RISK ASSESSMENT AND CONTROL IN BIOTECHNOLOGY

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Biotechnology processes may result in a hazard to health or to the environment. The approach taken to the assessment and control of these potential hazards has been based on the fundamental principles of occupational hygiene. A brief description of the risk assessment and control strategies for handling naturally occurring or genetically modified organisms is given.

Biotechnology, Genetic Modification, Control Strategies.

INTRODUCTION

Biotechnology can be defined as the industrial or commercial application of living organisms and biochemical processes. As such it encompasses many age-old processes including the production of bread, cheese, wine and beer. More recently its scope has been extended to the production of antibiotics and vaccines through the culturing and fermentation of naturally occurring organisms. Within the last two decades, biotechnology has been revolutionised by the introduction of the techniques of genetic modification (genetic manipulation or recombinant DNA). These new techniques have resulted in the commercialisation of new products that previously either could not have been obtained or only obtained in insufficient yield or purity. Examples include pharmaceutical products such as human insulin, growth hormone and vaccines such as that against Hepatitis B. Genetic modification is also of importance to the antibiotic, food and drink industries as it gives the possibility of improving the performance of 'traditional' process organisms. The arrival of genetic modification has however focussed attention on the hazards to health and the environment from biotechnology. This has highlighted the need to ensure that the hazards from the organisms used in biotechnology processes are adequately assessed and controlled.

BIOTECHNOLOGY PROCESSES

A typical biotechnology process involves a number of stages; inoculum preparation, large-scale growth of the organism in a bioreactor, separation of the organism from its growth medium and product purification. These stages are summarised in Figure 1.

Inoculum preparation is the stage at which the required organism in the correct state and quantity is prepared to start the large-scale phase. Depending on the scale of this large scale phase it may be preceded by growth in successive stages. For example successful growth on a 50,000L scale may require preceding stages of 5 and 500L. The large scale growth phase in a bioreactor allows organisms to grow under conditions which are controlled so as to maximise the yield of desired product. Most processes are carried out on a batch basis but a number of continuous processes have been developed. The first stage in downstream processing is usually the separation of the organism from the medium in which it has been grown. The organism may be inactivated at this stage. The organism or the medium may then be processed further to obtain the desired final product.

RISK ASSESSMENT

In contrast to conventional chemical processes a biotechnology process operates under 'moderate' conditions. Organisms are typically grown in dilute solutions under moderate pH, at close to ambient temperature and pressure and have by-products that are usually biodegradable. Consequently the major hazards of the conventional chemical industry, including exothermic reactions, large inventories of flammable or toxic materials, high temperatures, pressures, and toxic by-products are usually avoided (Russel(1)).

The organisms used in biotechnology processes may however pose a number of potential health hazards including; infection hazard (pathogenicity), toxic, allergenic or other biological effect of the organism or cells, its components or its naturally occurring metabolic products and toxic, allergenic or other biological effect of the product expressed by the organism. Exposure can be by inhalation, ingestion, inoculation or transmission through broken skin and mucous membranes. The effect on health depends upon the route of entry, the organism, the dose received and the status of the exposed person's immune system.

As in any industrial process the risks to health from handling living organisms and their products must be evaluated and the necessary action taken to remove or reduce those risks. The chemical industry is well used to assessing the risks to health from chemical processes and deciding on appropriate controls. When handling conventional chemical agents compliance with exposure limits may be one indication of whether everything has been done so far as is reasonably practicable to control exposure. In many of the processes involved in biotechnology there are few relevant exposure limits and there are none that apply to living organisms. There is a lack of adequate, quantitative data on the risks to health from these biological agents and their products. In the absence of such data the Health and Safety Executive (HSE) and the Advisory Committee on Genetic Modification (ACGM) and the Advisory Committee on Dangerous Pathogens (ACDP) have used the general principles of occupational hygiene to develop principles of assessment and control for biological agents. For laboratory work the ACGM(2) and the ACDP(3) have specified four hazard categories, together with corresponding containment levels. Although ACGM and ACDP have reached a similar end point in terms of laboratory containment, the methods of risk assessment have been rather different.

In addition ACGM have defined assessment considerations for work on a large scale which are applicable to research and development work as well as commercial manufacture.

ACDP have categorised microorganisms into one of four hazard groups, based on a assessment of the available data on; pathogenicity of the organism to man, hazard to laboratory workers, transmissibility in the community, and availability of effective prophylaxis or treatment. Although ACDP have not considered large scale operations specifically the assessment of risk for such work should take into account both the suggested hazard category and factors such as the virulence and transmissibility of the microorganism as well as the titre or concentrations handled and the operations undertaken. In addition there is a need to consider any allergenic or toxic properties of the organism or its products.

