THIRD REVIEW CONFERENCE OF THE PARTIES
TO THE CONVENTION ON THE PROHIBITION OF THE
DEVELOPMENT, PRODUCTION AND STOCKPILING
OF BACTERIOLOGICAL (BIOLOGICAL) AND
TOXIN WEAPONS AND ON THEIR DESTRUCTION

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BACKGROUND DOCUMENT ON NEW SCIENTIFIC AND TECHNOLOGICAL DEVELOPMENTS
RELEVANT TO THE CONVENTION ON THE PROHIBITION OF THE DEVELOPMENT,
PRODUCTION AND STOCKPILING OF BACTERIOLOGICAL (BIOLOGICAL) AND TOXIN
WEAPONS AND ON THEIR DESTRUCTION

Prepared by the Secretariat

1. In paragraph 23 of its report (BWC/CONF.III/1), the Preparatory Committee
for the Third Review Conference of the Parties to the Convention on the
Prohibition of the Development, Production and Stockpiling of Bacteriological
(Biological) and Toxin Weapons and on their Destruction decided to request
each of the Depositary Governments, as at the First and Second Review
Conferences, to submit to the Review Conference information on new scientific
and technological developments relevant to the Convention. The Committee
further decided to invite States parties who wished to do so to communicate to
the Secretary-General of the United Nations their views on new scientific and
technological developments relevant to the Convention. In both cases, the
Committee stated, this information should cover the applications being made of
new scientific and technological developments and their relevance to the
various aspects of the Convention. The Committee also decided that this
background documentation should be circulated not later than two weeks before
the opening of the Review Conference.

2. The present document contains the information provided by Depositaries
and other States parties to the Secretariat, as of 26 August 1991, pursuant to
paragraph 23 of the Preparatory Committee's report.

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Australia

Impact of recent advances in science and technology on the Biological Weapons Convention

In the period since the Second Biological Weapons Convention Review Conference in 1986 the continued exploitation of biotechnology in the fields of medicine and agriculture has lead to major advances in the production, harvesting and preservation of micro-organisms and plant and animal cells. National capabilities for the large-scale production of micro-organisms and cell products have increased as the industrial potential in the exploitation of biotechnology has been recognized. While this has been beneficial from the point of view of public health and agriculture, it has also the potential, if misused, to provide the expertise and experience needed for developing and producing BW agents.

Production

Cell culture

In recent years there has been a major commitment to developing new animal cell culture methods that increase cell density and product concentration. Many complex biological compounds of therapeutic interest such as some hormones and antibodies cannot be produced in bacterial fermentation, even using genetic engineering techniques, because these proteins require glycosylation or other post translational modifications for proper activity. They must therefore be produced in animal cells. Viruses, being obligate parasites also must be grown in cell culture and with increasing vaccine production, large-scale virus production is needed. Until relatively recently the normal method of choice for growth of animal cell lines was as a monolayer on a solid surface in roller bottles. The process is labour intensive and requires frequent aseptic changes of growth medium. As the need for large-scale production of cells lines increased, many ways have been found to intensify animal cell growth. Growth of cells on the surface of beads suspended in growth medium has enabled the scaling up of production to stirred tank reactors of several hundred litres capacity. Yield has been improved still further by replacing the beads with porous microcarriers which will support higher cell densities per unit of reactor volume, can give more than 20 times greater cell yield compared with simple airlift reactors and have the added advantage of protecting cells against shear in agitated culture. Hollow fibre perfusion systems have been designed to overcome the problems with
substrate concentration gradients that can occur with immobilized cells. In these systems the cells are maintained within fibres and the nutrients and products pass freely through pores in the fibres. This gives a very high cell density, eliminates problems of washout and can give simultaneous separation of product and cells, in the case of soluble products. In addition serum free and low protein growth media are becoming more widely available and more versatile in the range of efficacy. This substantially reduces the cost of media required.

These new techniques simplify virus production and allow large yields from relatively small facilities.

Fermentation technology

Fermentation technology has improved dramatically in recent years. Automation of process control not only enables optimization of production, it also reduces the manpower required to operate large-scale fermenters. The inclusion of implant cleaning systems has also reduced the labour requirements and has allowed much more rapid turn-around time which also increases overall productivity. The development of continuous flow fermenters has enabled the size of a fermenter to be reduced approximately 1,000 times over conventional batch fermenters to give equivalent production. Cross-flow filtration can be used for continuous separation of inhibitory metabolites from cells, thus giving greater cell productivity. These improvements have had the effect of increasing the yield of cells or secondary metabolites (antibodies, toxins) and of increasing productivity so that more can be produced per unit volume per unit time or alternatively smaller units can be used.

These techniques are now incorporated into commercially available production technologies which have brought biotechnology industries into more widespread use. They simplify production and reduce the cost of cell products such as proteins and viruses and can, if misused, make BW production more feasible.

Harvesting

As the size and productivity of fermenters has increased, so has the need for improved harvesting systems. Continuous flow centrifuges are much faster than batch centrifuges and are commonly used for harvesting viruses. This equipment is, however, expensive and labour intensive. The use of cross-flow membrane filtration is being investigated as an alternative. This process has a lower labour requirement and, additionally, does not generate aerosols, as
centrifuges can. There is still a problem with membrane fouling but this is likely to be overcome in the near future.

Dissemination and delivery

Reaction against the adverse environmental effects of chemical pesticides has resulted in considerable research into biological control of agricultural pests. Many of the problems encountered in applying biological control are similar to problems that would be encountered in BW. Agents are readily inactivated by exposure to air, or to ultraviolet light (sunlight) or by desiccation. Methods utilizing microencapsulation of agents have helped to overcome these problems. Microencapsulation involves covering particles or droplets or an organism or toxin with a thin but very protective coat which will release the agent when it reaches a target environment (e.g. inner lung). The size and the smoothness of microcapsules can be controlled so that they will disperse well and carry properly through or with the air to their target. Such control would also allow easier, standardized weaponization or spraying methods for a range of agents. The inclusion of UV protective pigments such as those that have recently been discovered in marine soft corals and sponges would provide further protection of aerosolized micro-organisms. The dissemination methods for biological pesticides have also improved, with the development of low-volume spraying devices and jet engined sprayers. These methods are as applicable to BW agents as to microbial pesticides.

Genetic engineering

The major impact genetic engineering has had that is relevant to the BWC is the possibility of large-scale production of toxins. Most potent toxins have formerly been available only in very small quantities, and then only upon isolation from vast amounts of biological material. The cloning of the genes coding for production of toxins into micro-organisms has enabled production of kilogram quantities of these toxins under controlled conditions in fermenters. The developments in fermentation technology mentioned earlier have made it possible to utilize relatively small facilities for production.

It is possible that genetic engineering could be used to increase the resistance of potential BW agents to antibiotics, UV or to desiccation although it is more likely that resistance to the latter two factors would be provided by the formulation used for delivery of the agents. The scenario of producing more pathogenic BW agents by genetic engineering is unlikely.
Organisms evolve capabilities based on the environment in which they develop. Insertion of new genes into an organism is likely to prejudice its survival. There is no guarantee that the inserted gene will be expressed in the new host, no way of preventing the possibility of mutations after release which would thus circumvent vaccines used to protect an aggressor and no easy way to test the organisms prior to use.

