-acceptors to, and degradation products from, microbes dwelling in various SFR locations. This interaction is judged to be the most important in relation to most other interactions

involving microbes. The amount of hydrogen and carbon dioxide is of importance to the state of microbes in the SFR. At high concentrations, hydrogen may be inhibitory and carbon dioxide is important for the activity of autotrophic microbes, such as methane-forming microbes. Oxygen is not expected except during the initial phase. Oxygen is a very potent e-acceptor and as a result, will rapidly be used up for microbial activity. Microbial activity generally increases with temperature in the interval between $-15\text{ }^{\circ}\text{C}$ and $+113\text{ }^{\circ}\text{C}$. The species distribution changes over temperature as the growth interval for a single microbe generally comprises no more than $20\text{--}30\text{ }^{\circ}\text{C}$.

Microbial growth is possible in the waste that has been solidified with cement. The growth may be significant if an advective flow supplies the microbes with e-acceptors, removes degradation products and leaches the concrete. Under stagnant hydraulic conditions, the effect will be insignificant. Specific species of bacteria can produce sulphuric and nitric acid, which may deteriorate cement and concrete. Microbial growth is possible as biofilms on the surfaces of bitumen-contained waste. The growth may be significant if there is an advective flow to supply the microbes with e-acceptors and remove degradation products. The bitumen will be degraded at a slow rate if conditions are anaerobic. Degradation is much higher under aerobic conditions. The effect will be insignificant under stagnant hydraulic conditions. Steel-contained bitumen waste is most susceptible to this process. Non-solidified waste has the greatest potential for microbial degradation. The organic carbon content is very high and pH will be less alkaline than in cement-solidified waste. The large mobility of microbes and waste components inside a container suggests that gas production may become significant here. Microbial growth is possible on the outside of cement containers. Again, the growth may be significant if an advective flow supports the microbes with e-acceptors, removing degradation products and leaching the concrete. The effect will be insignificant under stagnant hydraulic conditions. The species of bacteria that can produce sulphuric and nitric acid may deteriorate the cement and concrete containers also from the outside. Microbial corrosion is a well-established process and pitting corrosion by sulphate-reducing bacteria (SRB) may rapidly corrode holes into the steel containers. The growth and pitting corrosion may be significant if there is an advective flow to support microbes locally with sulphate and organic substrates.

In the concrete backfill, too, microbial growth is possible, and may be significant in the presence of an advective flow. The effect will, however, be low under stagnant hydraulic conditions. If present, sulphuric and nitric acid-producing bacteria will deteriorate cement and concrete. Microbial growth is possible on concrete structures and may be significant in the presence of an advective flow. Compacted bentonite with a density of $> 1800\text{ kg/m}^3$ has been demonstrated to be unfavourable for microbial growth. The main constraints are the low water availability, low porosity and the stagnant transport conditions allowing only diffusive transport of nutrients, energy sources and degradation products. However, the SILO will have a compaction at 1000 kg/m^3 and will contain 85–90% sand at both the top and the bottom. This bentonite barrier will not restrict microbial activity. It is possible that transport of microbes will be somewhat restricted in the pure bentonite, owing to pore size limitations. Growth of microbes in vaults and backfill may alter the hydraulic conditions, if the growth is significant. Biofilms may form and clog the preferential flow pathways. This process would probably be possible only downstream of the SFR in the “plume” containing dissolved material from the interior of the SFR, which can be used for significant growth. Microbial growth will produce a series of different degradation products that will alter the groundwater composition inside and downstream of the SFR. Dissolved carbon dioxide and methane, acids, ferrous iron, sulphide and complexing organic

agents may form, while sulphate, nitrate, ferric iron, hydrogen and oxygen will be reduced owing to microbial consumption. The activity of microbes under anaerobic conditions may eventually lead to a significant production of gas, mostly methane. Some hydrogen and carbon dioxide may be consumed during autotrophic methane production, but most of the methane will be liberated from the degradation of the organic wastes in the SFR. Microbial activity may, if vigorous, generate a great deal of heat. The SFR environment will, however, not be so rich in easily degradable organic carbon and e-acceptors that the temperature will be influenced.

Microbial activity may influence radionuclides in many different ways. The production of organic acids and other complexing agents may increase the mobility of radionuclides. The microbes may also act as sorbents for radionuclides and mobilise or immobilise them, depending on whether they are attached or unattached. In addition, microbes may change the chemical form of radionuclides in the course of their metabolism. The methylation of metals, turning inorganic species to organic, and carbon fixation of $^{14}\text{CO}_2$ to organic carbon, and degrading organic ^{14}C -carbon to $^{14}\text{CO}_2$, is one example. Such transformations alter the mobility of the elements.

Sammanfattning

I SFR lagras avfall som till stor del utgörs av organiska ämnen. Detta avfall utgör därmed en möjlig energi- och näringskälla för mikroorganismer. Mikrober kan bryta ned avfallet till olika komponenter som i varierande omfattning kan skapa problem i den mån nedbrytningen är omfattande. Miljön för mikrobiellt liv i SFR är emellertid speciell eftersom den inte direkt kan jämföras men någon av de miljöer som mikroorganismer naturligt anpassat sig till. Det är huvudsakligen soptippar och olika typer av landutfyllnader som kommer SFR närmast i likhet. I dessa förekommer mikrobiell aktivitet med nedbrytning av avfallet och bildning av restprodukter som följd. Erfarenheter från studier av sådana "analogier" samt från försök utförda under förvarsliga förhållanden kan ge en god vägledning om vad som kommer att ske i SFR.

Mikrober kan bryta ned bitumen, som används för att binda en del av avfallet, men sådan nedbrytning är mycket långsam under syrefria förhållanden. Denna process kommer därför inte att påverka säkerheten i SFR. Vissa mikroorganismer kan, främst i närvaro av syre, bildas syror som i princip kan påverka cement och betongkonstruktionerna i SFR. SFR kommer emellertid att vara syrefritt under förvarstiden och syrabildning blir därför försumbar. Sulfatreducerande bakterier har förmåga att bilda sulfid, som kan verka korrosivt på metaller. Under speciella förhållanden, med lokala grundvattenflöden nära metallytor och med tillskott av organiska ämnen från förvaret kan viss gropfrätning ske. Det bedöms dock bli en förhållandevis ovanlig process. Många mikroorganismer kan utnyttja vätgas som energikälla och därmed bidra till att reducera halten av denna gas som i SFR huvudsakligen bildas vid korrosion av metall. Cellulosa är ett utmärkt substrat för många mikroorganismer och det kommer att vara den huvudsakliga kol- och energikällan för mikrob-katalyserade processer i SFR. Nedbrytning av cellulosa kommer att påverka den kemiska sammansättningen av grundvattnet i SFR. Oxiderade elektron acceptorer kommer att reduceras. Många mikroorganismer kan producera komplexbildande ämnen som kan ha en accelererande inverkan på transport av radionuklider. De kan också påverka radionuklidens rörlighet genom att oxidera eller reducera dem, eller genom att sorbera dem till sitt cellhölje. Radionukliderna kan då komma att transporteras med mikrober istället för att fastna på berg eller andra radionuklid-sorberande faser. Slutligen bildar många mikrober gaser i sin metabolism, främst koldioxid och metan. Under gynnsamma förhållanden kan det bildas stora mängder gas, men experiment utförda i England tyder på att gasbildningen avtar efter ett initialt bildningsskede.

1 Microbes and microbial processes – general principles applicable to the SFR

Waste disposed of in the Swedish final repository for low and intermediate radioactive operational waste (SFR) comes in a variety of forms and typically contains a significant microbially degradable component. Cellulose is an obvious such constituent. Ion exchange resins with organic matrices, various garbage and scrap materials and bitumen can likewise undergo microbial degradation, provided the conditions for microbial activity are favourable. Microbial degradation activities may cause unwanted effects in the SFR. Possibly the worst case scenario is the microbial production of gas in amounts that may damage storage containers and force contaminants into the groundwater. Large-scale microbial production of complexing agents from organic components in the repository, microbial concrete dissolution, microbial methylation of radionuclides and microbial radionuclide dissolution and mobilisation are other possible unwanted events. The proper evaluation of the risk of these various events requires a significant base in microbiology, including data from published experimental work. Textbook evaluation alone is not sufficient because of the unique environmental conditions the microbes encounter in the SFR. These repository environments are in many respects “new” to the microbes in the sense that they do not exist in nature. The microbes are, therefore, not fully adapted to deal with the various constraints of life that are created in the SFR. Still, under specific conditions, specific parts in the SFR may favour microbial activity that may negatively influence SFR performance. The aim of this report is to evaluate whether there exist microbial processes that may threaten the integrity of the SFR.

1.1 The microbe – what is it?

A microbe is a living entity which contains all it needs in order to perform a life cycle, including feeding, growth and reproduction, in one single cell. The size of microbes varies significantly, from the smallest bacterium with a diameter of about 0.2 μm to some unicellular animals and plants which may reach 1 mm or more in diameter. The organisms on Earth cluster in three major domains, viz. *Bacteria*, *Archaea* and *Eukarya*. All organisms in the domains *Bacteria* and *Archaea* are microbes. Most of the branches of the domain *Eukarya* are microbial as well, including fungi, unicellular animals and algae. Microbes can therefore be found virtually everywhere in the tree of life (see SKB TR 00-04, Section 2.1, for details). They constitute the absolute dominating diversity of life on our planet. Biochemically, much of this diversity is contradictory to multicellular life, the diversity of which is largely morphological. The enormous biochemical diversity among the microbes explains their huge adaptability to almost any environment where temperature allows life, viz. from $-15\text{ }^{\circ}\text{C}$ to $+113\text{ }^{\circ}\text{C}$.

A typical bacterium is a very robust creature that generally survives extremely well in the niche it is adapted to live in. Different bacteria are adapted to different conditions and as a group, the bacteria cover all possible combinations of environmental circumstances. The domain *Bacteria* comprises many millions of species. This vast diversity of unknown species represents an uncertainty with respect to possibly unknown microbial processes of importance to the SFR.

Microorganisms in the domain *Archaea* were regarded as bacteria until molecular data revealed that they belong to a domain which is totally different from all bacteria as well as all plants, animals and fungi. A unifying characteristic of organisms in this domain is their ability to adapt to what is called “extreme conditions”. Different species of *Archaea* are active under different conditions. Some *Archaea* are adapted to extreme pH levels as low as 1 or as high as 12, and some may even survive under conditions of lower/higher pH. The high pH conditions in the SFR are, consequently, not a conceptual hindrance to microbial activity. Another group of *Archaea* of importance to the repository is the methanogens. They produce methane gas from hydrogen and carbon dioxide, or from one- or two-carbon organic compounds, such as formate, methylamine, methanol or acetate.

1.2 The microbe – what can it do?

The microbe can be regarded as a multi-functional catalyst. It may speed up reactions that normally occur very slowly or not at all under repository conditions. Under optimal conditions, it has the ability to multiply exponentially, with a doubling time of < 15 minutes. The microbes consequently can adjust their population numbers rapidly, according to the prevailing conditions. The SFR environment will be far from optimal in general terms, but local spots may offer appropriate conditions for growth, as will be analysed later in this report.

1.2.1 Microbial growth and activity

The most common way of culturing microorganisms in the laboratory is by using a batch culture. A culture vessel is supplied with all constituents necessary for growth, and inoculated with the microbe of interest. A typical batch growth curve can be registered (Fig. 1-1). First, there is an adaptation phase during which the cells adjust to the conditions in the culture vessel. Then the cells start to divide and grow exponentially to high counts, doubling their number at regular time intervals. Finally, when some limiting component is used up, or when a toxic component forms at too high a concentration (e.g. alcohol in fermentation cultures), growth is arrested. Figure 1-1 shows that the batch-grown cells basically are active only during the exponential growth phase. The batch culture represents a closed system with no input or output of components from the system. It can be compared to the various SFR containers with bitumen or concrete casted waste and the SILO. Some diffusion of components may occur but this will generally be a very slow process. The SFR will enter a lasting stationary state with no growth once the conditions for growth become unfavourable. However, the batch approach would predict some significant microbial growth when groundwater fills up the SFR.

The batch system is a superb tool for many research purposes in the laboratory but it does not mimic the life of microbes in natural environments. Such environments generally consist of a huge number of open systems with a continuous input and output of matter in between. Rather than being based on a batch culture, models of microbial processes in the SFR may alternatively be based on a continuous culture situation, as described below.

The hard rock aquifer environment in the SFR can be considered as an open system. A particular fracture will have a water composition that reflects the origin of the water and various reactions between the solid and liquid phases that occur along the flow path.

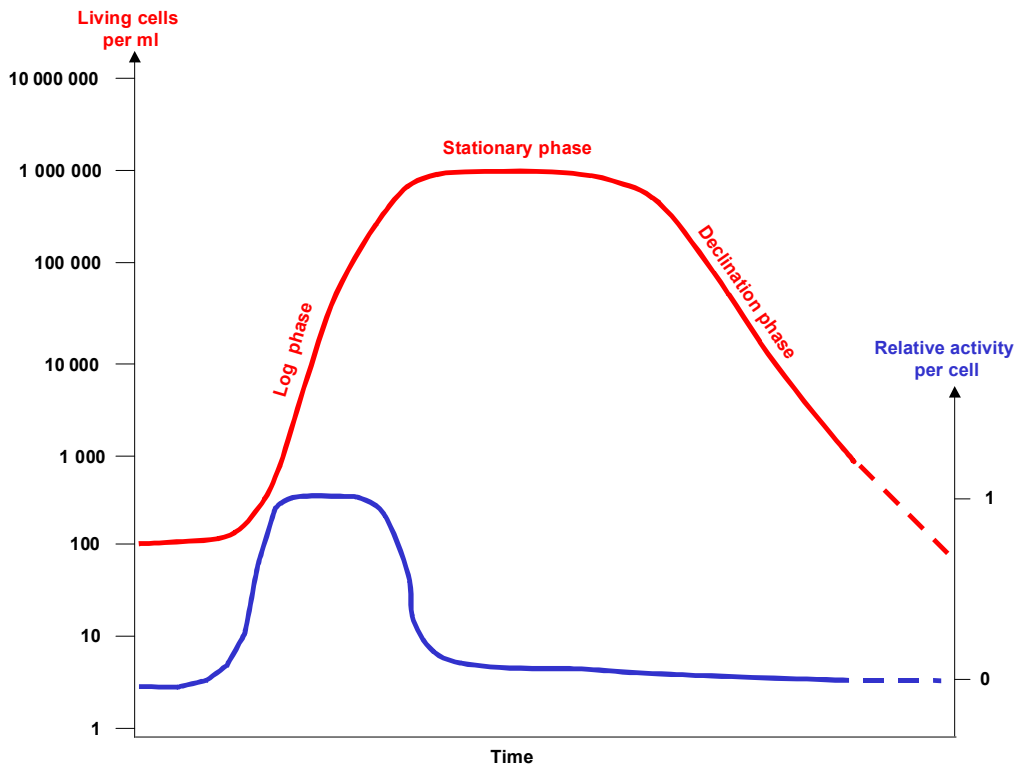


Figure 1-1. A schematic representation of microbial growth in a closed batch culture. The microbes are basically active only during the exponential growth phase, when they double their numbers during a specific time period. The doubling time can be as short as 15 minutes for some easily culturable microbes or it may last many hours or even weeks for microbes more difficult to work with.