In contrast ACGM has assessed individual experiments involving genetically modified organisms using a ranking system which is based on the assignment of risk values (negative exponentials) for each of the factors; access or probability of the organism colonising the body, expression that is the anticipated or known level of expression of inserted genes and damage, the Ill-health effect due to the gene product of the genetically modified organism. The product of these factors gives the overall risk value for the experiment which is then related directly to the appropriate containment level. When genetically modified organisms are used on a large scale it differs from most laboratory work in that the organism used and any product is usually defined prior to the start of production. In addition to considering the risk assessment for the construction of the genetically modified organism in the laboratory additional factors must be considered and are described by ACGM(4). These include the genetic stability of the organism as well as the consequences of the organism entering the environment.

The Hazard categories for Biological agents are summarised in Table 1.

TABLE 1 - Hazard Risk Categories for Biological Agents

Laboratory Scale		Large Scale
ACGM	ACDP	ACGM
1	1	Good Large Scale Practice
2	2	Large Scale 1
3	3	Large Scale 2
4	4	Large Scale 3

Organisms in Group 4 and Large Scale 3 may cause severe human disease and are a serious hazard to workers and possibly the general public. Organisms assigned to Hazard Group 1 (ACDP and ACGM) and those meeting the criteria for good large scale practice (GLSP) are not normally associated with ill-health in man. Some organisms, whilst not representing a threat to man, may be pathogenic to plants and animals with the potential for causing heavy economic losses. These and other environmental concerns need to be taken into account during risk assessment.

The vast majority of biotechnology processes use organisms that do not present a pathogenic or toxic risk, they may however pose an allergenic risk. In many instances the most serious risk to health is from the product. Consequently the distinct unit operations of a biotechnology process are usually assessed individually, and the characteristics of each operation dictate the physical containment required.

CONTROL MEASURES

Guidelines on the control measures required for large scale processes using genetically modified organisms have been published by HSE and ACGM. Little or no guidance advising on the precautions necessary for large scale processes using 'traditional' organisms is available. However as the health hazards from genetically modified organisms are of the same nature as those from 'traditional' organisms it is possible to apply the same principles of control.

Biotechnology processes can potentially lead to organisms contaminating equipment, room surfaces and air as well as entering the environment outside the workplace. These 'releases' can occur either as a result of routine procedures or following an accident. Consequently precautions are necessary to minimise the hazard to workers, the general public and the environment. These include engineering controls to provide primary and secondary containment to a level appropriate to the nature of the risk, together with good operator technique and good safety management. A further objective of these control measures may be to ensure adequate product quality. The majority of biotechnology processes warrant only conditions of minimal containment. For organisms meeting the GLSP criteria and others of equivalent low risk then the controls required may involve no containment measures beyond those required for process needs. For this group of organisms however, as well as for all other hazard groups, certain fundamental principles of good occupational and environmental safety have been described by the Organisation for Economic Cooperation and Development(5).

Fundamental Principles of Control

Exposure. Workplace and environmental exposure to organisms, their components or products should be kept as low as reasonably practicable.

Controls. Engineering controls should be exercised at source, with appropriate personal protective equipment provided when necessary. Such control measures must be adequately maintained, examined and tested.

Monitoring. The workplace and the environment should be monitored, as appropriate, for the presence of process organisms.

Training. Suitable instruction and training on the risks to health and the precautions which need to be taken should be given to operators.

Local Rules. Local rules for the protection of operators and the environment should be formulated and implemented.

Elimination or substitution of the hazard are primary objectives. If possible a less hazardous organism or strain should be used for the process. Additional emphasis to considering elimination or substitution may be given when the capital, maintenance and operational costs of running a facility with the standard of containment required for high hazard organisms are considered. This concept of 'biological containment' has been applied with some success to large-scale work involving genetically modified organisms. It involves using well-characterised, disabled or "genetically crippled" organisms that have been specifically developed to minimise any hazards to man or the environment if physical containment is breached. If the hazard cannot be eliminated then emphasis is placed on control at source through good engineering design of the processing equipment. The provision of protective equipment for the operator must be regarded as the last resort. These are certainly not novel ideas, but embody traditional occupational hygiene principles which have been applied for many years to chemical agents. Primary containment is intended to protect personnel and the facility from exposure to the organism being handled, and consists of the immediate physical barriers to release. In the case of a bioreactor this could include the provision of appropriate seals and gas filters to contain the fermentation process. Secondary containment is intended to protect the environment external to the laboratory or process plant from exposure to the organism being handled. This could include the provision of appropriate waste treatment and room air filtration.

Hierachy of Control Measures

The hierachy of measures for preventing or controlling exposure to biological agents and their products is summarised in the Approved Code of Practice for the Control of Substances Hazardous to Health Regulation 1988(6).

Elimination. Exposure can be prevented by eliminating the use of the hazardous organism or substance by for example work with gene fragments of the AIDS virus not the whole virus

Substitution. If use of the organism cannot be eliminated then it may be possible to use biologically disabled strains such as Escherichia coli K-12 in place of the wild-type.

Enclosure. Exposure can be controlled by using enclosed equipment, for example closed fermentation systems or class III biological safety cabinets.