Conclusions

The recognition of the range of novel, diverse and complex products of therapeutic and agricultural value that can be produced from microbial, animal and plant cell culture has brought about many changes that have direct relevance to the Biological Weapons Convention. The commercial value of these products has resulted in the widespread acquisition of national capabilities in biotechnology. Coupled with this is the rapid improvement in the technology so that the level of skill and the manpower required to operate the newer automated fermenter systems has been reduced as has the size of the facilities needed to produce useable quantities of products. As a result, many nations now have capabilities in the field of biotechnology, capabilities that can, if misused, be applied to BW production.

Canada

Canada is pleased to provide the document entitled "Novel Toxins and Bioregulators: the Emerging Scientific and Technological Issues Relating to Verification and the Biological and Toxin Weapons Convention". 1/

The document itself, in its preface on page (iii), contains a traditional disclaimer. However, this should be understood to apply only to any references therein relating to policy issues, as contrasted to scientific and technological developments.

Czechoslovakia

The CSFR is aware of new scientific and technological developments which, if they fall into the wrong hands, could be misused for the production of biological and toxin weapons. For this reason we consider it vital to closely monitor any future developments, especially in the fields of molecular biology

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1/ A limited distribution of this document in English only has been made available to States parties and observers to the Third Review Conference. Additional copies are available from the Permanent Mission of Canada at Geneva.
and biotechnology and their possible applications for BW purposes. In this respect we emphasize the importance of the provisions of Article X of the Convention. It is worth mentioning in this respect that the Second Review Conference urged that cooperation under Article X should be actively pursued both on bilateral and multilateral levels.

Denmark

With respect to communication of views on new scientific and technological developments and their applications relevant to the Convention, Denmark has no specific comments to offer, but wishes, however, to draw attention to the text of Article 1 of the Convention – where in subparagraph 1 it is stated: "... microbiological agents, or toxins whatever their original or method of production".

It seems that the text of this subparagraph fully covers all substances – including those eventually produced by modern biotechnology if used for the purpose of biological warfare.

Denmark welcomes, however, a discussion also on this subject at the Third Review Conference to be held in Geneva, 9-27 September 1991.
Background Paper on
New Scientific and Technological Developments

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Summary and Conclusions

There has been a rapid progress in many areas of molecular biology and biotechnology during the period 1986-1991. Using molecular biology, mechanisms of virulence and infection have been identified and the same techniques may also permit deliberate manipulations of these mechanisms. Thus, there is a potential danger that new or genetically modified BW agents may be created. There are very few new infectious agents that have been discovered and there has been some progress in the area of slow infection. The first field trials with genetically modified microorganisms have been carried out and this will also provide data for construction of models for the spread of microorganisms in the environment. As the techniques of molecular biology have advanced, projects, which previously were believed to be unrealistic, have now been initiated. Perhaps the most challenging of these is the Human Genome Project. It has the aim to sequence the complete human genome and thereby gain knowledge of all human genes, which will have wide implications for diagnosis and treatment of hereditary diseases. The vast knowledge gained in this area may also have applications in the area of biological and toxin weapons in the future. There has been a rapid development in the techniques for detection. Work is in progress for development of rapid detection equipment. Rapid progress has also been seen in the area of identification. One example is the use of monoclonal antibodies but also development of new techniques, such as the polymerase chain reaction, biosensors, as well as of probes for RNA hybridization, all of which have been shown to have a great potential for increasing sensitivity. Human pathogens previously undetected have thereby been characterized, for example hepatitis C. Potent antiviral and antibacterial agents, some of which have broad spectra, have been developed. Potentially useful methods for antimicrobial therapy are studied such as the use of ribozymes and of anti-sense RNA. Recently, "humanized" monoclonal antibodies have been developed. Molecular biology has allowed improved characterization of the immune defence, e.g. cytokines. Development of component and recombinant vaccine is a promising area although only a few vaccines have been registered lately. The area of process design for production of microorganisms and biochemicals has also progressed. Achievements in the area of protein engineering and catalytic antibodies have opened up new possibilities. More cost-effective
large scale culture systems for production of all kinds of biochemicals and microorganisms in more modest sized plants.

One of the problems with molecular biology research is that microorganisms created or manipulated in civilian research may be used as biological warfare agents. It also implied an increased technical potential to produce harmful molecules or organisms for use as biological or toxin weapons. The major technical innovation during the period is the PCR technique with applications mainly for identification. In comparison with 1986 there has been a further progress in the techniques mentioned previously. During this period the volume of R & D has significantly expanded. The commercial exploitation of biotechnology has become a reality and major multinational companies in biotechnology and pharmaceuticals are using the new techniques. It can also be noted that most nations now have, or are initiating national R & D programmes in the area of biotechnology. Even if most of the research has been directed towards improved protection compared with for example studies of enhanced virulence, it could possibly also lead to improved offensive capabilities. The background document of the Second Review Conference (BWC/conf II/4/, August 18th, 1986), stated that rapid technical developments had taken place between 1980-86. As previously mentioned, progress has been at least as rapid during the period 1986-1991. Sweden still considers the relevant conclusions of the 1986 report* to be valid. In the view of Sweden the analysis of the scientific and technological developments during the period 1986-1991 shows that it is still covered by Article I in the BWC. However, the number of potential B agents, toxins or other toxic biochemicals, has continued to increase as well as the possibilities to modify or create new agents. The border between defensive and offensive research has become more indistinct and the time from research to industrial application is becoming shorter. Due to this, it is important to continuously monitor research and development in this and related areas.

* Background document of new scientific and technological developments relevant to the Convention on the Prohibition of the Development, Production and Stockpiling of Bacteriological (Biological) and Toxin Weapons and on their Destruction, BWC/CCNF/II/4/, 1986-08-18.
Introduction

The present review covers scientific and technological developments of relevance to the Convention on the Prohibition of the Development, Production and Stockpiling of Bacteriological and Toxin Weapons and on their Destruction during the period from the Second Review Conference 1986. This report is submitted in accordance with the recommendations in the Final Document of the Second Review Conference.

Disease-causing mechanisms

A pathogenic microorganism, is highly adapted and may cause disease because its survival strategy includes a requirement for infection. Well-established infectious agents have during evolution in general reached a state of balanced pathogenicity in the host, and cause the smallest amount of damage compatible with the need to enter, multiply and be discharged from the body. It has though been shown in a mice model that a mutated strain of *Yersinia pseudotuberculosis* may become as virulent as *Yersinia pestis*, the causative agent of plague. This result has provided the basis for the hypothesis that *Y. pestis* has evolved from *Y. pseudotuberculosis*. Less virulent strains of *Y. pestis* may have been harbored by non-human hosts, rats and fleas, during endemic phases, whereafter mutations rendered strains virulent and these strains caused the plague epidemics. This type of knowledge will pose a threat that microorganisms by a similar, deliberate manipulation could obtain enhanced virulence and become potential BW agents.