A new composition may be the result of two fractures meeting and of their water mixing. These processes may be slow but there is a continuum of varying geochemical conditions in hard rock aquifers at repository depth, and the SFR with all its alien construction components will add variance to these conditions. Microbes are experts on utilising any energy in the environment that becomes thermodynamically available for biochemical reactions. A slow but steady flow of organic carbon from the waste, for instance isosaccharinic acid (ISA) from alkaline degradation of cellulose, may support a continuum of microbial growth and activity in the repository. Alternatively, a flow of reduced gases such as hydrogen from corrosion processes and methane from microbial degradation processes of organic waste components may be utilised.

Continuous growth of microbes can be studied in the laboratory using a chemostat. The culture vessel is continuously supplied with energy by a slow inflow of nutrients. The inflow is balanced by an outflow that removes waste products and some cells. The number of cells will therefore remain constant in the chemostat. The microbes will, however, be active (Fig. 1-2). Unlike the batch system, a chemostat system is open as it has an influx and outflow of matter. The continuous culture situation of the chemostat is applicable to any hard rock aquifer or SFR waste container experiencing a flux of matter through advective groundwater flow. The flows may be very slow, but over geological time scales, they may be significant.

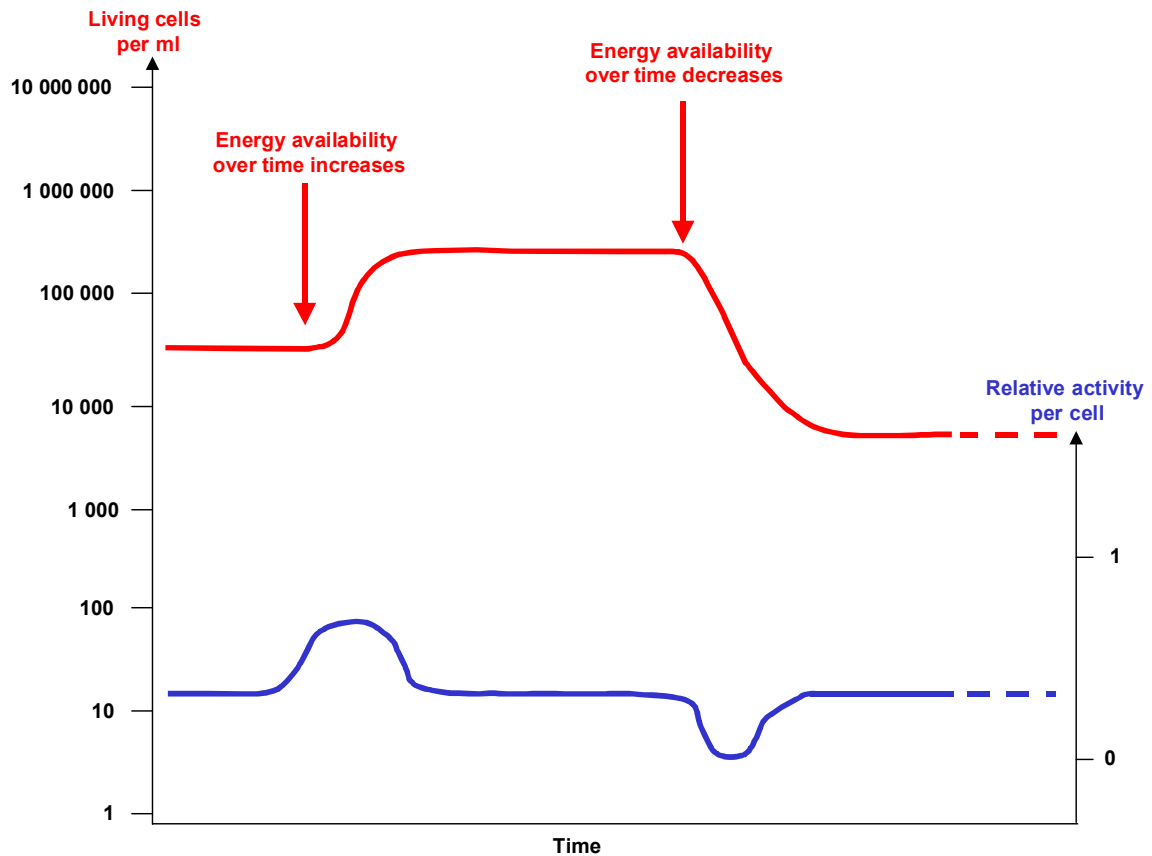


Figure 1-2. A schematic representation of microbial growth in an open, continuous culture system. The microbes are continuously active except for periods when there is a decrease in availability of energy over time.

1.2.2 Biofilms

A special case is the possible occurrence of microbes that grow attached to aquifer surfaces, a phenomenon that has repeatedly been observed in groundwater from deep hard rock aquifers (Ekendahl et al. 1994; Pedersen et al. 1996). Such biofilms will increase their cell numbers until they reach steady state, as previously described for continuous growth of unattached microbes.

1.3 The microbe – why does it transfer electrons?

Metabolising microbes perform many different fermentative and respiratory electron transfers, as part of their energy-harvesting metabolism. Their decomposition of organic material depends on the sources of energy and on the electron acceptors (e-acceptors) present. In the SFR, organic carbon, hydrogen, methane and reduced inorganic molecules are possible sources of energy. During microbial oxidation of these energy sources, the microbes use e-acceptors in a certain order (Fig. 1-3). First, oxygen is used, followed by nitrate, manganese, iron, sulphate, sulphur and carbon dioxide. Simultaneously, fermentative processes may supply the respiring microbes with hydrogen and short organic acids. The solubility of oxygen in water is low, but for

many microbes, oxygen is the preferred e-acceptor. This is because the microbes get much more energy per organic molecule if the molecule has been oxidised, than they do with the other e-acceptors listed in Figure 1-3. Consequently, microbial activity will contribute significantly to the SFR becoming anaerobic soon after it is closed and filled with water. The respiratory processes in Figure 1-3 require continuous access to oxidised e-acceptors from the environment. Electrons are then transferred from the substrate, that is, organic constituents in the SFR are transferred to the acceptor that becomes reduced and “useless” to microbes in an anaerobic environment. In the case of absence of external e-acceptors, many microbes turn to fermentative processes. They comprise an internal electron transfer within the substrate, and the need for an external electron transfer is therefore neutralised. For example, the fermentation of glucose results in ethanol, which is more reduced than is glucose, and carbon dioxide, which is more oxidised than is glucose.

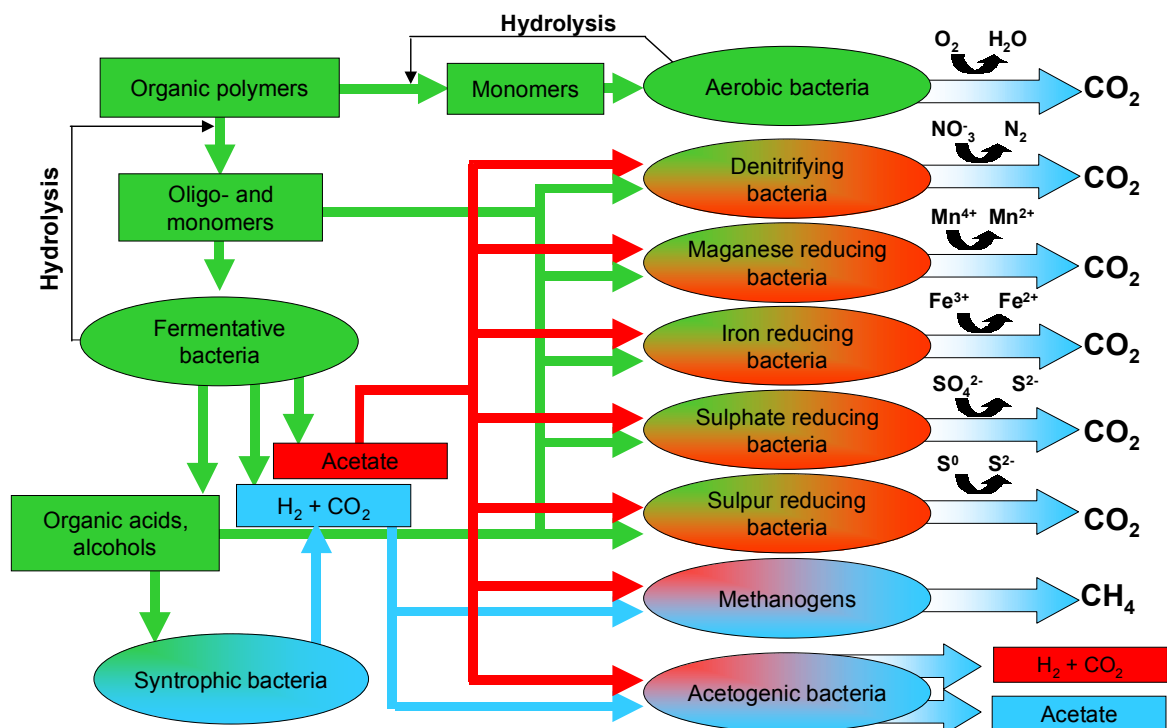


Figure 1-3. The degradation of organic carbon can occur via a number of different metabolic pathways, characterised by the principal e-acceptor in the carbon oxidation reaction. A range of significant groundwater compounds are formed or consumed during this process. Of great importance for SFR disposal is the turnover of gases such as carbon dioxide, hydrogen and methane.

1.4 The microbe – what does it need?

The active microbe needs a liquid water environment, temperatures of between $-15\text{ }^{\circ}\text{C}$ and $113\text{ }^{\circ}\text{C}$, nutrients for its synthesis of cell constituents and, most importantly, one or more utilisable energy sources.

1.4.1 Water

The requirement for water is absolute; active life without water is not possible. A suitable measure of availability of water, or of water content, is the thermodynamic water activity (a_w) of a system in equilibrium (Potts 1994). The water activity of a solution is related to the relative humidity of air in equilibrium with a solution or a water-containing SFR material. In other words, at a given temperature, a_w is the ratio between the vapour pressure of the solution (e.g. concrete) (P_s) and that of water (P_0).

$$a_w = P_s / P_0 \quad (1)$$

Thus, water activity is a measure of the relative tendency of water to escape from the system, compared with pure water, and can adequately be described by the relative humidity that the system can maintain in equilibrium. Microorganisms can grow over a large range of a_w s (0.999–0.75), but most favour an a_w of saltwater (0.98) or more. The cell wall and the cell membrane of most microbes are freely permeable to water, meaning that the water activity outside a microbe must be balanced by the water activity inside. Microbial growth and activity will, consequently, only be possible in the SFR at places where the water activity is large, in other words, generally where groundwater has entered the waste. The interior of bitumen-contained waste cannot be degraded by microbes owing to the lack of water.

1.4.2 Temperature

Microbes can live at temperatures between $-15\text{ }^\circ\text{C}$ and $113\text{ }^\circ\text{C}$. Different species are adapted to different temperatures. The SFR is not expected to generate heat, and therefore, microbial activity will proceed at relatively low temperatures in the $10\text{--}15\text{ }^\circ\text{C}$ interval. This would favour microbes occurring naturally in the groundwater and this may restrict the activity of microbes coming with the waste from high temperature sites.

1.4.3 Nutrients for growth

All living cells synthesise cell components and require specific components for doing so. Nitrogen is needed for amino acid synthesis and can be obtained from nitrogen gas or from various organic and inorganic sources. Phosphorus is another substance that is needed. Phosphorus can be obtained from certain minerals, such as apatite. The cells also require tiny amounts of many trace elements, metals and sometimes also organic constituents, such as vitamins.

1.4.4 Energy and carbon

Microbes require energy for their life processes. This energy can be obtained from organic or inorganic sources. Most types of naturally occurring organic compounds can be degraded by microbes for their extraction of energy. Commonly, organic energy sources are concomitantly used as a source of carbon for synthesis of cell constituents. Many inorganic compounds can likewise be utilised as energy source. One of the most effective inorganic compounds is molecular hydrogen, which is an excellent source of energy used by many different microbes. Sulphide, ferrous iron and manganese(II) are other energy sources used by various microbes. Their carbon for cell synthesis is extracted from carbon dioxide, which is reduced with electrons from the energy source. The SFR is very rich in compounds that may serve as energy sources for microbes. Large amounts of utilisable energy in the form of organic material and hydrogen from anaerobic corrosion of iron and aluminium are available in the SFR, as is carbon dioxide in the groundwater.

2 The prospect for microbial life in the SFR repository – environmental considerations

2.1 The repository environments

2.1.1 The waste types

The SFR comprises four disposal environments, namely the two concrete tank caverns (BTF), the cavern for low-level radioactive waste (BLA), the cavern for intermediate-level radioactive waste (BMA), and the SILO for intermediate-level radioactive waste. At the end of 1999, a total of 2919 tons of ion exchange resins, 2734 tons of garbage and scrap, 527 tons of ash, a 106 ton reactor lid and 7 tons of sludge were deposited in the SFR (Skogberg 2000b). The main components by weight in the SFR are therefore ion exchange resins and garbage and scrap materials. In addition to the waste itself, container and solidification materials, steel concrete, cement and bitumen are added to the SFR, commonly in masses much larger than the waste disposed of. The absolute dominating component in the SFR, then, is concrete, followed by bitumen and steel. Table 2-1 gives the maximum weights of the disposal packages. The BLA and BTF caverns receive very large containers (10–20 m³), while BMA and SILO containers generally are smaller (0.2–1.73 m³). Table 2-2 a-d lists the types of waste disposed of in the SFR, distributed over the disposal area, container material and solidification material. The BLA and BMA waste is a mix of most components deposited, while the BTF and SILO wastes are generally ion exchange resins.

Table 2-1. Distribution of container and waste material over the four different SFR sub-repositories, given as the maximum weight of each material deposited in a container during 1999.

Material	BLA	BTF	BMA	SILO
	(kg)	(kg)	(kg)	(kg)
<i>Containers</i>				
Iron/steel	2700	647	660	400
Cellulose	310	0	9	9
Remaining organic material	0	0	10	10
<i>Waste</i>				
Iron/steel	7400	0	180	480
Aluminum/zink	500	5	6	12
Cellulose	4000	0	156	8
Ion exchange resins	4600	1400	650	670
Bitumen	3400	0	820	960
Sludge	60	60	115	333
Evaporated concentrate	0	0	161	0
Remaining organic material	3000	66	580	444

Data are taken from Skogberg 2000a.

Table 2-2 a. Waste container types deposited in SFR concrete tank caverns since 1988.