Local Exhaust Ventilation. If complete enclosure is not practicable then local exhaust ventilation such as Class I and II biological safety cabinets may be appropriate.

Reduction of Exposure. Good operator technique, including appropriate disinfection and waste control policies are an essential part of reducing potential exposure.

Personal Protective Equipment. Personal protective equipment includes protective clothing, respiratory protective equipment and eye protection, as appropriate.

Hygiene Facilities. Providing handwash basin, and shower facilities where appropriate.

A range of control measures may be required to control the hazards from a particular organism. A fundamental feature of the approach taken to control is the importance of good operator technique. This includes following good hygiene practices such as no eating or drinking in the containment facilities. Adequate training in techniques to prevent aerosol formation, including the correct use of equipment is also essential.

Examples of containment approaches for handling living organisms in biotechnology processes are given in Table 2. For GLSP organisms and those of equivalent low risk the controls required will vary from industry to industry. As an example whilst both the brewing and pharmaceutical industries may use organisms of intrinsically low risk in certain processes, different containment and hygiene measures may be required.

TABLE 2 - Examples of Containment Approaches for Biotechnology Processes other than for GLSP and Equivalent Low Risk Organisms

	Large Scale Containment Categories 1	2	3
Primary Containment			
Process physically separated from workplace	Yes	Yes	Yes
Treatment of process exhaust gases	Minimise release	Prevent release	Prevent release
Sample collection/material addition/ organism transfer	Minimise release	Prevent release	Prevent release
Equipment Seals	Minimise release	Prevent release	Prevent release
Secondary Containment			
Controlled Area	Optional	Yes	Yes and purpose built
Ventilation: mechanical	Optional	Yes	Yes
negative pressure	No	Yes	Yes
exhaust air HEPA filtered	No	Yes	Yes
input air HEPA filtered	No	Optional	Yes
Process Spillage Containment	Yes	Yes	Yes
Facility Sealable for Fumigation	No	Optional	Yes
Process Effluent Treatment	Treated	Inactivated	Inactivated
Sink/Shower Effluent Treatment	No	Yes	Yes

During the initial downstream processing stages living organisms may be present for example during centrifugation and homogenisation. For these parts of the process a similar approach to that adopted for the assessment and control of the risks during growth in the bioreactor should be used. The subsequent extraction, purification and finishing stages either use well established industrial techniques or developments of these techniques. These include dialysis, chromatography as well as filtration, blending, filling and freeze or spray drying. The approach taken to the assessment and control of the risks to health from these stages of a biotechnology process is identical to that used in the conventional chemical industry.

Product Protection

An important difference between biotechnology processes and conventional chemical processes is the need in most situations to ensure sterility of the process equipment. That is not only must the worker and the environment be protected from the organism being handled but the bioreactor may need to be protected against contamination by other organisms present in the workplace. The level of protection against contamination is dependent upon the process, with for example brewing only requiring a hygienic environment whereas the production of pharmaceuticals may require a sterile environment. In many cases any measures taken to ensure sterility of the process may also increase the level of containment. However it may be necessary to give careful consideration to the design of the secondary containment to ensure that the bioreactor or downstream processing area remains aseptic whilst also protecting the areas outside this part of the facility from exposure to organisms or product.

CONCLUSIONS

The approach taken to the assessment and control of the risks to health from biotechnology processes has been based on the fundamental principles of occupational hygiene. An important feature of the control strategies used by the industry is 'biological containment' that is the elimination or substitution of hazardous organisms with organisms that have reduced infective, toxic or allergenic risks. This, together with the 'benign' conditions that are required for the growth of most living organisms used in biotechnology means that the major hazards (including the use of toxic chemicals, fire and explosion risks) associated with conventional chemical processes are avoided. Some concern remains in the public mind however, over the hazards to man and to the environment, in particular from the increasing commercialisation of the products from the 'new' biotechnology of genetic modification. In order to ensure that public confidence in the biotechnology industry is maintained it is important that any risks to worker, public and environmental safety continue to be adequately assessed and controlled.

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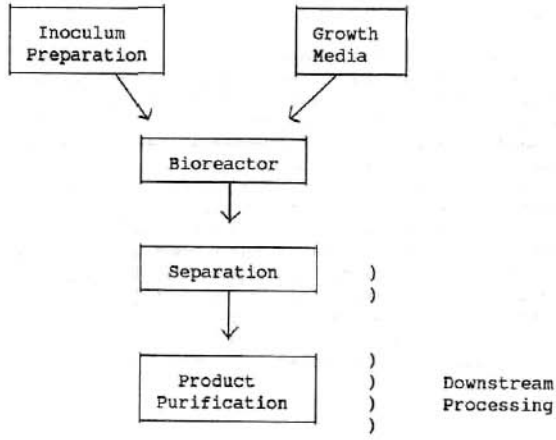

Figure 1 Typical Stages of a Biotechnology Production Process.