When an individual is exposed to a combined infection by two types of microorganisms at the same time, a more severe disease than that caused by each of the microorganisms might result. The effect of such co-infection is dependent on the nature of the two microorganisms. Enhancement has been seen in model systems including bacteria, viruses or both. Virus or bacteria may support the effect of each other and one type of organisms might express modified or stronger properties leading to a more severe infection. One example is the higher incidence of bacterial infection in connection with common cold caused by viruses. Hypothetically, an infectious process could be intensified by the simultaneous spread of modified organisms together with other organisms.
Slow infections is the general groupname of six diseases - three in humans and three in animals. The diseases are confined to the central nervous system and are supposed to arise spontaneously (genetically inherited) or after contact with infectious materials. The presumed human infectious agent has been denoted prion - a proteinaceous infectious particle. All attempts to isolate a nucleic acid in connection with the prions have failed. The pathogenesis of the disease is so far not characterized. There has recently been discussions concerning the spread of a similar slow infection in cattle. Although the risk for spread to humans has been suggested to be extremely low or non-existing, there has been a great public concern.

Release of genetically engineered microorganisms in the environment

Before release of genetically engineered organisms, several considerations have to be made. First, the persistence of the organism and effects on the ecosystem must be known. Competitiveness and confinement of the organisms is also important. In addition, the potential of genetic events such as gene alteration and gene transfer must be assessed. In order to gain knowledge about these factors, field tests have been performed the last five years. The first small-scale field test was performed with a mutant of the bacterium *Pseudomonas* which protects against frost-damages. Thereafter, several field tests have been conducted or are in progress with different genetically engineered microorganisms. Research in this area has also resulted in increased interest in the development of more refined dispersal models for biological aerosols. A regular follow-up of the area is of importance. The knowledge gained on persistence and ecological effects when releasing genetically modified organisms would also be of value if considering the effects of releasing B agents in the environment.

By using genetic engineering it may also be possible to program the survival of a released bacterial population. Some conditional suicide systems have been reported which are based on a cell killing function and appropriate regulatory sequences. By varying the ratio of two regulatory genes it was possible to construct clones where killing of a predetermined fraction of the cells per unit time occurred. This system is aimed for use when genetically modified organisms are released in the environment.
Recently, by the advent of recombinant DNA technology, the cloning of toxin genes and their expression in plant-associated microorganisms has provided potentially powerful alternative strategies for the protection of crops against insect damage.

Gene technology and man

The Human Genome Project has been initiated on a multinational basis with the aim of sequencing the complete human genome, as well as several other selected genomes. The work will involve the identification of 3000 million bases of DNA and at least 50 000 genes. The technical problems are immense as well as the costs, which have been estimated to 3 billion dollars. A deadline of 15 years for the project has been set. This will, though, require dramatic technical advances in order to keep the schedule. Immediate benefits of the project will be the identification and localization of genes causing hereditary diseases and simplification of the development of pharmaceutical drugs for treatment of hereditary diseases. It will also greatly refine prenatal diagnosis of these diseases. The vast knowledge that accumulates may also have applications in the area of biological and toxin weapons in the future.

New methods for detection and diagnosis

Recent advances in molecular biology have enabled a more precise determination of bacterial identification than previously has been possible. Using techniques such as the polymerase chain reaction and hybridization to ribosomal RNA (rRNA), have lead to the remarkable discovery that most environmental microbes are refractory to in vitro cultivation by current techniques. It is not unreasonable to assume that this is also the case for many microbes colonizing the human body. In recent years, such organisms have been identified. Examples are bacteria identified in AIDS patients with bacteremia. Obviously, molecular biology will allow for identification of a more precise and possibly more extensive array of already known genera of viruses and bacteria. However, the identification of new, highly infectious agents which causes global epidemics will be rare, although not highly unlikely, as shown by the number of individuals infected by HIV, an agent
not known 10 years ago. Another interesting example is the identification of hepatitis C.

The development of the polymerase chain reaction (PCR) is an important technological advancement in molecular biology. By the amplification procedure of nucleic acid, the detection limit may under some circumstances be one microorganism. The potential of the technique is enormous in diagnosis of microorganisms, as well as in the detection and identification of genes of interest. Some expectations have been fulfilled. These include the demonstration of DNA/RNA from hitherto unknown or uncultivable microorganisms in samples from patients. Moreover, RNA/DNA from HIV and other viruses has been demonstrated in a much greater number of individuals than that demonstrated by traditional diagnostic methods. The great sensitivity of the technique has, however, led to disputes over whether the findings are clinically relevant or not. Beside identification, the technique offers advantages for detection of mRNA in clinical specimens, for sequence analysis, detection of DNA polymorphism, and in cloning of genes.

An approach has been developed to diagnose microorganisms based on hybridization against rRNA with specific oligonucleotides. If, despite the great number of RNA molecules in each cell, the sensitivity is too low, the RNA hybridization technique may be combined with the use of the polymerase chain reaction, thereby greatly increasing the sensitivity of the test. There is a rapidly increasing knowledge of RNA sequences, which are collected in commercial databases. Thereby the usefulness will be greatly increased in the future. RNA sequences will probably be available for many possible BW agents.

Although there is much interest in the development of biosensors in general, progress in the development of receptor-based biosensors has been slow compared to sensors based on alternative sensing systems. With the growing knowledge of receptors for toxins and other biological active compounds, new biosensors will be developed. Biosensors will be good detection devices for all kinds of signal substances, such as toxins, B and C agents that the human body responds to. Thus, application of biosensors is a most interesting method for detection of BW agents or toxins in the future, although the expectations of the past decade have not been fulfilled.
Monoclonal antibodies have been used for identification, diagnosis and for purification of sensitive substances. A great number of monoclonal antibodies are available for identification of blood cells, tumor cells, microorganisms and various other substances. Recently, however, techniques have become available to "humanize" antibodies by using genetic engineering to transfer the binding part from a mouse antibody to a human antibody. Also, techniques have been introduced to produce monoclonal antibodies of human origin.

Another problem is to produce large quantities of antibodies to a reasonable cost. One solution to this problem is the use of recombinant microorganisms. However, an intact antibody is too big to be produced in bacteria. Instead fragments of antibodies, "mini-antibodies", are generated which have the advantage to be easily manipulated. Also "hybrid-molecules" can be created. Such a hybrid-molecule can consist of a "mini-antibody" and an enzyme or a toxin.

New antimicrobial and immunomodulating agents

In the past five years, four major and truly different categories of antibiotics have been introduced: fluoroquinolones, monobactams, augmented penicillins and imipenems. This is in sharp contrast to the slow progress made from 1960 to 1980. Of these antibiotics, monobactams, augmented penicillins and imipenems have only narrow indications. In contrast, fluoroquinolones have several advantages. Besides an outstanding antibacterial activity, they have low toxicity, low costs, and no transferable resistance. They have excellent properties for treatment of practically all important enteric pathogens.

A considerable number of antiviral drugs have been introduced during the last five years for clinical use, for example ribavirin, acyclovir and azidothymidine. Also interferons have been used for treating viral infections also in combination with antiviral drugs.