Deposition site	Container type	Solidification material (weight)	Size and weight	Waste content (weight)	Radio-activity	Additions	Waste type
BTF	concrete	–	3.3 m x 1.3 m x 2.3 m (16 tons)	dewatered ion exchange resins and filter material (2–2.5 tons)	low	metal oxides sodium sulphate (2%)	B.07
BTF	armed concrete	–	3.3 m x 1.3 m x 2.3 m (16 tons)	dewatered ion exchange resins and filter material. Small amounts of metal hydroxides and sodium sulphate may occur (2–2.5 tons).	low	–	O.07
BTF	steel-concrete barrel	–	(300–450 kg)	ash from incineration of low-level radioactive waste (76,5 kg)	low	–	S.13
BTF	–	–	105.6 tons	reactor lid	low	–	R.99:2

Data are taken from Skogberg 2000b, and Riggare et al. 2001.

BTF = concrete tank cavern.

Table 2-2 b. Waste container types deposited in SFR caverns for low-level radioactive waste since 1988.

Deposition site	Container type	Solidification material (weight)	Size and weight	Waste content (weight)	Radio-activity	Additions	Waste type
BLA	steel	–	standard container (10–20 tons)	Solid garbage and scrap metals	low	–	B.12, F.12, R.12
BLA	½ standard container	–	6 m x 2.4 m x 1.3 m (maximum 20 tons)	36 200 l barrels of solid garbage and scrap metals, some glass, insulation, and organic material (36 x 100 kg = 3600 kg)	low	–	S.14
BLA	standard container	bitumen (3420 kg)	standard container (10 tons)	36 barrels of the F.05 type with ion exchange resins, small amounts of filtering material and evaporation concentrate (4680 kg)	low	Si-based antifoam and starch	F.20
BLA	standard container	bitumen (5400 kg)	standard container (10 tons)	36 barrels of the B.05 type with ion exchange resins, small amounts of filtering material and evaporation concentrate (1800 kg)	low	emulsifier and sodium sulphate	B.20

Data are taken from Skogberg 2000b, and Riggare et al. 2001.

BLA = cavern for low-level radioactive waste.

Table 2-2 c. Waste container types deposited in SFR caverns for intermediate-level radioactive waste since 1988.

Deposition site	Container type	Solidification material (weight)	Size and weight	Waste content (weight)	Radio-activity	Additions	Waste type
BMA	steel barrel	bitumen (150 kg)	59 cm x 88 cm (200 kg)	ion exchange resins and evaporate concentrate (50 kg)	intermediate	emulsifier and sodium sulphate (3–4 kg)	B.05
BMA	steel barrel	bitumen (95 kg)	59 cm x 88 cm (250 kg)	ion exchange resins, small amounts of filtering material, and evaporation concentrate (130 kg)	intermediate	Si-based antifoam and starch	F.05
BMA	steel	bitumen (820 kg)	1.2 m x 1.2 m x 1.2 m (2080 kg)	ion exchange resins, filtering material and small amounts of evaporation concentrate (774 kg)	intermediate	–	F.17
BMA	steel	cement	1.2 m x 1.2 m x 1.2 m (2380 kg)	ion exchange resins, filtering material and small amounts of evaporation concentrate (536 kg)	intermediate	cement additions	F.15
BMA	steel or concrete	cement	1.2 m x 1.2 m x 1.2 m (maximum 5000 kg)	garbage and scrap metals	intermediate	–	F.23, O.23, R.23
BMA	armed concrete	cement	1.2 m x 1.2 m x 1.2 m (maximum 5000 kg)	ion exchange resins and filter material (130 kg)	intermediate	cement additions	O.01.9
BMA	armed concrete	cement	1.2 m x 1.2 m x 1.2 m (maximum 5000 kg)	ion exchange resins and filter material (130 kg)	intermediate	cement additions, calcium oxide	R.01
BMA	armed concrete	cement	1.2 m x 1.2 m x 1.2 m (maximum 5000 kg)	sludge from decontamination and from cleanups, large variations in content (115 kg)	intermediate	cement additions, calcium oxide	R.10
BMA	armed concrete	cement	1.2 m x 1.2 m x 1.2 m (maximum 5000 kg)	ion exchange resins and filter material (250 kg)	intermediate	cement additions, calcium oxide	R.15
BMA	containers	–	–	odd waste, containers with ash from wet incineration of condensation waste	intermediate	–	F.99:1

Data are taken from Skogberg 2000b, and Riggare et al. 2001.

BMA = cavern for intermediate-level radioactive waste.

Table 2-2 d. Waste container types deposited in the SFR SILO since 1988.

Deposition site	Container type	Solidification material (weight)	Size and weight	Waste content (weight)	Radio-activity	Additions	Waste type
SILO	steel barrel	bitumen (150 kg)	59 cm x 88 cm (200 kg)	ion exchange resins and evaporate concentrate (50 kg)	intermediate	emulsifier and sodium sulphate (3–4 kg)	B.06
SILO	steel	bitumen (960 kg)	1.2 m x 1.2 m x 1.2 m (2080 kg)	ion exchange resins, filtering material and small amounts of evaporation concentrate (600 kg)	intermediate	–	F.18
SILO	armed concrete	cement	1.2 m x 1.2 m x 1.2 m (maximum 5000 kg)	ion exchange resins and filter material (130 kg)	intermediate	cement additions (> 4%)	O.02
SILO	armed concrete	cement	1.2 m x 1.2 m x 1.2 m (maximum 5000 kg)	ion exchange resins and filter material (130 kg)	intermediate	cement additions, calcium oxide	R.02
SILO	armed concrete	cement	1.2 m x 1.2 m x 1.2 m (maximum 5000 kg)	ion exchange resins and filter material (250 kg)	intermediate	cement additions, calcium oxide	R.15

Data are taken from Skogberg 2000b, and Riggare et al. 2001.

SILO = Silo for intermediate-level radioactive waste.

2.1.2 Microbial environments in the SFR repository

The SFR can be regarded as a set of closed or semi-closed batch environments placed in an open, continuous system, viz. the cavern/rock/aquifer environment (cf. Section 1.2.1). The BLA and BMA caverns are filled with waste packages that are mostly solidified with concrete or bitumen (Table 2-2b, c). Most of the BTF packages, however, are not solidified (Table 2-2 a). Many of the packages contain organic material and materials that may corrode and produce hydrogen. Hydrogen and organic material can principally be consumed and degraded by microbes, at rates that are determined by the environmental conditions. The package temperature will be within the range where life is possible. The complex nature of the waste in most of the packages will offer any nutrient a microbe could ask for. Water will intrude most of the packages, with the exception of waste solidified in bitumen. Energy and carbon will be present in the waste in large amounts, offering substrate for microbial conversion to various metabolic products (see Fig. 1-3). It is obvious that the SFR would become a huge bioreactor unless microbial activity was restricted in various ways.

2.2 Evaluation of transport processes in the repository, of importance for microbial activity

A typical landfill has its content of organic material in common with the SFR, and contaminants and microbial degradation products certainly migrate from such a site (Christensen et al. 1994). It therefore appears important to understand in what respect the SFR will differ from a normal landfill and to establish whether these differences will make the migration of contaminants from the SFR, especially radionuclides, significant or non-significant. Any microbial process rate ultimately depends on transport processes. Required substances must reach the microbe and waste products from the metabolism must be removed to avoid toxic effects. This can be accomplished in several ways. Firstly, diffusion is a slow but possible transport mechanism. The concentration dependence of diffusive processes may, however, strongly limit the rate at which microbial activity can proceed. Secondly, attached microbes, so-called “biofilms”, utilise flow transport and their metabolic rate may become flow-dependent. Thirdly, many microbes are mobile, and can therefore move up and down concentration gradients towards nutrients and away from toxic compounds. These transport processes will be discussed below in more detail, with the SFR closed/semi-closed package environments and open cavern systems in focus.

The waste packages can be regarded as parcels with solid components that may act as a source of energy for microbes. Figure 2-1 illustrates two such adjacent packages in the BTF, BLA or BMA. The containers are stored with a slot of a certain distance, D , between them. This allows a groundwater flow that will flow at various rates, depending on the geological situation. The groundwater flow at the SFR repository has been calculated (Holmén and Stigsson 2001). During the time the SFR is under the sea, the hydraulic gradients in the rock mass below the sea will be small, and the groundwater flow is expected to be small as well. In the rock mass surrounding the tunnels, the average specific flow is calculated to be within a range of 1-3 litre/(m², year). The total flow through the storage tunnels of the repository (SILO, BMA, BLA and BTF tunnels) has been calculated to about 40m³/year. In the case of a land raise, which would take the SFR above sealevel, the average specific flow in the surrounding rock masses will increase to about 7-9 litre/(m², year), and the flow through the storage tunnels will

increase to about 170 m³/year. Such flow scenarios, of under and above sea level cases, will be discussed below, with respect to microbial activity in the SFR.

Many organic carbon sources can be degraded by microbes via two different processes, fermentation and respiration (Fig. 1-3). Principally, fermentation of the organics in the SFR will not require the transport of components from outside the SFR, while respiration will require a continuous supply of e-acceptors with groundwater from outside the SFR.

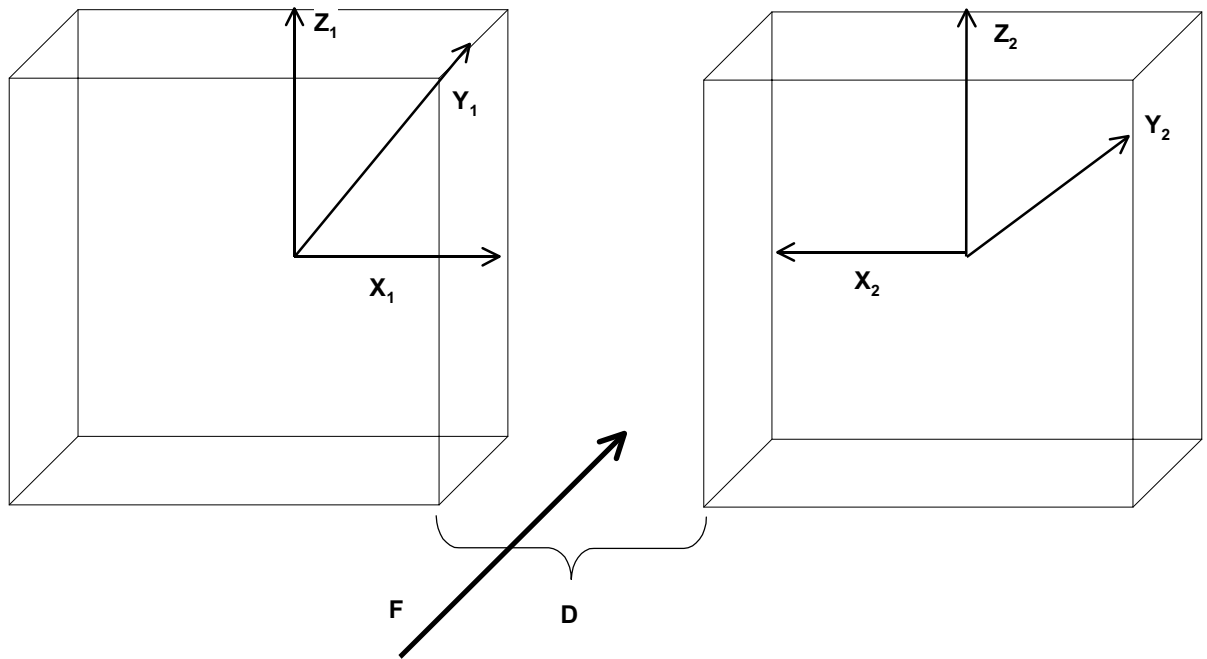


Figure 2-1. Containers in the BTF, BLA and BMA caverns will be stored with some distance, D , between them. They will have different sizes, X , Y , and Z , as listed in Table 2-2. Groundwater will move between the containers at a flow rate, F .

2.2.1 Diffusion in the repository

Diffusion is one of three fundamental processes that may contribute to the rate of change of matter or energy present at a given position in the SFR. The other two processes are active transport of matter, for example by advection or convection, and *in situ* production, for example by radioactive decay. In geochemistry, diffusion is defined as the process by which atoms, molecules, ions or lattice vacancies move in a solvent from one position to another under the influence of a chemical potential gradient. In most cases, chemical potential is directly related to concentration, and diffusion can usually be considered as a process related to concentration gradients. Ficks' first law states that the diffusional flux of matter, F , is proportional to the spatial concentration gradient (Stuwe 1999).

$$F = -D * \partial C / \partial x \quad (2)$$

In the equation, F is the volumetric flux of matter transported per cross-sectional area and per unit time. The proportionality constant, D , is called the “diffusivity coefficient”

and has the unit of m^2/s . The distance of diffusion can only be shown in terms of a concentration profile and varies according to the components and media studied. Diffusion is usually much slower in solids than in liquids. The bitumen-contained waste in the SFR will therefore react much more slowly with the environment than will the concrete- and steel-contained waste. Diffusion will only be important for microbial activity over short distances, that is, through cell capsule material, through the cell membrane, and through biofilms when active transport of matter occurs in the SFR. The distance from most of the waste to the surrounding rock aquifers is so great, and most of the concentration gradients are so low, that diffusion can be neglected as a significant long-distance transport process for microbial activity, in contrast to the advective flow transport.

2.2.2 Advective flow in the repository

There are several possibilities of a significant groundwater flow occurring through the SFR. One is that a flow may occur between containers, through cracks induced by corrosion, or along the vaults. The most significant event increasing the flow rates in the SFR will be the predicted land raise, but gas release from corroding metal may also cause local movement of water through the SFR. This report will evaluate firstly, which of the microbial processes relate to advective flow and secondly, the range of results assumed for the SFR. Basically, microbial activity will be directly correlated to the flow rate through a flow path. Most of the microbes will be attached to the waste and the proper situation to evaluate is how biofilms will react to what the flow may transport to and from the microbes. Figure 2-2 illustrates the relation between flow and microbial activity in a marine system. The biofilm productivity is strongly dependent on the flow at low flow rates. This is so because biofilm growth depends on transport of growth stimulation components in the flowing water and on removal of growth-limiting waste products from their metabolism. The flow in Figure 2-2 is in the cm s^{-1} interval, which is much faster than what can be expected in most hard rock aquifers at repository depth. The steep inclination of the productivity curve at low flow rates suggests a strong flow dependence of biofilms at the rates expected in rock aquifers. The flow rate of groundwater in the SFR will be the most important growth-controlling factor, once energy for microbial activity is made available in the waste.

2.2.3 Mobility of microbes in the repository

Many microbes have the ability to move in specific directions. Microbes can take out directions in chemical gradients and can move upwards (an attractant) or downwards (a repellent) in such gradients. The velocity with which microbes swim is in the range of $20\text{--}100 \mu\text{m s}^{-1}$. This velocity may take a microbe $1.5\text{--}7.5 \text{ m}$ a day, provided it swims in the same direction the whole day. The mobility of microbes will probably not transport significant amounts of material or radionuclides. Rather, it ensures that microbes will inhabit any space in the SFR, which has degradable material that can be reached by mobile bacteria. The porosity must be in the micrometer range, however. Once on its degradation site, the microbe will be transport-controlled, as discussed below.