Drug design refers to the computer graphic modeling of small non-peptidic, organic molecules: antibiotics and enzyme inhibitors are examples. Unlike protein engineering, in which individual amino acids or discrete parts of a protein molecule are swapped or rearranged, drug design is closer to the traditional pharmaceutical development protocols. One example of drug
design is computer-aided design of antiviral agents. Clinical trials are under way with the most promising compounds. Effective drug design will in the future enable the development of improved and new pharmaceutical substances.

We now know that our immune system is controlled by a group of proteins, called cytokines, that act as immunohormones. Today, cytokines are primarily used in the areas of oncology, infectious diseases and bone marrow transplantations. The use of cytokines may probably increase dramatically in just a few years to treat various diseases. The interest will also be focused on cytokines with the ability to mediate an adjuvant effect when administered with vaccines. The use of such combinations will improve the possibility of protection against possible BW agents.

RNA with catalytic activity, ribozymes, has the potential of being used as therapeutic agents directed against viral RNA. Insertion of genes expressing ribozymes directed against RNA of pathogens, seems to be within reach using current technology. If ribozyme-catalyzed cleavage of specific RNA could be extended from in vitro to in vivo use, ribozymes might be extremely useful as prophylactic and therapeutic agents. Thus, ribozymes are promising antiviral agents even though the knowledge today is far too incomplete.

Antisense RNA are small molecules that pair to mRNA and control their expression, by blocking the informational flow from RNA to protein. Plants expressing antisense RNA against viral mRNA may e.g. become resistant to viral infection. Such a novel defence mechanisms against viruses has been shown to be very effective also in E. coli. The results mentioned demonstrate that the use of antisense genes to manipulate gene expression may have future applications.

Toxins in low doses and their possible utilization as therapeutic agents against several diseases is at present investigated. Another strategy is a specific targeting of the toxin to disease-causing cells by linkage of the toxin to a binding structure (to appropriate antibodies or other ligands, such as cytokines, by means of chemical conjugation or by recombinant DNA technology). Plant toxins and bacterial toxins have been used for construction of chimeric toxins. Diphtheria toxin has also been attached to a lymphocyte-specific cytokine. This results in increased knowledge of how to modify toxins and direct them against specific cells or organs.
Vaccine development

During 1986 to 1991 several types of vaccines have been investigated. Among these, component vaccines and recombinant vaccines appear to be the most promising. Component vaccines contain purified proteins or carbohydrates which constitute immunodominant antigens. Vaccines against Hemophilus influenzae and Streptococcus pneumoniae are two successful examples. Another interesting example is the use of an orally administered, attenuated form of cholera toxin as a vaccine against cholera.

There has recently been a rapid development in the area of recombinant vectors. Organisms such as vaccinia virus and Salmonella are at present investigated for their ability to harbor foreign genes and express vaccine antigens. In particular, the use of such recombinant vaccines against parasites, e.g. malaria and bilharzia, is the focus of an intense research. A useful vaccine, however, will probably not be available during this decade. Research is also being carried out on using these vectors to develop multivalent vaccines against a number of infectious agents. The use of such expression systems may also pose threats if toxins are included in the construction.

The use of molecular biology will probably clarify virulence mechanisms and enable identification of useful antigens to be included in vaccines from a wide array of microorganisms. Furthermore, research on vaccine carriers are also in progress. This will in a long perspective also allow for the development of suitable vaccines against potential BW agents against which no vaccines are available today. However, only a few vaccines have been registered during the last decade, so the development of such vaccines may last for a long time.

Industrial applications

Nowadays, microbiological fermentation technology allows efficient production of large quantities of microorganisms, bioproducts and cells. The need for new improved continuous bioreactors and separation techniques are growing. This means that the development will accelerate and that improved technology will be available on the market in the near future.
The new techniques simplify the large scale production of all kinds of bioengineered products and organisms in more modest sized plants. They also become more cost-effective. This development also means that the large scale production of many different BW agents may be simplified.

Expression of heterologous genes in living cells is often subjected to a number of limitations. For example, the protein product can be unsaturable in a given cell or, in some cases, it can be toxic to the cell. Due to this, cell-free translation system is being developed for production of polypeptides.

Recent advances in molecular biology enable large quantities of relatively rare, biologically active proteins, with a variety of medical and industrial applications, to be produced reliably. One drawback to their use is that they are inherently unstable. Stabilization can be achieved by manipulations of the proteins.

Catalytic antibodies, similarly to enzymes, may be used to catalyze reactions by stabilizing intermediate products of the reaction. The technique may have wide industrial applications when refined. The antibodies can be obtained by raising an immune response against a small molecule designed and chemically synthesized to mimic the intermediate product of the reaction of interest. It may thus be used as an alternative to organic synthesis. Protein engineering can be used for optimizing the activity of catalytic antibodies. The antibodies will be capable of catalyzing transformation of substrates which are not recognized by any enzyme.

Protein engineering is the manipulation of protein structures. This enables appropriate structural modifications or analog constructions and also de novo design of desired structures for specific functions. The possibilities to manipulate toxins or bioregulators, such as proteins and peptides, or to produce them in pure form in large quantities, opens up new perspectives for the future that has to be considered with implications for the Convention.

Microencapsulation techniques are subject to an increased attention within chemical, biological and medical research fields. Alternative microencapsulation techniques are generated on the research level, but complications arise when attempts are made to transform new processes to industrial level. These techniques could be used to improve stability of possible BW agents or toxins for dissemination in water and air.
United Kingdom of Great Britain and Northern Ireland

1. INTRODUCTION

1.1 Article XII of the Convention on the Proliferation of the Development, Production and Stockpiling of Bacteriological (Biological) and Toxin Weapons and on their Destruction (BWC) provided for a conference of States Parties to be held to review the operation of the Convention within five years after entry into force. Inter alia, this review was to take into account any new scientific and technological developments relevant to the Convention.


1.3 The Preparatory Committee for the Review Conference which met in April 1991, again requested the Depositary nations to prepare background papers on new scientific and technological developments relevant to the Convention, for distribution to all States Parties before the meeting of the Third Review Conference in September. The Committee also requested that this information should cover the applications of these developments and their relevance to various aspects of the Convention.

1.4 This paper, which follows the structure of the 1980 Co-depositary's paper (1) and the 1986 UK paper (2), sets out the views of the UK on developments since 1986.

(1) BWC/CONF 1/5 8 February 1980: United Nations.

(2) BWC/CONF 2/4 Add 1-2.
2. GENERAL DEVELOPMENTS RELEVANT TO THE BWC

2.1 The term genetic modification (GM) is used in this paper instead of the 'recombinant DNA techniques' of the 1980 Co-depository's paper (1) or the 'genetic engineering' of the 1986 UK paper (2) to reflect current practice in terminology in relevant European Commission (EC) Directives. The principles of the classical techniques of genetics and of the techniques of GM were outlined in the 1980 paper (1) and do not need to be repeated here. The term biotechnology is defined as the application of scientific and technological principles to the processing of materials by microorganisms, and this embraces GM techniques, fermentation, and downstream processing. To facilitate comparison with the previous papers, the same subject sub-headings are used.