2.2.4 Processes independent of transport

A fermentation process comprises an internal electron transfer within the substrate. For instance, when glucose is fermented to alcohol and carbon dioxide, the glucose molecule is split and electrons are moved to the alcohol from which carbon dioxide will be made. There is no need for external electron donors or acceptors. Fermentation processes may, theoretically, occur in the SFR without the need of transport of external

components, until the limiting concentration of a fermentation product arrests growth. For example, ethyl alcohol limits fermentation processes at 10–15 weight %. Hydrogen is another fermentation product which blocks fermentative metabolisms at low concentrations, although the limiting concentration varies significantly between species and fermentation processes. Hydrogen produced by corrosion processes will also act as a limiting factor on microbial activity.

A second metabolic route for microbial growth in a stagnant waste container system is respiration of organic components, according to Figure 1-3, using e-acceptors present in the waste. Intrinsic e-acceptors will be used up and the growth process will be arrested.

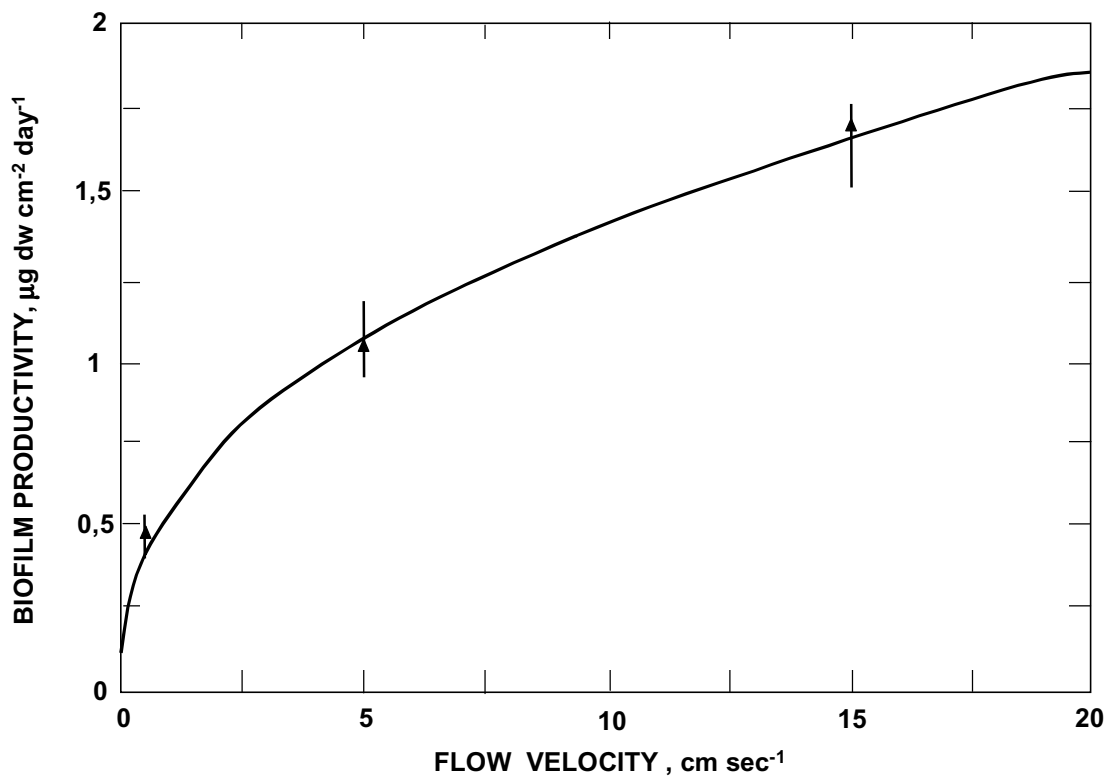


Figure 2-2. The biofilm productivity of a marine biofilm was studied at different flow rates. An initial strong dependence levelled out as the flow increased. It was concluded that biofilms are strongly transport-dependent at low flow rates and that this dependence decreases with an increase in the flow rate (Pedersen 1982).

Intact containers in the SFR will offer limited growth conditions for microbes, provided water is present. Dry waste does not degrade. Figure 2-3 illustrates two intact, adjacent waste containers, according to Figure 2-1. A significant amount of energy for microbes will be available in the organic material present in most containers, and the additions to the concrete also contain degradable material (Table 2-2). A limited amount of e-acceptors will be present in the groundwater that fills the SFR, and possibly, in the waste, but at a low flow situation, transport will be limited to diffusion, which is a slow, concentration-dependent process. The intrinsic e-acceptors will be consumed by microbes during their oxidation of the organic material. New e-acceptors can only reach the microbes via diffusion, a process which may be neglected (see Section 2.2.1). The respiratory processes will be followed by fermentative processes, progressing until a

limitation point is reached. These growth processes will follow a typical batch curve, as illustrated in Figure 1-1. The amount of end-products will be approximately the same at growth arrest, independent of how much energy is available in the waste, because it is the end-products' toxicity that will limit microbial activity. Experiments have been performed with waste containers buried in shallow land (Macdonald et al. 1997). The microbial activity in these containers depended on water content and they all exhibited batch-type cumulative gas product curves.

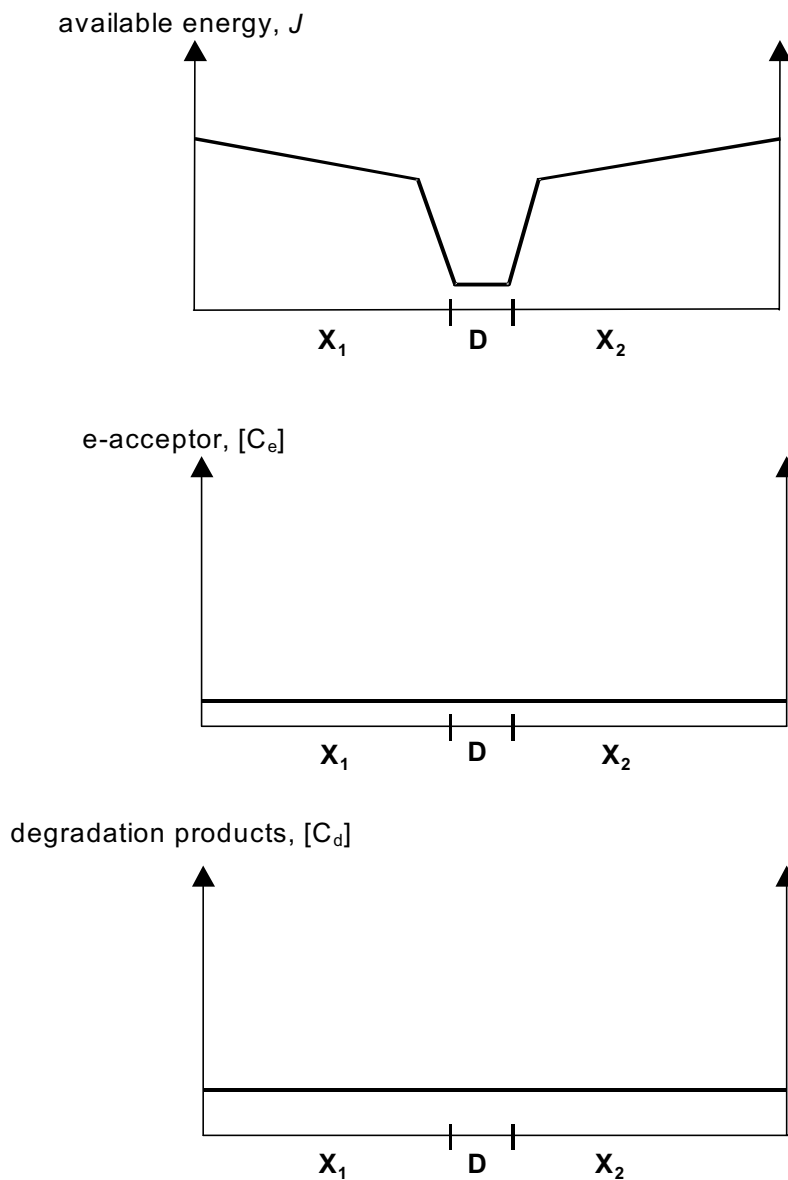


Figure 2-3. A schematic representation of available energy, e-acceptors and degradation products in a stagnant SFR system of the type illustrated in Figure 2-1 with a flow rate of $F=0$. Little energy will be available between the containers. The intrinsic e-acceptors will be consumed. Fermentation processes will be arrested by the build-up of toxic degradation products, limiting the continuing microbial creation of degradation products.

2.2.5 Transport-dependent processes

The prospects of microbial growth in the SFR will be different if a flow situation appears. An advective flow of groundwater from outside the SFR (Fig. 2-4) between containers in the BTF, BLA and BMA caverns (Fig. 2-1) will replenish e-acceptors and remove degradation products. These components would still need to diffuse into the containers unless cracking or corrosion has opened up flow paths through the containers. A flow situation with groundwater flowing through the SFR differs from the no-flow situation in that e-acceptors will be replenished much closer to the waste, than if they had to diffuse from outside of the vaults (Fig. 2-5). This flow situation would possibly occur in discrete parts of the SFR as a result of corrosion-induced cracking of concrete or other processes that break up the container integrity. The SILO will be different in that grouting between the containers will make such a flow impossible, unless the bentonite barrier fails and corrosion-induced cracking allows a flow.

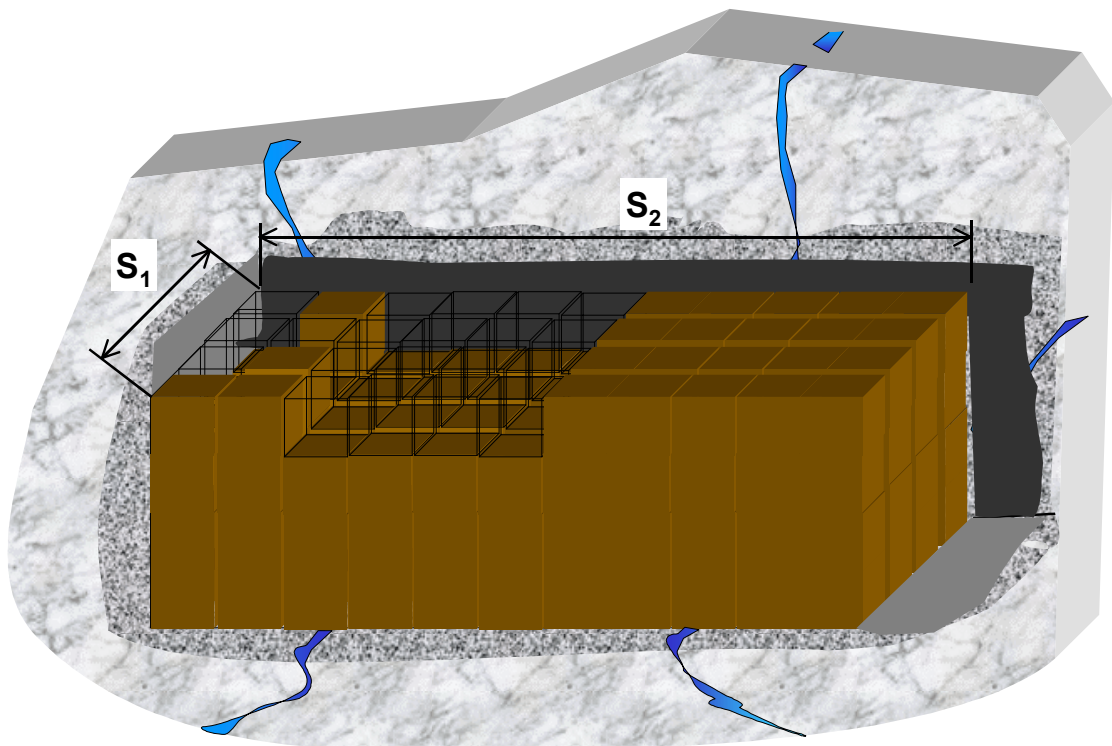


Figure 2-4. The flow situation in the BTF, BTF and BMA caverns. Fractures connect the vaults to the groundwater. A preferential flow path may form along the vaults or through the storage space, between containers. With preferential flow paths, only parts of the waste will be exposed to the flow.

A scenario of groundwater flowing between containers and occasionally through containers would allow a series of continuous microbial processes not possible in a stagnant situation. The continuous supply of new e-acceptors and removal of degradation products (Fig. 2-6) would unlock the growth-limiting factors discussed above, inducing a continuous microbial growth situation of the type represented in Figure 1-2. Microbial biofilms may build up, as reported for different groundwater systems (Pedersen and Ekendahl 1992; Ekendahl and Pedersen 1994; Pedersen et al. 1996) and drinking water (Pedersen 1990). Eventually, such biofilms may clog up preferential flow paths and force the flow in new directions. A continuous waste

degradation process in a preferential flow path may continuously release radionuclides to the flow. Degradation of ^{14}C -organic carbon could lead to production of ^{14}C -methane which will escape via the flow to the biosphere, either dissolved or as gas bubbles. Other radionuclides, for example from ion exchange resins, may be released from the waste by microbial activity and transferred to the flowing groundwater. The microbes will, through their metabolic activity, consequently add a radionuclide release factor to any migration model involving a case of advective flow.