2.2 Since 1986 there have been no major innovations among GM techniques but rather the continued refinement of techniques and an ever widening application to areas of microbiology and biology in general. Techniques for the high level expression of genes and for the modification of proteins are now relatively commonplace. Expression vectors for additional host species including an increased number of eukaryotic species have been developed, which increases the possibilities of having readily available the tools to effectively modify particular characteristics of any chosen microbial species. Modification (increase) of virulence has been reported for several pathogens; thus, an increase in the virulence of Yersinia pseudotuberculosis after a double mutation has been described, and baculovirus modified by insertion of the gene for the neurotoxin of the North African scorpion showed increased virulence for some species of insect.

2.3 The increasing use of biotechnology has led indirectly, or in some cases such as the industrial growth of genetically modified organisms, led directly to the imposition of regulatory guidelines or statutory controls by governments. In some respects increased national scrutiny of safety in laboratories and industry, and resulting national control regimes, may have tended to reduce the number of laboratories in developed nations that have expertise on the more exotic pathogens that cause concern for the BWC. In such nations, it is now so costly to build containment laboratory facilities to meet the required standard for handling dangerous pathogens and concurrently the pressure on research budgets has been such that academic and public health research on some pathogens which are not priority public health concerns for that particular country has tended to decline in the last decade.
2.4 The successful global eradication of smallpox has clearly demonstrated the continuing relevance of vaccination to combating disease, and global vaccination programmes against other diseases have expanded in the last five years. Under the World Health Organisation Expanded Programme for Immunisation, 60 million children are now vaccinated annually against diphtheria, pertussis, tetanus, polio, measles, and tuberculosis. GM techniques and advances in genetics are making new types of vaccines feasible, and there is considerable research towards genetically modified live attenuated vaccines able to immunise simultaneously against multiple antigens. However, for many diseases lack of knowledge of the basis of pathogenicity and also of the various facets of the immune response continues to limit the potential of new vaccines. Information relevant to understanding the human response to infectious disease may come from the multinational Human Genome Project which is working towards data bases on gene linkage and on polymorphism. The economics of vaccine production and sales including the risks of litigation provide little incentive for the commercial pharmaceutical sector to develop new vaccines. International collaboration between public sector organisations continues to be an important feature of R&D on new vaccines, and several vaccines now at the field trial stage have been developed outside developed nations, including vaccines for leprosy (Venezuela and India), leishmaniasis (Venezuela and Brazil) and dengue haemorrhagic fever (Thailand). GM continues to be a powerful tool in R&D work on vaccines. An example of the use of GM to produce sub-units that may have potential for future use as vaccines is the expression in Escherichia coli of a 31 kilodalton protein of Brucella. Use of GM to produce a live vaccine is one idea being considered for the development of an oral vaccine against anthrax for use with livestock or wildlife, by introducing the gene for the Protective Antigen into a non-anthrax bacterial species. Genetically modified vaccines are already starting to appear for animal applications as an improvement on existing vaccines that do not always offer full protection. An example in the UK is the development of an animal vaccine that uses fowl pox virus as a vector to carry genes for Newcastle Disease antigens.

2.5 Antibiotics have continued to improve towards compounds with a wider spectrum of activity. Although we do not yet have an antibiotic that kills all known bacteria, new compounds that approach this are imipenem which is however available only for parenteral use and is costly, and some of the new 4-quinolones. Research on anti-viral agents has continued and ribavirin has showed success in clinical trials of therapy of infections caused by some hantaviruses. There has thus been a minor improvement in the prospect for defensive measures against bacterial and rickettsial BW agents, and some lesser progress towards therapy of virus disease.
2.6 The application of GM techniques to the development of new proteins, commonly referred to as 'protein engineering', has continued to flourish. These activities fall into two groups; the modification of natural proteins such as enzymes in order to alter important characteristics such as stability and substrate specificity; and the creation of completely new types of molecule. An example of the latter is the successful creation of a synthetic enzyme having chymotrypsin-like properties.

2.7 The commercial exploitation of GM and other biotechnology techniques has accelerated in developed nations, and the medical, veterinary and agricultural applications remain very much to the fore but with increasing applications in the foodstuffs and cosmetics industries. There has been an increase in teaching and research programmes on the principles involved in applications of biotechnology, including post graduate courses specifically aimed at students from developing countries. Study areas include process biotechnology and specifically the scale up of operations using GM materials, the genome engineering of plants in order to improve disease resistance, and the biocontrol of plant pathogens and of insects including by infection with fungi. Increased emphasis and sophistication in downstream processing in commercial production operations largely for economic reasons has increased the potential for large scale production of proteins and small regulatory molecules. Continuing study of the genetics of crop species is leading to strains with improved disease resistance, and in the long term this will reduce the prospects of an aggressor waging economic warfare by attacking crops.

2.8 The use of labelled antibody techniques for the specific identification of pathogenic microorganisms and toxins has become increasingly routine in public health laboratories, and advances in knowledge of the molecular basis of antigens has led to antibody reagents of improved specificity, with a continuing trend from classical polyclonal antibodies towards monoclonal antibodies. There have been steady advances in the development of test methods based on criteria other than antigenic characteristics, and the potential for diagnostic laboratories to use a combination of such tests and antibody based tests should provide a useful improvement in the confidence of the ultimate identification conclusion. The use of the polymerase chain reaction (PCR) to amplify the specific reaction of gene probes with DNA sequences of the target microorganism is a particularly promising technique which provides the potential for specifically detecting as few as 10-100 microorganisms. Advances in diagnostic technologies directed at public health applications, which in some countries are the subject of considerable research efforts in academic, government, and commercial laboratories, may be expected to have had useful spin off into defence capabilities for identification and diagnosis of putative BW agents.
2.9 In conclusion, in the period since the BWC entered into force the
techniques of GM remain the most significant development among the scientific
and technological activities that have relevance for the BWC. There has been
steady refinement of those biotechnology aspects other than GM that an
aggressor nation could misuse in developing an offensive BW capability;
important among the capabilities that could be misused are techniques for the
large scale production of natural or modified microorganisms and toxins that
are now established in a considerable number of countries. Further advances
in capabilities of producing microorganisms and other biological agents are
to be expected. On the other hand, GM and other advances in biotechnology
have increased the potential for developing effective defensive measures
against possible BW attacks. These defensive measures include development of
multi-specific vaccines, and of techniques for identification and for
diagnosis, and techniques for field detection. Increased emphasis on health
and safety in the workplace has led to an expansion of the production of
civilian protective equipment including respirators, and this industrial base
could in some cases be easily adapted to provide protective equipment
suitable for biological defence.

3. NEW INFECTIOUS DISEASES

3.1 The two previous co-depository papers (1, 2) considered the
implications for the BWC of the newly recognised diseases Marburg, Ebola,
Lassa, and Legionnaires disease, and in 1986 it was then concluded that none
of these diseases had any special relevance to the BWC. The 1986 paper (2)
also drew this conclusion for human immuno deficiency virus (HIV), the
causative agent of AIDS. It is worth noting that recent evidence indicated
the incubation period of AIDS to be even longer than had previously been
thought. The 1986 paper also drew attention to the new arboviruses that had
been isolated since 1980, and concluded that whilst the arbovirus group
contains several well established candidate BW agents, and more may exist,
there was no indication that any of those isolated since 1980 could offer
compelling advantages as BW agents. Since 1986, the advance in knowledge
about these diseases or the additional new arboviruses that had been isolated
since then do not necessitate modification of these conclusions. However, it
must be recognised that the continuing increase in knowledge and expertise
relating to these newly recognised diseases and arboviruses in the public
health context with the passage of time, can only increase the potential for
misuse of such micro-organisms.