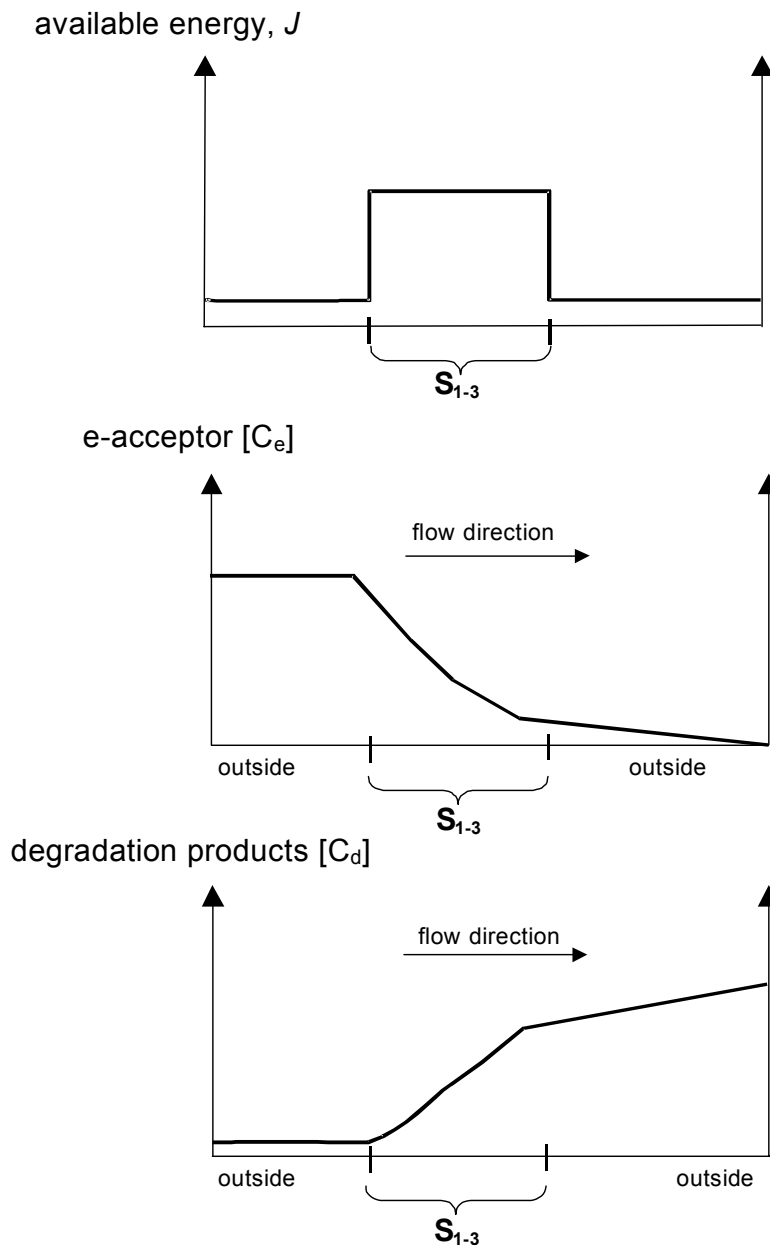


Figure 2-5. A flow through the BLA, BTF or BMA caverns would resemble the situation typical of any waste disposal site. The waste constitutes a source of energy for microbial activity. Electron acceptors are supplied with the flowing groundwater and degradation products spread downstream of the repository.

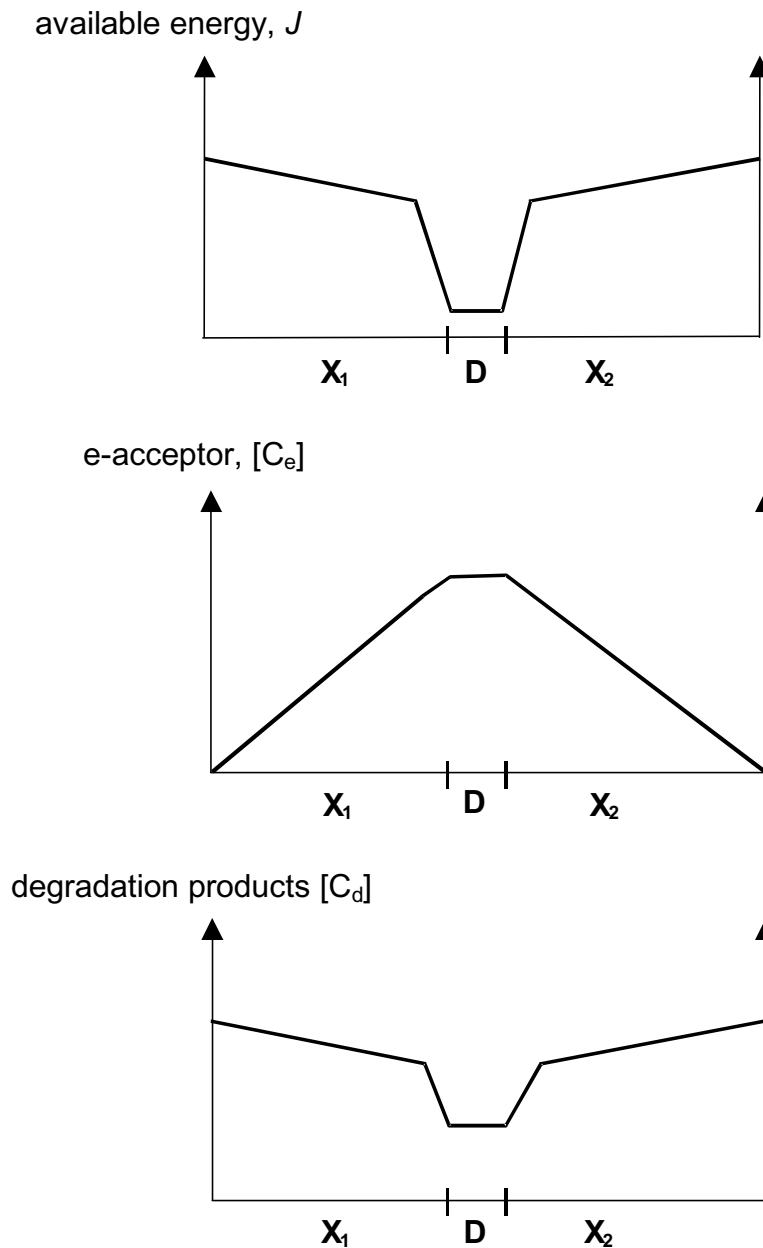


Figure 2-6. A schematic representation of available energy, e-acceptors and degradation products in an SFR system of the type illustrated in Figure 2-1, with the flow rate of $F > 0$ (cf. Fig. 2-3). Little energy will be available between the containers but the flow will bring a continuous supply of new e-acceptors which can diffuse into the concrete containers, and move through cracks in the concrete and holes in corroded steel containers. These e-acceptors can be consumed during respiration of organic waste. Fermentation processes will continue because the flow will prevent a build-up of toxic degradation products, causing a continuing microbial flow of degradation products downstream of the repository (cf. Fig. 2-5).

2.3 Repository content and possible microbial activities

The waste disposed of in the SFR consists of bitumen, concrete, metals, cellulose, fabrics, plastics, wood, rubber, insulation material, ion exchange resins, filter material, ashes, cement additions and miscellaneous material (Table 2-2). The microbial degradability of these materials varies significantly. In general, cellulose is the most easily degradable organic material in the SFR, followed by wood, fabrics and rubber. Bitumen has a low degradability in anaerobic environments and metals are not degraded at all. Ion exchange resins are generally difficult to degrade, and inorganic ion exchange matrices will not be degraded. Still, ion exchange waste may be a significant source of energy to microbes since they may have picked up organic components from the system the resins served in. Such material may be relatively easily degraded. This list should be expanded according to the material actually present, as shown in Table 2-2.

The degradation of waste material in the SFR is not harmful *per se*; rather, it is the degradation products that may cause problems, once degradation proceeds. Gas production and complexing agent formation may enhance radionuclide transport. These processes and others will be discussed in detail in Chapter 3. In contrast to waste degradation, microbial degradation of the waste containers themselves may indeed be harmful. Bitumen degradation may release solidified radionuclides, and concrete degradation may influence the mechanical stability of the containers and possibly lower the pH inside the containers, thereby increasing the prospect of radionuclide transport. Finally, microbial pitting corrosion may rapidly corrode holes into metal containers. These processes are discussed in Chapter 3.

3 Evaluation of the prospects of microbially induced processes in the repository

In this report, the transport of material to and from the microbes which will inevitably inhabit the SFR has been judged crucial to how they will perform. An evaluation of how microbial processes will progress in the SFR should, therefore, be correlated with flow scenarios. The cases discussed below will look at microbial processes in a stagnant situation and in various situations with a groundwater flow. It is very difficult to predict in detail what types of microbial processes may, or may not, develop in the SFR without support from experimental data. Such data are available to varying degrees, with a great deal of variation in the experimental conditions applied. The applicability of published data must therefore be carefully inspected before any conclusions may be drawn based on them.

3.1 Bitumen degradation

Various studies have been performed to test the microbial degradation of bitumen. In a book on nuclear waste disposal, edited by Wolfram et al.(1997), it is suggested that the microbial degradation of bitumen in the SFR will be a very slow or insignificant process. Roffey and Nordqvist (1991) report on aerobic and anaerobic degradation of bitumen with carbon dioxide production. They found aerobic and anaerobic rates of 0.6–1.5 $\mu\text{moles CO}_2 \text{ month}^{-1} \text{ mg}^{-1} \text{ bitumen}^{-1}$ and 1.1–1.5 $\mu\text{moles CO}_2 \text{ month}^{-1} \text{ mg}^{-1} \text{ bitumen}^{-1}$, respectively. Alkaline conditions somewhat reduced the degradation activity. Fungi were not detected although they have previously been reported to aerobically degrade bitumen (Ait-Langomazino et al. 1991). The conditions in the SFR are expected to be anaerobic and some investigations indicate a much slower degradation rate under anaerobic conditions. Wolf and Bachofen (1991) report that bitumen-degrading organisms are ubiquitous. The organisms these authors studied formed biofilms on the surface of the bitumen and degraded bitumen at a rate of 20–50 g bitumen $\text{m}^{-2} \text{ y}^{-1}$ under aerobic conditions. Anoxic conditions yielded a 100 times smaller degradation rate. Under anaerobic conditions, methane and carbon dioxide were formed.

Combining the figures in Roffey and Nordqvist (1991) with those of Wolf and Bachofen (1991) for a degradation time of 1 year yields the following result:

$$18 \text{ mmoles CO}_2 \text{ g}^{-1} \text{ bitumen}^{-1} \times 50 \text{ g bitumen m}^{-2} \text{ y}^{-1} = 0.9 \text{ mole CO}_2 \text{ m}^{-2} \text{ y}^{-1}$$

Under aerobic conditions, 20.2 L CO_2 per m^2 per year will be produced. With the anaerobic degradation rate (Wolf and Bachofen 1991), this number will be reduced to 0.2 L $\text{CO}_2 \text{ m}^{-2} \text{ y}^{-1}$. The gas production number (Roffey and Nordqvist 1991) was obtained under aerobic conditions and would be lower without oxygen. The 0.2 L $\text{CO}_2 \text{ m}^{-2} \text{ y}^{-1}$ can, therefore, be regarded as a conservative figure.

It is clear from the research cited and calculations above that bitumen can be degraded by microorganisms in the SFR once bitumen surfaces are exposed to groundwater. The degradation will, however, only proceed on bitumen surfaces once they become exposed to groundwater. Because of the lack of water (Table 3-1) no microbial activity will occur inside a solid block of bitumen. The degradation rate will be very slow under

stagnant flow conditions, but it will increase with increasing flow. Biofilms will form on surfaces exposed to flowing groundwater, producing carbon dioxide and methane. If solidified waste becomes exposed, it will likewise become degradable. Bitumen degradation will, however, most probably not occur as long as the steel/concrete containers (Table 2-2) remain intact, because they protect the bitumen from flowing groundwater. Bitumen degradation will, consequently, be connected with corrosion of the steel and concrete containers and with an establishment of a flow over the exposed bitumen surface. Taking all the above listed facts into consideration it seems safe to conclude that the degradation rate of bitumen in the SFR and the gas production connected to such degradation will be very slow, even from the 1000-year time perspective.

Table 3-1. Transport influencing properties of the packages

Solidification material	State	Water content	Porosity
Bitumen	solid	none	very low
Concrete/cement	solid/liquid	low	low
None	solid/liquid	high	high

3.2 Concrete degradation

Sulphuric, nitric and organic acid-producing bacteria may degrade concrete (Diercks et al. 1991, Rogers et al. 1997). The sulphuric and nitric acid-producing bacteria produce the acids during their litho-autotrophic oxidation of hydrogen sulphide and ammonia, respectively (see SKB TR 95-10, Sections 3.2.1 and 3.5.4). They are mostly known from aerobic environments, but anaerobic variants have been reported as well. They require ammonia and hydrogen sulphide for their acid-producing activity. Both these compounds are produced during the anaerobic metabolism of denitrifying and sulphate-reducing bacteria (SRB) (SKB TR 95-10, Sections 3.2.2 and 3.5.1). This reduction occurs during their consumption of organic compounds, which the SFR is rich in. The production in a two-step process of inorganic, concrete-degrading acids is, then, conceptually possible, if the conditions unexpectedly change to aerobic. More restrictions will, however, apply, making this a very unlikely scenario. First, the extremely alkaline conditions expected may inhibit these microbes and there would be the need for some kind of gradient conditions for this two-step reaction to evolve. Microbial degradation of concrete as a result of the production of inorganic acids will be more plausible in the outer parts of the SFR, where groundwater with circum-neutral pH meets the alkaline concrete repository, or in preferential flow paths that leach the concrete of alkaline components. Flowing groundwater would be a requirement. One such scenario may be concrete cracking as a result of anaerobic corrosion of solidified metal and armouring metals that will offer preferential flow paths for biofilm-forming litho-autotrophic microbes.

Organic acids can be produced by both bacteria (Diercks et al. 1991) and fungi (Perfettini et al. 1991). Anaerobic corrosion of iron and aluminium will produce hydrogen which is an excellent source of energy and electrons for microbes (SKB TR 95-10, Section 3.6). Acetogenic bacteria are common in deep groundwater (Haveman et al. 1999) and they produce acetic acid from organic compounds or hydrogen. Alkalophilic fungal strains have been found to increase cement porosity by 11.4% and reduce the bending strength by 78% through their production of gluconic and oxalic acids (Haveman et al. 1999). Direct contact between the fungal mycelia and the concrete is not necessary for effective dissolution of concrete to take place. These

acids are produced during the fungal degradation of organic components. Principally, fungi and bacteria will be present in most of the SFR material deposited and some may be able to produce organic acids. In contrast to the inorganic acid-producing microbes, the organic acid producers may be fermentative, which would reduce the requirement for transport of e-acceptors from outside the SFR. Organic acid production may occur inside containers once groundwater has invaded the containers. The pH, which initially is very high, may reduce such activity (Wenk and Bachofen 1995), but the absolute upper pH limit for microbial activity is unknown (SKB TR 95-10, Section 2.4 and SKB TR 97-22, Paper 11).

The prospect of concrete degradation will be different for the SFR sub-repositories. The BTF and BLA caverns with non-solidified waste will offer the best growth conditions for microbes, but they also contain the lowest levels of radionuclides. Waste in the BMA and SILO contains more radionuclides and is consequently better contained, with more concrete and bitumen. The bentonite barrier around the SILO will further restrict the possibility of a flow situation in this part of the SFR (except for the top lid part of the SILO).

3.3 Microbial corrosion of metal containers

It is known that bacteria may cause corrosion of metals. Biodeterioration of bitumen can in principle generate carbon dioxide, which in turn enhances steel corrosion. This would, however, be possible only inside steel drums containing a bitumen waste matrix, because normally, carbon dioxide is consumed by reactions with concrete in the repository. Inside a drum, it would be possible for microbes to generate carbon dioxide and lower the pH without the buffering effect of concrete. It is doubtful, however, that bitumen degradation will occur at significant rates in intact steel drums without the flow of water and the formation of biofilms on the bitumen surface, as discussed above (Section 3.1).

The major effect of abiotic corrosion processes in the SFR would be the gas produced. In the SFR, anaerobic, non-microbial corrosion is expected to totally dominate gas production (Roffey 1990). The only microbial corrosion process that may have an impact on the SFR would be pitting corrosion, commonly in conjunction with SRB. Local production of hydrogen sulphide in a biofilm on a metal surface may rapidly corrode a hole into thick metal. Steel casing failure in oil wells is one example where SRB cause severe technical problems. However, an oil well casing is exposed to strong flows of water and oil and offers excellent conditions for the growth of sulphate reducers.

Pitting corrosion of metals in the SFR would only occur on metal containers exposed to flowing groundwater that supplies the corroding microbial biofilm with sulphate and nutrients. The problem with pitting corrosion processes is that holes occur much sooner in a container than if an even corrosion process were to progress over the complete metal surface. The chain of unlikely events becomes long if microbial corrosion should damage the SFR integrity. A preferential flow path must occur in which SRB attach to and grow on an important metal surface barrier. Sulphate and organic material must be transported to this biofilm from the far field or from the waste, and pitting holes must corrode. The corroded holes must redirect the flow through a container where radionuclides become released (possibly by other microbes) and transported out of the container to the biosphere. Possibly, too, radioactive gas may escape out of a corroded

hole. None of the events in this chain of circumstances is conceptually impossible, but the chain of events is highly unlikely, unless strong groundwater flows become established through the SFR.

A rough calculation of the potential of corrosion caused by sulphide from SRB in the BLA cavern shows that a flow of 15 m³ groundwater per year with 100 mg SO₄²⁻ L⁻¹ (the freshwater period) would transport approximately 15 moles of sulphur to the waste, which can be reduced to 15 moles of sulphide. Assuming FeS formation on steel containers, this sulphide would be able to corrode 840 g, or 106 cm³ y⁻¹, iron. Over a 1000-year period, this would build up to 840 kg of iron, to be compared with the iron weight of one large BLA container, which is 2700 kg (Table 2-1). General corrosion caused by SRB does not seem to be a problem. Pitting corrosion is local, but will only occur if the bacterial production of sulphide is local, which again will only occur under restricted flow conditions where most of the 15 m³ sulphate-bearing groundwater per year transports over a very limited container area. In addition to sulphate, the groundwater must carry degradable organic compounds from other parts of the SFR or from the surface biosphere.

The reasoning above illustrates the difficulty to predict with high precision which microbial processes will develop in the SFR. Few processes are impossible, but the likelihood for them to develop in a certain chain of events will strongly depend on the environmental conditions that prevail. Again, transport-related factors, flow rate and transport regimes are vital for the proper evaluation of microbial processes that can develop in the SFR.

Table 3-2. The basic metabolic processes of hydrogen utilisation by various groups of bacteria in the presence of different hydrogen acceptors.

Electron and hydrogen acceptor	Metabolic process	Resulting compounds
H ₂ + oxygen	Aerobic respiration	Water
H ₂ + carbon dioxide	Carbon reduction cycle	Organic compounds
H ₂ + nitrate	Nitrate respiration	Nitrogen, nitrite
H ₂ + sulphate	Sulphate respiration	Sulphide
H ₂ + carbon dioxide	Acetate formation	Acetate
H ₂ + carbon dioxide	Methane formation	Methane
H ₂ + fumarate	Fumarate reduction	Succinate

3.4 Gas consumption

Hydrogen is expected to evolve in large quantities as a result of anaerobic corrosion of iron and aluminium. Hydrogen is an excellent source of energy for many microbes (SKB TR 95-10, Section 3.6) and can be reacted by microbes with an array of different compounds (Table 3-2). The resulting compounds may be sulphide, nitrogen, nitrite, methane, acetate and various other organic compounds. Many of the hydrogen utilisers use carbon dioxide as carbon source for their production of organic compounds. The concentration of free carbon dioxide will be limited under the prevailing alkaline conditions in the SFR, but can be locally significant if the groundwater flow leaches the concrete. A high hydrogen partial pressure may act as inhibitor on many hydrogen-utilising microbes, as accurately reviewed by Kidby and Rosevear (1997). The main concern for SFR performance, with respect to hydrogen consumption, would be the production of unwanted complexing agents, mostly short organic acids. The extent of

the processes in the SFR listed in Table 3-2 is very difficult to predict. They will, however, depend on the partial pressure of hydrogen and the availability of hydrogen acceptors, most of which would need to be renewed by transport processes to produce significant amounts of resulting compounds. The exception may be carbon dioxide, at least if the K_m 's (SKB TR 95-10, Section 2.1.2) for the present carbon dioxide fixation enzymes are small enough to compete with the high alkalinity.

3.5 Cellulose degradation

The SFR will eventually contain large amounts of cellulose in wood, cotton cloth, cardboard, paper, tissues, and most will be disposed of in the BLA and BMA (Tables 2-1 and 2-2). Cellulose is unstable under alkaline conditions and will degrade to water-soluble, low molecular weight compounds. In the presence of Ca^{2+} , a primary cation in cement pore water, (isosaccharinic acid) ISA, is the main degradation product (Van Loon et al. 1999). The degradation, the so-called "peeling reaction", does not come to completion because of stopping reactions that result in stable, alkaline end-groups. The products of alkaline cellulose degradation may act as complexing agents for radionuclides (Askarieh et al. 2000).

Microbes may degrade cellulose to carbon dioxide, or just convert it to complexants. The microbial production of gas will be further dealt with below, in the section on "Gas formation". In this process, ISA-degrading microorganisms have been reported, as summarised by Askarieh et al. (2000). Such microbial activity would be beneficial to the SFR as it would decrease the concentration of this complexing agent.

Microbial degradation of cellulose may lead to a number of possible problematic effects on the performance of the SFR. These processes, in which cellulose is the main energy source for the microbes, are discussed under the various headings below. A range of secondary effects may be triggered when microbes grow on cellulose and its degradation components.

3.6 Biogeochemical considerations

It is well known from land fill waste dumps that the plume of groundwater coming out from the waste has a composition much different to what went in, up the groundwater gradient through the waste (Christensen et al. 1994). Generally, the redox potential in the plume is much lower than it is in the ingoing groundwater and, depending on the type of waste, it carries reduced e-acceptors (see Fig. 1-3). Some of these may be beneficial to the SFR. Sulphide production will contribute to metal precipitation, including radionuclides that can react with sulphides, forming insoluble compounds. (Sulphide may also be detrimental, causing corrosion [see Section 3.3].) The formation of methane with ^{14}C -carbon or 3H hydrogen would, by contrast, add to radionuclide dispersion from the SFR. As concluded for most other microbial effects on the SFR, the extent of geochemical processes depends on the groundwater flow through the repository. Rough calculations can be made, taking the groundwater concentrations of electron donors listed in Figure 1-3 and recalculating them to the corresponding reduced compounds and carbon dioxide. The expected flow of groundwater over time and space can, then, be applied to find the maximum production of reduced e-acceptors and carbon dioxide via respiratory processes.

Fermentative processes can continue without external e-acceptors, but they require the removal of toxic compounds, as discussed in Section 1.2.1. Calculations of when

fermentative reactions stop as a result of toxic metabolites reaching a limiting level are more complicated, because the level varies from metabolite to metabolite. Generally, acids, alcohols and hydrogen may act as limiting factors, while carbon dioxide and methane production can go on until very high partial pressures have been reached. Such gas formation is further discussed in Section 3.11.

3.7 Formation of complexing agents

Microbial metabolism generates a large number of different compounds, many of which may act as complexing agents (SKB TR 95-10, Section 5.7.2-3). The breakdown of lignin and cellulose-containing material produces large macromolecular compounds with a wide range of complexing abilities (Birch and Bachofen 1990). Under optimal conditions and in the presence of oxygen, complex organic substrates are degraded via different metabolic pathways to the end-products carbon dioxide and water. In waste sites, including the SFR, sub-optimal conditions exist, viz. a low water potential, an anaerobic milieu, alkaline pH, and an imbalance in available nutrients. Under these conditions, metabolic pathways will be incomplete, facilitating the release of a variety of low molecular weight organic acids, alcohols and other anions (Table 3-3). It is clear from the literature (see, e.g., Birch and Bachofen 1990; Donick et al. 1996; Francis 1990; Gadd 1990; Ledin and Pedersen 1996; Ledin, Pedersen et al. 1996; Ledin, Rülcker-Krantz et al. 1996; Volesky 1994) that many different metal-microorganism interactions exist. The SFR is a complex system which is too complicated to predict in detail regarding the effects of microbial processes on radionuclide mobilisation and immobilisation. Until more detailed knowledge has been obtained, significant production of microbial radionuclide-complexing agents should be anticipated. They will become important in scenarios where groundwater-mediated transport is in focus.

Table 3-3. Examples of the complexing action of microbial by-products.

Products of microbial origin	Action
Tricarboxylic acids	Complexation of Mn
Citric acid	Formation of complexes with Pu (ten with citric acid, six with isocitric acid)
Catechol	– Formation of various actinides – Implicated in the uptake of germanium into microbial cells
Oxalate, salicylate, acetate, lactate, pyruvate and polypeptides	Solubilisation of uranium
Uncharacterised low molecular weight anion found in leachates from seepage trenches	Solubilisation of ⁹⁹ Tc, ⁶⁰ Co, ²³³ U
Uncharacterised low molecular weight ions produced by <i>Bacillus thuringiensis</i> in Mo-deficient medium	Complexation of Mo
Uncharacterised organic ligands in saline, alkaline lake water	Maintained elevated concentrations of normally insoluble Th ⁴⁺ , Pa ⁵⁺ , U ⁶⁺ , and Pu ions

For complete references, see Birch and Bachofen 1990.

3.8 Clogging of preferential flow paths

Biofilm formation strongly depends on the flow rate and also, the content of nutrients in the flowing groundwater (cf. Fig. 2-2). The largest potential for biofilm formation will consequently be in preferential flow paths. The potential for clogging of a preferential flow path will be higher inside the SFR vaults and downstream of the repository, where the concentration of nutrients for biofilm growth will be larger than in pristine groundwater. Biofilms are commonly composed of cells and their exudates, such as various polysaccharides. These extracellular compounds may precipitate dissolved inorganic solids which will add to the long-term clogging effect because they are not susceptible to the microbial degradation that may occur when the flow ceases. Permanent clogging will be the result. There are several possible outcomes of clogging of the preferential flow path. Firstly, until a new flow path becomes active, the flow will cease upstream. Hydraulic gradients will rule where the groundwater will flow. If new flow paths are formed and new biofilms develop mainly inside and downstream of the SFR, as suggested above, the network of clogging flow paths would eventually direct the groundwater flow outside the SFR, which would be beneficial. By contrast, a negative influence can be expected if a few flow paths through radionuclide-dense areas of the SFR remain open. As discussed earlier in this report, flow-directed transport will enhance microbial processes and their effects on radionuclide speciation in the SFR.

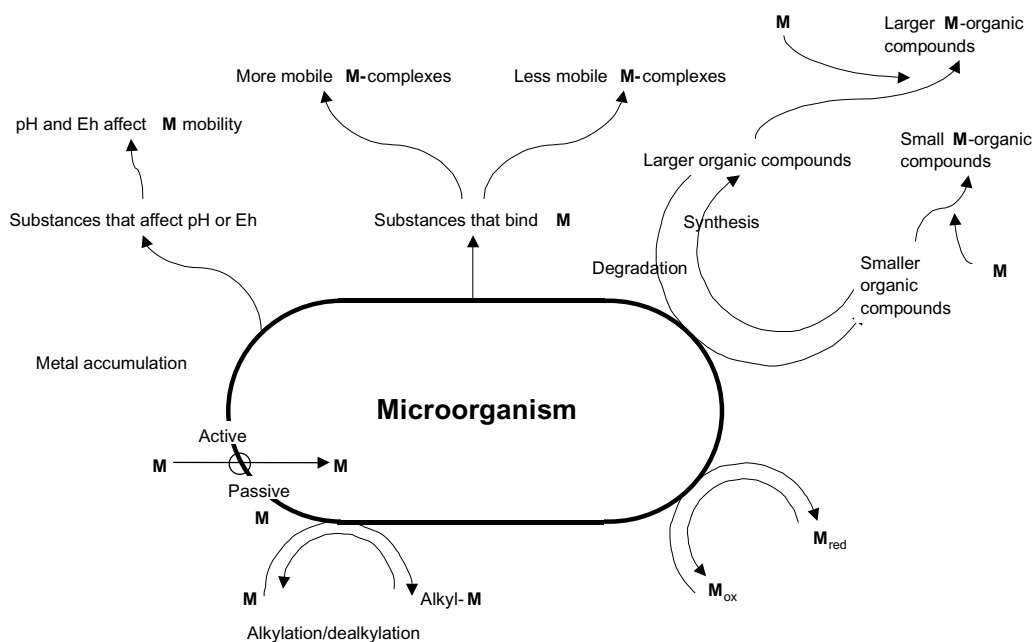


Figure 3-1. Principal ways in which microorganisms can influence metal mobility. (*M* = Metal)

3.9 Radionuclide dissolution and mobilisation

Several different situations may develop with respect to the distribution of radionuclides between mobile and immobile phases in the SFR (Fig. 3-1). Principally, similar situations may apply to both metal and non-metal radionuclides. Free-living microbes constitute mobile, suspended particles, which may have a radionuclide-sorbing capacity

greater than that of the surrounding environment. If the majority of the microbes grow in biofilms on surfaces, the transport of radionuclides may be reduced. The bacterial production of complexing agents has been discussed above. Local production of acids, as discussed under “Concrete degradation”, may dissolve radionuclides in addition to leaching the concrete (Fig. 3-2). It is plausible that microbes in the SFR dissolve and mobilise radionuclides, but this process will have an influence on radionuclide migration to the biosphere in relation to the flow situation through the SFR, with a flow in contact with organic, radionuclide-containing waste.