3.2 Understanding of the pathogenicity and epidemiology of infectious
diseases and intoxications is still often inadequate for predicting major
natural outbreaks of disease, and thus public health organisations continue
to be caught off guard on occasion. Diseases that have risen to new
prominence are cryptosporidiosis from water supplies, Escherichia coli
0157 causing haemorrhagic colitis, and this year the outbreak of cholera in Peru. Unexpected outbreaks of Q fever (in the UK) and diphtheria (in the USSR) also occurred in the period since the last paper. The improvement of the routine reporting of such disease incidents to bodies such as the WHO is encouraged.

3.3 In conclusion, the decade from 1980 has not seen the emergence of new infectious diseases with characteristics particularly relevant to the BWC. Such a wide range of pathogens with a combination of widely known characteristics that may make them attractive for development into weapons exist that in practice potential aggressors are unlikely to consider agent choice to be a constraint. In this decade there has been an entirely laudable increase in laboratory and epidemiologic expertise with many microbial species especially viruses highly pathogenic for man, as well as with a number of toxins, in support of national or multinational programmes aimed at the control and eradication of disease. This increased effort has resulted in improvements in laboratory capabilities for microbial identification and for diagnosis of disease, as well as in measures for prophylaxis and therapy. Nevertheless, it is a corollary of this increased global expertise that the opportunities for misuse of the knowledge may be said to have increased.

4. **Toxins**

4.1 A wide variety of toxins, not only bacterial toxins, have now been isolated and can be more readily produced than was the case in 1986. For many of them the molecular basis of toxicity is becoming better understood. One consequence of this increased level of knowledge is that toxins can be considered for specific therapeutic applications; e.g. botulinum toxin has recently been fully licensed for treatment of various dystonias, and a technique still in the research stages is the use of ricin targeted against tumour cells by means of specific antibodies—a concept that has been called the 'magic bullet'. As a result of studies of structure-function relationships novel toxins could be generated which are not found in nature. For example, the A and B sub-units of Shiga toxin and Shiga-like toxins have been combined. Such toxins may have altered toxicological or immunological properties which may confer advantages for therapeutic applications.

4.2 The increasing examples of the successful microbial synthesis of cloned proteins since 1986 further increases the potential for large scale production of toxins by this method, while the disadvantages of scaling up chemical synthetic routes remain. However, special problems in the expression of small peptides may complicate the use of GM techniques for production of such molecules in microorganisms. The ability to clone cassettes of genes also raises the possibility of the use of GM to produce non-protein toxins.
4.3 Cyanobacteria, formerly known as blue green algae, have caused a small number of incidents of intoxication in people using recreation water reservoirs in the UK. Because of this, any reservoirs showing an algal bloom are now closed to recreational use. Research is underway on the toxins involved and on their detection.

5. **INDUSTRIAL MICROBIOLOGY**

5.1 The use of GM materials in industrial microbiology has flourished and in many countries early fears over the safety of GM applications have subsided leading to the development of more realistic regulatory policies. There are moves towards international harmonisation of GM industrial regulations, including more formal acceptance of guidelines from bodies such as the WHO. Nevertheless there are still many countries where there is still very little government regulation of industrial microbiology.

5.2 GM techniques have been increasingly applied to eukaryotic organisms including yeasts and filamentous fungi, and for example some enzymes are now accessible from eukaryotes. The interest in exploiting these higher organisms, for example their expression advantages for proteins, has led to increased study of the special process engineering problems in scale manufacture of eukaryotes. The potential for the large scale production of toxins and smaller regulatory molecules by fermentation techniques is thus considerably increased.

5.3 In fermentation processes the use of real time sensors such as biosensors to give feedback loops under microprocessor control has allowed study and optimisation of production processes, and remote control of fermenters even by satellite links is now a possibility. Economic pressures have led to changes in design of industrial processes, with more attention on recovering materials from sidestreams and on processing effluents, usually by the application of well established technology. Government pressures to safeguard the environment have led to increased emphasis on demonstrating the safe disposal materials including the treatment of potentially hazardous effluent gases by incineration or filtration.

5.4 The view taken in the 1986 paper (2) that the Single Cell Protein industry should not be used as a global indicator of national industrial microbiology capability continues to be valid. In the Soviet Union, which far outstripped the West in the scale of SCP production in the 1970s and 1980s, public protests over environment pollution arising from SCP plants and other kinds of fermentation plants (eg plants making antibiotics and lysine) has recently led to the closure of several factories and abandonment of a number of new construction projects. In November 1989 the Soviet Government issued a decree calling for a halt to SCP production by 1991, but it is not
clear what action has resulted. In the face of soaring oil prices and
competition from cheap soya bean supplies, SCP production is no longer
considered economically viable in Western countries. Another industrial
activity that is very uneven worldwide is the fermentation of biomass to
produce fuels or energy. The greatest national commitment to producing
alternative transport fuels from biomass continues to come from Brazil. In
the UK, the potential outside the sewage sector is still largely untapped,
but electricity generated from gas generated in anaerobic digestion of
landfill waste now amounts to tens of megawatts.

5.5 There can thus be no doubt that the proliferation of legitimate
civilian industrial microbiology activities, and the continuing development
of the underlying theory and equipment, has increased the potential worldwide
for developing and producing biological weapons in contravention of the BWC.
The increased attention on analysis of risks to the environment and the move
towards improved physical containment means that again in principle it may
now be easier for an aggressor to acquire expertise and equipment suitable
for the illicit production of BW pathogens or toxin. Moves in several
countries towards better monitoring of exports of certain microorganisms and
for relevant dual use production equipment will act to limit this trend.

6. MICROBIAL CONTROL OF PESTS

6.1 In attempts to improve the efficacy of bioherbicides and insecticides,
there has been a significant increase in studies of the behaviour of the
particular microbial species when released into the environment. A parallel
field that is expanding is the release of Pseudomonas species having
impaired ice nucleation for use in the protection of crops against frost
damage.

6.2 The bacteria Bacillus thuringiensis is still the most widely
used entomopathogen, but there is increasing research on about 20 of the
several hundred entomopathogenic fungi known. Of the viruses, baculoviruses
continue to receive the most attention. Aspects of the use of these
microorganisms that are being studied include production, problems of low
pathogenicity, and constraints posed by temperature and humidity in the
field. Novel and improved formulations are being developed to extend shelf
life and to extend residual activity in the field eg by the use of
ultraviolet protectants. Formulations are also being developed to ensure
compatibility with existing agricultural spray devices, to remove the need
for dedicated high cost equipment. Because Lepidoptera are far more
of a problem in tropical areas, microbial control technology including
'cottage industry' production of pathogens is being developed in those areas
with the help of the developed nations.
6.3 In research on the use of fungal herbicides there has been an emphasis on the role of environmental factors in limiting the effective spread of disease in the target weed population.

6.4 In conclusion, although economic factors have tended to limit the expansion of the uses of biocontrol of pests, there has been increased study of factors relevant to effective dissemination. Such knowledge could in principle be misused by an aggressor intending to attack crops or, less likely, beneficial insect species eg those useful for pollination of crops. Some aspects of the dissemination technology would also be relevant to the deliberate release of organisms or toxins harmful to humans or animals.

7. CONCLUSIONS

7.1 The two previous papers (1, 2) concluded that the rapid pace of scientific and technological developments in areas closely related to the BWC demonstrated that implementation of its provisions had not hindered activities for peaceful purposes. In respect of developments since 1986, it is the UK view that there continues to be no impediment.

7.2 The UK continues to support the view that the BWC fully covers all biological agents whether naturally occurring or not, including any resulting from use of GM techniques.

7.3 The 1986 paper felt there was by then an increased potential for the large scale production of BW agents with enhanced military utility. The current UK view is that worldwide the increase in knowledge of many of the pathogenic species of microorganisms, and knowledge of toxins and other biological agents, and the continuing pace of developments in civil biotechnology areas, have further increased the possibilities for production and hostile use of biological agents, whether naturally occurring or not.

7.4 Increasing developments of the techniques of GM and biotechnology and continuing expansion of activities in industrial microbiology and microbial control of pests has further increased the capability worldwide for the production and dissemination of microorganism, of their products, and of other types of biological agents. Such developments in the civil sector are relevant to the BWC because they could be abused to support an offensive BW programme.

7.5 However, there has also been some increase in the potential for improving defensive measures against possible hostile use, particularly by taking advantage of civil developments in identification and diagnosis technologies, and by developing appropriate vaccines.
1. Introduction

1.1 In preparation for the 1991 Review Conference on The Convention on the Prohibition of the Development, Production and Stockpiling of Bacteriological (Biological) and Toxin Weapons and on their Destruction (BWC), the Preparatory Committee requested the depositary nations to prepare national papers on new scientific and technological developments relevant to the Convention.

1.2 Since the last Review Conference in 1986, the major breakthroughs have been the advances in basic research and the application of those advances in the fields of medicine, agriculture and industry. Of special interest to the BWC are the applications in detection and identification and the production advances that have ensued. The number of countries which are developing a biotechnology capability continues to grow and the industry has expanded both in scope and products developed. These trends continue to have practical significance for the BWC.

1.3 Our review of technological developments must continue to provide a broad overview of the field since advances are not limited to any one field. Advances in the industrial applications remain the most noteworthy since these advances provide new, simpler and more rapid production methods which is always a BWC concern. At the same time significant advances have been made in detection and identification technology due to biotechnology. This represents a positive development since faster, more precise detection of potential violations remain a concern. Our concern expressed in 1986 remains that while promising great benefits for mankind these advances could be used to produce new substances or modify old ones and lead to a significant toxin and new biochemical weapons threat.

2. Advances in industrial application of biotechnology

2.0 In a number of areas, the industrial application of biotechnology has relevance to the Convention.

2.1 Altered organisms. Biotechnology enables the development of microorganisms and products with new, unorthodox characteristics. These new microorganisms have a variety of uses, for example, in developing
environmentally safer pesticides or new treatments for cancer. However, in examining these developments from the point of view of the BWC, we cannot ignore the potential misuse of biotechnology to produce new biological agents or to improve certain characteristics in those already known. Transferring certain genetic traits into naturally infectious microorganisms can potentially create organisms of greater virulence, antibiotic resistance, and environmental stability. Changing the microbes genetically could alter their immunogenicity, thereby rendering vaccines and serodiagnostic techniques useless. Otherwise harmless microorganisms could be altered to produce toxemia or disease, although the host would continue to recognize these microorganisms as innocuous and therefore not defend against them.

2.1.1 Bioengineering of microorganisms has other implications as well. Bacteria and yeasts, genetically altered to produce products, are miniature factories by virtue of their ability to reproduce rapidly. Examples are the production of products by insertion of genes into tobacco mosaic virus and subsequent product extraction. Large quantities of compounds, previously available only in minute amounts, thus become available. Such a method of production is becoming more commonplace in civilian industry.

2.1.2 In addition, it is now possible to identify genes which have desirable properties and transfer them between host microorganisms. A nearly infinite variety of biological compounds designed for specific uses and given specific characteristics is possible. Given the technical progress in this area, future developments should be of concern to the Review Conference. Within the next decade, the potential for misuse of ongoing developments in biotechnology could be most pronounced in the following areas:

2.1.3 **Microbial pathogens** could be genetically engineered to maximize infectivity and pathogenicity. Likewise they could be modified to increase or decrease their environmental stability and persistency.

2.1.4 **Toxins.** Naturally occurring protein toxins could be made in host organisms by modifying their DNA. Plant and/or fungal toxins could be mass produced. If used as an agent, the origin of these toxins could be difficult to pinpoint, given that they are already in the environment, albeit at low concentrations. Improvements in biotechnology since the previous Review Conference leads us to believe that production of potent toxins, which until now were available only in minute quantities, and only upon isolation from...
immense amounts of natural biological materials, can now be produced in kilogram quantities which could be militarily significant.

2.1.5 Peptides. Peptides have been called "the antibiotics of the year 2000" because these biological materials may represent a new class of miracle drugs. Peptides are precursors of proteins made up of amino acids. They are interesting molecules for many reasons. They are active at very low concentrations (one part per billion or trillion) which makes their detection very difficult. They can be successfully modified as agonists (more active products) or antagonists (having a contrary activity). For example, modification of LHRH, a fertility hormone, by substituting a single amino acid has yielded a product 50 times more potent. Another modification of this same peptide yields a product useful in the treatment of prostate cancer.

2.1.6 Their range of activity covers the entire living system, from mental processes (e.g. endorphins) to many aspects of health such as control of mood, consciousness, temperature control, sleep, or emotions, exerting regulatory effects on the body. Even a small imbalance in these natural substances could have serious consequences, inducing fear, fatigue, depression or incapacitation. These substances would be extremely difficult to detect but could cause serious consequences or even death if used improperly.

2.1.7 The predictable modification of peptide and protein structure and function (i.e. protein engineering) is in its infancy. Computer-aided molecular design will rapidly develop, enabling molecules to be manipulated for varying degrees of physiological activity, specificity and stability. Technologies permitting the direct chemical synthesis of peptides and proteins in large yields will, in the more distant future, augment or replace microbial production of these molecules.

2.2 Advances in production. As mentioned above, once a suitable recombinant organism has been engineered, exploiting it becomes a matter of using established procedures. Biological production technology has proceeded to the point where large quantities of biological products can be produced quickly in small facilities. An example is the production of bovine growth hormone, a compound developed to increase milk production. Two genetically altered pesticides are about to be approved for commercial use which represent a "second-generation" of biopesticides which will not degrade quickly in the environment due to a microencapsulation process. Long-term refrigerated storage, in some cases, will not be needed because large quantities can be
produced very quickly starting from a minute seed stock. Several relevant technological considerations regarding biological production are discussed below.