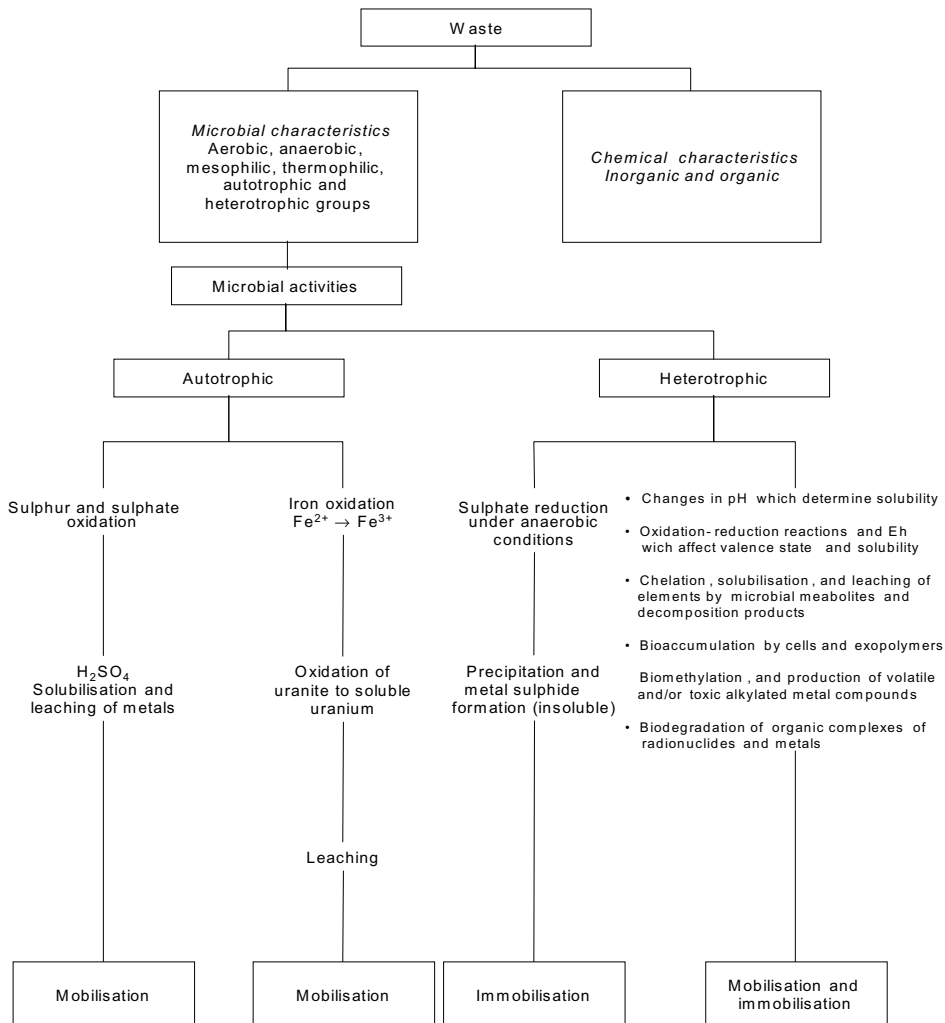


Figure 3-2. Microbial transformation of toxic metals and radionuclides.

3.10 Methylation and alkylation of radionuclides

The methylation and alkylation of radionuclides can be regarded as a special case of radionuclide mobilisation (Fig. 3-2). As part of their metabolism, microorganisms perform several different methylation reactions (Francis 1990). The radionuclide is not only mobilised; it is also made more toxic by the addition of organic groups. The

organic character of methylated and alkylated radionuclides makes them more bio-available and thus, more toxic. Such processes may proliferate in a batch mode (Fig. 1-1) until a stationary phase is reached. If a flow is generated in the vicinity, these toxic compounds may reach the biosphere.

3.11 Gas formation

Gas generation from anaerobic biodegradation of organic material is inevitable, as can be deduced from Figure 1-3. Some methane and carbon dioxide can also be produced, with hydrogen as the source of energy (see Section 3-4). The main gases produced are carbon dioxide and methane. While carbon dioxide may be adsorbed by the cement, methane will be inert. Once the saturation concentration is reached, however, free gas will either accumulate in pockets in the SFR or migrate upwards, through fractures. The amount of degradable organic material in the SFR is significant and if all were completely degraded to methane, large volumes of gas would escape from the SFR. A major concern, in addition to mechanical damage caused by high gas pressure, is that contaminated water could be forced out of containers and that ^{14}C and ^3H -containing gas would escape to the biosphere. Consequently, the prospect of vivid gas formation in the SFR is a very important aspect of waste management, which needs to be evaluated.

NIREX has invested large efforts in researching gas formation in underground repositories. A model, GAMMON, has been developed and laboratory and field-scale experiments have been performed. A series of reports describe this work (Agg, Billington et al. 1997; Agg, Purdom et al. 1997; Coutts et al. 1997; Kidby and Rosevear 1997; Macdonald et al. 1997; Purdom et al. 1997). The model approach describes gas generation by the coupled processes of metal corrosion (hydrogen) and microbial degradation of cellulosic materials to carbon dioxide, methane, hydrogen, hydrogen sulphide and nitrogen in low- and intermediate-level waste. Testing the model (Agg, Billington et al. 1997; Macdonald et al. 1997) has shown that moisture content is an important factor in that dry drums develop less gas than do drums with a higher moisture content. The addition of cement also reduced the gas production. There were some discrepancies between the model and the data obtained, but overall, the confidence in GAMMON has increased. The model predicts semi-batch type of gas production curves, with rapid production of gas, at the start and later, a slow exponential increase in the cumulative amount of gas generated. Data have been collected over a period of 4 years and the data obtained describe a typical batch growth gas production that stops at a specific cumulative gas level. The drums used were closed containers, so that the experiments were analogous to large batch cultures. Up to 140 litres of gas were produced in a single 220 litre drum over a period of 1239 days. The experiments suggest that microbial gas production will cease before a destructive gas pressure is reached. The reasons for this may be that toxic levels of metabolites formed, that key substrates were used up or that inhibitory hydrogen concentrations had evolved. The continuous production of gas in parts of the SFR will require transport processes that remove metabolites and replenish key substrates, nutrients and e-acceptors.

3.11.1 Gas formation in the repository

The problem of understanding the potential of microbial gas formation in the repository is very complicated. As previously mentioned, NIREX has invested much work in the development of the model GAMMON (Agg, Purdom et al. 1997) which predicts gas generation in low- and intermediate-level radioactive waste repositories (Agg,

Billington et al. 1997). A brief inspection of the GAMMON input parameters (see Appendix) reveals the great complexity of accurately predicting microbial gas generation in a waste repository. The SFR is as complicated in those respects as any NIREX facility will be. Consequently, a proper prediction of gas generation in the SFR will require a similar approach. The best approach would be to apply GAMMON to the SFR. It is not possible to make reliable predictions for a selection of the few parameters listed in the Appendix. Such predictions certainly would generate results, but the degree of precision would be extremely low.

4 Microbial processes in the SFR interaction matrix

The SFR interaction matrix contains many possible interactions between microbes and various other elements. Below follows a brief discussion on possible interactions and how they relate to each other.

4.1 Environmental effects on the microbial state

01.12b Waste/cement → Degradation of organics → Gas

The microbial degradation of organic material conditioned in concrete may produce gas. The first step may be alkaline hydrolysis of cellulose, followed by fermentation of monomers and oligomers to gas. The gases formed may be carbon dioxide, methane, hydrogen, hydrogen sulphide and nitrogen. The process is restricted by alkaline conditions and hydrogen produced by corrosion. This process interaction depends on 11.15.

01.15 Waste/cement → Microbial activity → Biological state

The types of waste and its composition will influence microbial activity and the growth of microorganisms in the waste containers. The most favourable position for microbes, with respect to available energy, is inside the containers. However, restricted availability of e-acceptors and the build-up of toxic degradation products may limit the types and amount of microbes that may proliferate inside the containers. This interaction depends on 11.15 and 15.05.

02.15 Waste/bitumen → Microbial activity → Biological state

Bitumen can be degraded by microbes and is therefore a possible substrate for microbial activity. The growth of microbes would predominantly occur on bitumen surfaces inside steel canisters, locally generating great numbers of microbes. This interaction depends on 11.15.

09.15 Vaults and backfill → Microbial activity → Biological state

The backfill and the space in vaults may offer less stagnant and alkaline conditions for microbes, compared with inside the containers and the SILO. However, the amount of available energy will be much less here than in the waste. A great amount of microbial activity is only expected if waste, or degradation products from the waste (including alkaline degradation products), reaches these areas. This interaction depends on 10.15 and 11.15.

10.15 Water composition → Microbial activity → Biological state

The amount of organic carbon, nutrients and e-acceptors in the groundwater interacting with the waste influences microbial activity. Of great importance is the availability of e-acceptors such as oxygen (not expected), nitrate, ferric iron, manganese(IV), sulphate and carbon dioxide. The SFR is rich in nutrients and energy and these components will not be limiting for microbial activity *per se*.

11.15 Water hydrology → Advection → Biological state

The magnitude, direction and distribution of groundwater flow in the different SFR components will influence the transport of microbes and, much more importantly, the transport of e-acceptors to, and degradation products from, microbes dwelling in various SFR locations. This interaction is judged to be the most important in relation to most other interactions involving microbes.

12.15a Gas → Microbial activity → Biological state

The amount of hydrogen and carbon dioxide is of importance for the state of microbes in the SFR. At high concentrations, hydrogen may act as an inhibitor and carbon dioxide is important for the activity of autotrophic microbes, such as methane-forming microbes. Oxygen is not expected except in the initial phase. Oxygen is a very potent e-acceptor and as a result, will rapidly be used up with microbial activity.

13.15 Temperature → Microbial activity → Biological state

Microbial activity generally increases with temperature in the interval between $-15\text{ }^{\circ}\text{C}$ and $+113\text{ }^{\circ}\text{C}$. The species distribution changes over temperature as growth of a single microbe generally takes place within as small an interval as $20\text{--}30\text{ }^{\circ}\text{C}$.

4.2 Effects of the microbes on the state of the repository

15.01 Biological state → Microbial growth → Waste/cement

Microbial growth is possible in the cement-solidified waste. The growth may be significant if an advective flow provides the microbes with e-acceptors, as well as removing degradation products and leaching the concrete. Under stagnant hydraulic conditions, the effect will be insignificant. Specific species of bacteria can produce sulphuric and nitric acid which may deteriorate cement and concrete.

15.02 Biological state → Microbial growth → Waste/bitumen

Microbial growth is possible in the bitumen-contained waste. Biofilms may form on the surfaces of the waste. The growth may be significant if an advective flow provides the microbes with e-acceptors and removes degradation products. The bitumen will be degraded at a slow rate if conditions are anaerobic. Degradation is much higher under aerobic conditions but under stagnant hydraulic conditions, the effect will be low. Steel-contained bitumen waste is most susceptible to this process.

15.03 Biological state → Microbial growth → Non-solidified waste

The non-solidified waste will provide the largest potential for microbial degradation. The organic carbon content is very high and the pH will be less alkaline than in cement-solidified waste. The large mobility of microbes and waste components inside a container suggests that gas production may become significant here.

15.04 Biological state → Microbial growth → Concrete packing

On the outside of cement containers, microbial growth is possible. The growth may be significant if an advective flow provides the microbes with e-acceptors, removes degradation products and leaches the concrete. Under stagnant hydraulic conditions, the effect will be insignificant. Specific species of bacteria can produce sulphuric and nitric acid which may deteriorate cement and concrete containers from the outside.

15.05 Biological state → Microbial growth → Steel packing

Microbial corrosion is a well-established process and pitting corrosion by sulphate reducing bacteria may rapidly corrode holes into steel containers. The growth may be significant in the form of pitting corrosion if an advective flow supports microbes locally with a supply of sulphate and organic substrates.

15.06 Biological state → Microbial growth → Concrete backfill

In the concrete backfill, microbial growth is possible. The growth may be significant if an advective flow supplies the microbes with e-acceptors, removes degradation products and leaches the concrete. The effect will be low under stagnant hydraulic conditions. Specific species of bacteria can produce sulphuric and nitric acid which may deteriorate cement and concrete.

15.07 Biological state → Microbial growth → Concrete structures

Microbial growth is possible on concrete structures. The growth may be significant if an advective flow supplies the microbes with e-acceptors, removes degradation products and leaches the concrete. The effect will be low under stagnant hydraulic conditions. Specific species of bacteria can produce sulphuric and nitric acid, which may deteriorate cement and concrete.

15.08 Biological state → Microbial growth → Bentonite barriers

Compacted bentonite with a density of $> 1800 \text{ kg/m}^3$ has been demonstrated to be unfavourable for the growth of microbes. The main constraints are the low water availability, low porosity and stagnant transport conditions, allowing only diffusive transport of nutrients, energy sources and degradation products. However, the SILO will have a compaction at 1000 kg/m^3 and the top and bottom will consist of 85–90% sand. The bentonite barrier will not restrict microbial activity. It is possible that the transport of microbes will be somewhat restricted in the pure bentonite, due to pore size limitations.

15.09 Biological state → Microbial growth → Vaults and backfill

Microbial growth in vaults and backfill may alter the hydraulic conditions, provided the growth is large. Biofilms may form and clog the preferential flow pathways. This process would probably be possible only downstream of the SFR in the “plume” containing dissolved material from the interior of the SFR, which can be used for significant growth.

15.10 Biological state → Microbial growth → Water composition

Microbial growth will produce a series of different degradation products that will alter the groundwater composition inside and downstream of the SFR. Dissolved carbon dioxide and methane, acids, ferrous iron, sulphide and complexing organic agents may form, while the amount of sulphate, nitrate, ferric iron, hydrogen and oxygen will decrease owing to microbial consumption.

15.12 Biological state → Microbial growth → Gas

The activity of microbes under anaerobic conditions may eventually lead to significant production of gas, mostly methane. Some hydrogen and carbon dioxide may be consumed during autotrophic methane production, but most of the methane will be liberated from the degradation of the organic wastes in the SFR.

15.13 Biological state → Microbial growth → Temperature

Microbial activity may, if vigorous, generate a great deal of heat. The SFR environment is, however, not so rich in easily degradable organic carbon and e-acceptors that it will influence the temperature.

15.16 Biological state → Microbial transport → Radionuclides and toxicants

Microbial activity may influence radionuclides in many different ways. The production of organic acids and other complexing agents may increase the mobility of radionuclides, as described in Section 15.10. The microbes may also act as sorbents for radionuclides and may mobilise or immobilise them, depending on whether they are attached or unattached. In addition, as part of their metabolism, microbes may change the chemical form of radionuclides. The methylation of metals, turning inorganic species to organic, the carbon fixation of $^{14}\text{CO}_2$ to organic carbon, and the degradation of organic ^{14}C -carbon to $^{14}\text{CO}_2$, is one example. These transformations will alter the mobility of the elements.

5 References

Agg P J, Billington R S, Gunn Y, 1997. Preliminary testing of the gas generation model GAMMON using data from drums containing low-level waste simulant. NIREX Report NSS/R299 1–43.

Agg P J, Purdom G, Wiblin D, 1997. GAMMON (version 1A): a computer program addressing gas generation in radioactive waste repositories. NIREX Report NSS/R338 1–102.

Ait-Langomazino N, Sellier R, Jouquet G, Trescinski M, 1991. Microbial degradation of bitumen. *Experientia* 47, 533–539.

Askarieh M M, Chambers A V, Daniel F B D, FitzGerald P L, Holtom G J, Pilkington N J, Hess J H, 2000. The chemical and microbial degradation of cellulose in the near field of a repository for radioactive wastes. *Waste Management* 20, 93–106.

Birch L, Bachofen R, 1990. Complexing agents from microorganisms. *Experientia* 46, 827–834.

Christensen T, Kjeldsen P, Albrechtsen H-J, Heron G, Nielsen P H, Bjerg P L, Holm P E, 1994. Attenuation of landfill leachate pollutants in aquifers. *Environmental Science and Technology* 24, 119–202.

Coutts D A P, Grant W D, O'Kelly N, Pugh S Y R, Malpass J R, Rosevear A, Walter E R, Widdowson D, 1997. Microbiological research in support of the NIREX near-field chemistry program. NIREX Report NSS/R328 1–58.