2.2.1 Mammalian cell culture. Recent advances in mammalian cell culture make possible the growth of mammalian cells on the surface of minute beads, rather than on the inner surface of glass roller bottles. These cell culture systems provide the ideal environment for the growth of viruses. The new technique greatly simplifies virus production and allows large-scale yields in facilities of very modest size. As another example of advances in this field, the amount of tissue culture media needed to produce antibodies has been reduced a hundredfold by the use of encapsulated hybridomas. Such developments are eroding the distinction between production facilities and small laboratories.

2.2.2 Continuous flow fermentors. The introduction of computer controlled, continuous flow fermentors has dramatically increased productivity. Most likely the size of fermentors operating by batch process can be reduced a thousandfold by conversion to a continuous flow process.

2.2.3 Safety and environmental standards. Pharmaceutical plants around the world increasingly have incorporated safety and environmental release provisions akin to those which were once unique to BW production facilities, making it increasingly difficult to distinguish between permitted and prohibited activities.

2.2.4 Hollow fibre technology. Hollow fibre technology provides an example of the industrial production potential of the new technologies. This technology permits a far greater concentration of cells with a markedly increased rate of recovery in a shorter time than previously obtained using roller bottles. This equipment occupies less than one-twentieth the volume of the previous technology. Concentration and purification of a wide variety of protein substances can now be accomplished in large-scale liquid chromatography columns yielding pure, highly fractionated proteins. In the isolation of such cellular biomaterials as pyrogens, a similar transformation has taken place. Separation and reconstitution of the product can now be accomplished in about an hour using new compact ultrafiltration methods, whereas older methods took as much as four days.

2.2.5 Though not without constraints, developing biological and toxin weapons is an easier task than developing adequate defences against them. However,
the very advancements in biotechnology that have caused increased concern have also put new tools in the hands of those conducting permitted biological defence research.

2.2.6 In particular, gene splicing techniques can have an impact on development of effective vaccines against those disease agents already identified. In particular, gene splicing techniques can have an impact on development of effective vaccines against those disease agents already identified. Using vectored vaccine, rapid protection against multiple disease agents can be accomplished with a single injection. The polymerase chain reaction methodology has made detection of RNA or DNA at very low concentrations a reality. The application of this technology makes detection of single gene copies a possibility for even unsophisticated laboratories because the reagents are available in kit form. Biosensors and test kits are being developed to detect chemical agents, toxins, and pathogenic organisms.

Detectors which couple biological recognition sites such as receptors, antibodies, enzymes, or DNA probes, to electronic or optical microsensors are called biosensors. They combine the ability of the recognition site to detect agents in multicomponent mixtures at femtomolar concentrations with the signal amplification and analysis capability of microsensors. Biosensors have been designed to detect classes of toxins and chemicals, thus conferring a broader spectrum detection capability. While fieldable biosensors are still 3-5 years away, immunological test kits for toxin and bacterial spore detection have been fielded. Nevertheless, these positive developments represent a partial and incomplete response to the potential dangers resulting from advances in biotechnology.

2.3 Improvements in equipment, speed of production and quality of product are a common occurrence in the history of the commercial development of any new technology. Many oil utilizing microorganisms produce surface active compounds which can emulsify oil in water and facilitate recovery of the oil. A microbial glycolipid emulsifier has been produced in large quantities and shown to act as a powerful dispersant of oil in water. Unlike chemical surfactants, it is non-toxic and biodegradable. Other strains of microorganisms have been developed which have the potential for use in biodegradation of chemical nerve agents, mustard, explosives, and hazardous wastes such as PCB's. Other commercial developments in the area of agricultural research using biotechnology have occurred in crop research.
Cereals, forages, fibre crops and fruits and vegetables that are highly adapted to environmental stresses such as drought, cold, heat and toxic soil minerals are being developed.

In animal research, scientists are developing effective diagnostic tests for diseases of beef and dairy cattle, pigs, sheep, chickens, and turkeys, and producing new and more reliable preventive measures against livestock diseases. Other researchers are using the tools of biotechnology to grow bacteria that can break down toxic wastes. Because of this large number of technical innovations in biotechnology, especially in the area of industrial microbiology, the BWC has become more difficult to verify since its signature in 1972. Developments intended to increase production, decrease cost and create safer conditions for handling biological materials have blurred former distinctions important for purposes of verification — for example, between a large production facility and a laboratory. Also, capabilities to break out of the Convention in a very short time have increased.

3. Outbreaks of infectious diseases

3.1 As noted in our last report, acquired immune deficiency syndrome (AIDS) represents a newly recognized epidemic illness since the Review Conference in 1980.

3.2 AIDS. AIDS in a short period of time has become a major worldwide health problem. AIDS is caused by human T-cell lymphotrophic virus (HTLV-III), a retrovirus. The disease results from virus infection and destruction of T-helper cells, an important component of the immune system that helps the body ward off disease. Without these cells the patient is susceptible to a wide variety of opportunistic pathogens such as pneumocystis, fungi, and mycobacteria.

3.2.1 AIDS is a classic example of a new disease that has now become pandemic and which arose either from a mutational event of an existing human virus or introduction of an animal (monkey) virus into the human population.

3.3 EBOLA VIRUS. In November 1989, an outbreak of haemorrhagic fever occurred in cynomolgus monkeys (Macaca fascicularis) imported into the United States from the Philippines via Amsterdam and New York. The United States Army Medical Research Institute of Infectious Diseases isolated Ebola virus from affected monkeys. This was the first time that any strain of Ebola virus had been isolated from non-human primates that had not been infected experimentally. The origin of the infection was subsequently traced
to the Philippines. Although the initial outbreak occurred in Virginia, infections were subsequently reported in Pennsylvania and Texas in monkeys imported from the Philippines. Studies conducted by the Philippine Department of Health, the United States Navy, the United States Army Medical Research Institute of Infectious Diseases, and the Centers for Disease Control showed that a small percentage of animal handlers had developed antibody to the virus but no filovirus-related illness could be documented in any of the individuals. In 1976 outbreaks of Ebola virus infections in humans in Zaire and Sudan resulted in mortality rates of 88 per cent and 53 per cent respectively.

5. **Summary**

5.1 The past ten years have witnessed impressive strides in the fields of molecular biology and biotechnology. As the two juxtaposed words "molecular biology" imply, the distinction between biology and chemistry is becoming blurred. However, the United States continues to believe that Article I, which defines the scope of the Convention, has proved sufficiently comprehensive to have covered recent scientific and technological developments relevant to the Convention. In many ways, recent progress in biological technology affects the ease of concealment of manufacturing plants and the availability of new delivery systems, particularly for biological chemicals such as toxins and peptides. Verification of the Convention, always a difficult task, has been significantly complicated by the new technology. The confidence derived from the belief that certain technical problems would make biological weapons unattractive for the foreseeable future has eroded. The ease and rapidity of genetic manipulation, the ready availability of a variety of production equipment, the proliferation of safety and environmental equipment and health procedures to numerous laboratories and production facilities throughout the world, are signs of the growing role of biotechnology in the world's economy. But these very same signs also give concern for the possibility of misuse of this biotechnology to subvert the Convention.