Diercks M, Sand W, Bock E, 1991. Microbial corrosion of concrete. *Experientia* 47, 514–516.

Donick A, Ledin M, Pedersen K, Allard B, 1996. Mechanisms of accumulation of zinc and cadmium by *Cytophaga johnsonae*. *Biometals* 9, 169–175.

Ekendahl S, Arlinger J, Ståhl F, Pedersen K, 1994. Characterization of attached bacterial populations in deep granitic groundwater from the Stripa research mine with 16S-rRNA gene sequencing technique and scanning electron microscopy. *Microbiology* 140, 1575–1583.

Ekendahl S, Pedersen K, 1994. Carbon transformations by attached bacterial populations in granitic groundwater from deep crystalline bed-rock of the Stripa research mine. *Microbiology* 140, 1565–1573.

Francis A J, 1990. Microbial dissolution and stabilization of toxic metals and radionuclides in mixed wastes. *Experientia* 46, 840–851.

Gadd G M, 1990. Heavy metal accumulation by bacteria and other microorganisms. *Experientia* 46, 834–840.

Haveman S H, Pedersen K, Routsalainen P, 1999. Distribution and metabolic diversity of microorganisms in deep igneous rock aquifers of Finland. *Geomicrobiology Journal* 16, 277–294.

Holmén J, and Stigsson M, 2001. Modelling of the future hydrogeological conditions at SFR, Forsmark. SKB Report R-01-02.

Kidby D W, Rosevear A, 1997. The microbial basis of the gas generation program GAMMON. Nirex Report NSS/R369 1–58.

Ledin M, Pedersen K, 1996. The environmental impact of mine waste – roles of microorganisms and their significance in treatment of mine wastes. *Earth-Science Reviews* 41, 67–108.

Ledin M, Pedersen K, Allard B, 1996. Effects of pH and ionic strength on the accumulation of Cs, Sr, Eu, Zn, Cd and Hg by *Pseudomonas putida*. *Water Air and Soil Pollution* 93, 367–381.

Ledin M, Rülcker-Krantz C, Allard B, 1996. Zn, Cd and Hg accumulation by microorganisms, organic and inorganic soil components in multi-compartment systems. *Soil Biology and Biochemistry* 28, 791–799.

Macdonald C, Ambrose D, Brewer A J, Campbell D J V, James L, Sopp C, 1997. Investigations into the potential rate, consumption and cumulative amount of gas generated from the biodeterioration of simulated low-level waste. Nirex Report NSS/R322 1–17.

Pedersen K. 1982. Factors regulating microbial biofilm development in a system with slowly flowing seawater. *Applied and Environmental Microbiology* 44, 1196-1204.

Pedersen K, 1990. Biofilm development on stainless steel and PVC surfaces in drinking water. *Water Research* 24, 239–243.

Pedersen K, Arlinger J, Ekendahl S, Hallbeck L, 1996. 16S rRNA gene diversity of attached and unattached groundwater bacteria along the access tunnel to the Äspö Hard Rock Laboratory, Sweden. *FEMS Microbiology Ecology* 19, 249–262.

Pedersen K, Ekendahl S, 1992. Incorporation of CO₂ and introduced organic compounds by bacterial populations in groundwater from the deep crystalline bedrock of the Stripa mine. *Journal of General Microbiology* 138, 369–376.

Perfettini J V, Revertagat E, Langomazino N, 1991. Evaluation of cement degradation induced by the metabolic products of two fungal strains. *Experientia* 47, 527–533.

Potts M, 1994. Desiccation tolerance of prokaryotes. *Microbiological Reviews* 58, 755–805.

Purdom G, Rosevear A, Scott P E, 1997. Preliminary testing of gas generation model using data from experiments in small bottles and columns. NIREX Report NSS/R345 1–75.

Riggare P, Johansson C, 2001. Project SAFE – Low and Intermediate Level Waste in SFR-1 – Reference Waste Inventory, SKB Report R-01-03.

Roffey R, 1990. The Swedish final repository and the possible risk of interactions by microbial activities. *Experientia* 46, 792–794.

- Roffey R, Nordqvist A, 1991.** Biodegradation of bitumen used for nuclear waste disposal. *Experientia* 47, 539–542.
- Rogers R D, Hamilton M A, Veeh R H, McConnell J, 1997** A procedure to evaluate the potential for microbially influenced degradation of cement-solidified low-level radioactive waste forms. In: Wolfram JH, Rogers RD, Gazso LG (eds). *Microbial degradation processes in radioactive waste repository and in nuclear fuel storage areas*. Dordrecht, The Netherlands: Kluwer Academic Publishers, pp 43–53.
- SFRI, 1993.** Förvaringskedet. SSR 8.0-1, 1993-05-14.
- Skogberg M, 2000a.** SFR-1 Årsrapport över deponerad mängd, aktivitetinnehåll och övriga ämnen, 1999. SKB drift PM 00/03 rev 1. 2000-03-07.
- Skogberg, M, 2000b.** SFR – Sammanställning av mängd avfall deponerat under 1999. SKB drift PM 00/02, 2000-02-03, reg No. DS 242.
- Stuwe K, 1999.** Diffusion. In: Marshall C P, Fairbridge R W (eds). *Encyclopedia of Geochemistry*. Dordrecht, The Netherlands: Kluwer Academic Publishers, pp 133–140.
- Van Loon L R, Glaus M A, Laube A, Stallonme S, 1999.** Degradation of cellulosic materials under the alkaline conditions of a cementitious repository for low- and intermediate-level radioactive waste. II. Degradation kinetics. *Journal of Environmental Polymer Degradation* 7, 41–51.
- Volesky B, 1994.** Advances in biosorption of metals: selection of biomass types. *FEMS Microbiology Reviews* 14, 291–302.
- Wenk M, Bachofen R, 1995.** Characteristics of anaerobic mixed cultures isolated from alkaline and oligotrophic habitats. *Geomicrobiology Journal* 13, 33–44.
- Wolf M, Bachofen R, 1991.** Microbial degradation of bitumen. *Experientia* 47, 542–548.
- Wolfram J H, Rogers, R D, Gazso, L G. 1997** *Microbial Degradation Processes in Radioactive Waste Repository and in Nuclear Fuel Storage Areas*. NATO ASI Series, 1. Disarmament technologies – Vol 11. Dordrecht, The Netherlands: Kluwer Academic Publishers.

Appendix

TABLES FOR SECTION D

Table D1 List of GAMMON Input Parameters

Line no. ^(a)	Parameter	Category ^(b)	Units	Program variable
3	Microbial growth rates	B	hr ⁻¹	MU(8)
5	Microbial maintenance energies	B	g g ⁻¹ hr ⁻¹	MAINT(8)
7	Microbial death rates	B	hr ⁻¹	DIE(8)
9	Microbial growth yields	B	g g ⁻¹ hr ⁻¹	YIELD(8)
11	Constants of saturation and inhibition for microbial model	B	kg m ⁻³	KIN(6)
13	Constants of saturation and inhibition for corrosion model	B	kg m ⁻³	RATE(7)
15	Initial pH	A	–	PH0
	pH flag	A	Integer	IFPH
17	Polynomial coefficients of pH-dependence function for non-methanogens	B	–	KPH(7)
19	Polynomial coefficients of pH-dependence function for methanogens	B	–	KPHM(7)
21	Microbial hydrolysis rate	B	hr ⁻¹	HYDMIC
	Natural hydrolysis rate	B	hr ⁻¹	HYDNAT
	Cellular nitrogen fraction	B	–	CELLNIT
	Non-degradable biomass fraction	B	–	FNDBIO
	Cellulose dissolution rate	B	hr ⁻¹	KW
	NH ₄ ⁺ release rate	B	hr ⁻¹	RAMON
23	Metal corrosion rates (mild steel)	B	m hr ⁻¹	MMU(3)
25	Metal corrosion rates (stainless steel)	B	m hr ⁻¹	SMU(3)
27	Initial concentration: mild steel plate	A	kg m ⁻³	FEPL
	mild steel sphere	A	kg m ⁻³	FESP
	stainless steel plate	A	kg m ⁻³	SEPL
	stainless steel sphere	A	kg m ⁻³	SESP
	Rust index	B	–	RIN

Table D1 (continued)

Line no. ^(a)	Parameter	Category ^(b)	Units	Program variable
29	Initial activity in waste:			
	$^{14}\text{C}/^{12}\text{C}$	A	–	C14C12
	$\text{H}^3\text{HO}/\text{H}_2\text{O}$	A	–	HTOH2O
	Specific activities of active gases:			
	$^{14}\text{CO}_2$	B	Bq kg^{-1}	ACTCO2
	$^{14}\text{CH}_4$	B	Bq kg^{-1}	ACTCH4
31	H^3H	B	Bq kg^{-1}	ACTHT
	H^3HS	B	Bq kg^{-1}	ACTHTS
	Half-life (^3H)	B	hr	TH3
	Half-life (^{14}C)	B	hr	TC14
	Release rate of ^3H :			
	from mild steel	A	$\text{Bq m}^{-3}\text{yr}^{-1}$	TSTEEL
	from Magnox	A	$\text{Bq m}^{-3}\text{yr}^{-1}$	TMAG
	from stainless steel/zirconium	A	$\text{Bq m}^{-3}\text{yr}^{-1}$	TZIR
	Duration of release of ^3H :			
	from mild steel	A	yr	TS
from Magnox	A	yr	TM	
from stainless steel/zirconium	A	yr	TZ	
33	Initial concentration of $\text{Ca}(\text{OH})_2$	A	kmol m^{-3}	CAOH
	Henry's constant (H_2S)	B	atm	HH2S
	Henry's constant (CO_2)	B	atm	HCO2
	Gas constant	B	$\text{dm}^3\text{atm K}^{-1}\text{mol}^{-1}$	RCONST
	Repository temperature	A	K	TDEG
	Reference volume ^(c)	B	m^3	VOL
35	Concentration of SO_4^{2-} in groundwater	A	kg m^{-3}	SULIN
	Concentration of NO_3^- in groundwater	A	kg m^{-3}	NITIN
	Rate of resaturation of repository	A	$\text{m}^3\text{m}^{-3}\text{hr}^{-1}$	RFLOW
	Rate of groundwater flow through repository	A	$\text{m}^3\text{m}^{-3}\text{hr}^{-1}$	GFLOW
	Resaturation concentration of H_2O	A	kg m^{-3}	SAT
37	Initial concentrations of active gases:			
	$^{14}\text{CO}_2$	A	kg m^{-3}	C14O2
	$^{14}\text{CH}_4$	A	kg m^{-3}	C14H4
	H^3H	A	kg m^{-3}	HT
	H^3HS	A	kg m^{-3}	HTS

Table D1 (continued)

Line no. ^(a)	Parameter	Cate-gory ^(b)	Units	Program variable
39	Integer switches: 1. mild steel plate concentration is non-zero (1) or zero (0) ^(d) 2. mild steel sphere concentration is non-zero (1) or zero (0) ^(d) 3. output data for gases in kg m ⁻³ (1) or mol m ⁻³ (2) 4. release rate of ³ H from mild steel is non-zero (1) or zero (0) ^(d) 5. release rate of ³ H from Magnox is non-zero (1) or zero (0) ^(d) 6. H ₂ O concentration is fixed (0) or variable (1) ^(e) 7. Groundwater flow non-zero (1) or zero (0) ^(e) 8. O ₂ concentration is fixed (0) or variable (1) 9. release rate of ³ H from stainless steel/zirconium is non-zero (1) or zero (0) ^(d) 10. stainless steel plate concentration is non-zero (1) or zero (0) ^(d) 11. mild steel sphere concentration is non-zero (1) or zero (0) ^(d)	A A A A A A A A A A A	Integer Integer Integer Integer Integer Integer Integer Integer Integer Integer Integer	SWITCH1 SWITCH2 SWITCH3 SWITCH4 SWITCH5 SWITCH6 SWITCH7 SWITCH8 SWITCH9 SWITCH10 SWITCH11
41	Initial concentrations: CO ₂ (gas), CO ₃ ²⁻ , H ₂ S (gas), S ²⁻ , acetic acid, butyric acid, NH ₄ OH, OH ⁻ , Ca ²⁺ and H ⁺	A	kmol m ⁻³	YST(1-10)
43	Initial concentrations: cellulose recalcitrant cellulose soluble polysaccharides glucose-type monomer	A A A A	kg m ⁻³ kg m ⁻³ kg m ⁻³ kg m ⁻³	YST(11) YST(12) YST(13) YST(14)
45	Initial concentrations: H ₂ O, O ₂ , NH ₄ ⁺ (available), NO ₃ ⁻ (in waste) and SO ₄ ²⁻ (in waste)	A	kg m ⁻³	YST(15-19)

Table D1 (continued)

Line no. ^(a)	Parameter	Category ^(b)	Units	Program variable
47	Initial microbial concentrations	A	kg m ⁻³	YST(20–27)
49	Initial concentrations: CO ₂ , CH ₄ , H ₂ S, N ₂ , H ₂ , VFA and acetate	A	kg m ⁻³	YST(28–34)
51	Mild steel (plate): initial plate thickness	A	m	YST(35)
	initial concentration of rust	A	kg m ⁻³	YST(36)
	initial concentration of magnetite	A	kg m ⁻³	YST(37)
53	Mild steel (sphere): initial sphere radius	A	m	YST(38)
	initial concentration of rust	A	kg m ⁻³	YST(39)
	initial concentration of magnetite	A	kg m ⁻³	YST(40)
55	Stainless steel (plate): initial plate thickness	A	m	YST(41)
	initial concentration of rust	A	kg m ⁻³	YST(42)
	initial concentration of magnetite	A	kg m ⁻³	YST(43)
57	Stainless steel (sphere): initial sphere radius	A	m	YST(44)
	initial concentration of rust	A	kg m ⁻³	YST(45)
	initial concentration of magnetite	A	kg m ⁻³	YST(46)
59	Initial concentration of NH ₄ ⁺ (bound)	A	kg m ⁻³	YST(47)
61	Number of variables for which derivative output is required	C	Integer	NDER
62 ^(f)	Variables for which derivative output required	C	Integer	IDERIV(I), I=1,NDER
64	Error limit (also initial time step for numerical solver)	C	–	ESCAL
66	First output time	C	yr	STARTIM
	Last output time	C	yr	ENDTIM
	Number of output times per decade	C	Integer	NPTS
	Integer flag showing optional output required (1) or not required (0)	C	Integer	IPRNT
68	Tolerance for domain checking	C	–	H1

Notes for Table D1

- (a) The line numbers take into account the existence of the title and descriptive alphanumeric strings.
- (b) A: system-dependent parameter, B: system-independent parameter, C: job-definition parameter.
- (c) Reference volume should be 1 m^3 .
- (d) Switch should be set to 1 initially; reset to 0 by program when appropriate.
- (e) In the special case of repository resaturation, Switches 6 and 7 should initially be set to 1 and 0 respectively; they are reset by program to 0 and 1 respectively when resaturation is complete.
- (f) N.B. no alphanumeric description line precedes this line of data values.