



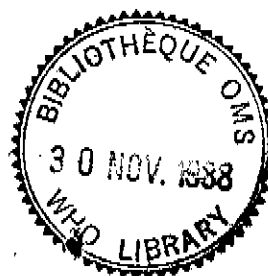
THE INTERNATIONAL PHARMACOPOEIA
THIRD EDITION
PHARMACOPOEIA INTERNATIONALIS

EDITIO TERTIA
Volume 4, Part 1

GENERAL REQUIREMENTS AND TESTING METHODS FOR DOSAGE FORMS

Contents

	<u>page</u>
1. Introduction	1
2. Tablets	2
3. Capsules	7
4. Parenteral preparations	11
5. Disintegration test for tablets and capsules	15
6. Uniformity of content for single dose preparations	18
7. Uniformity of mass for single dose preparations	18
8. Tests for sterility	20
9. Sterilization methods	24



1. INTRODUCTION

The publication of the third volume of the International Pharmacopoeia, third edition, almost completes the development of monographs for substances in the WHO Model List of Essential Drugs. The programme on the International Pharmacopoeia is at present focused on most widely used finished drug dosage forms, together with pharmaceutical aids employed in their manufacture.

This collection of general requirements and testing methods for pharmaceutical dosage forms constitute an advanced version of a section from the forthcoming fourth volume of the International Pharmacopoeia.

These requirements may be recommended for practical use in quality control in the absence of individual monographs for dosage forms which are now under development. With regard to the assay of dosage forms, a tentative general limit of $\pm 10\%$ may be recommended when the same analytical method as for the substance is applied, with a suitable pre-treatment if necessary. For certain drugs, simplified analytical techniques such as developed by the "Syndicat National de l'Industrie Pharmaceutique, France" could be considered.

This document is not issued to the general public, and all rights are reserved by the World Health Organization (WHO). The document may not be reviewed, abstracted, quoted, reproduced or translated, in part or in whole, without the prior written permission of WHO. No part of this document may be stored in a retrieval system or transmitted in any form or by any means - electronic, mechanical or other without the prior written permission of WHO.

The views expressed in documents by named authors are solely the responsibility of those authors.

Ce document n'est pas destiné à être distribué au grand public et tous les droits y afférents sont réservés par l'Organisation mondiale de la Santé (OMS). Il ne peut être commenté, résumé, cité, reproduit ou traduit, partiellement ou en totalité, sans une autorisation préalable écrite de l'OMS. Aucune partie ne doit être chargée dans un système de recherche documentaire ou diffusée sous quelque forme ou par quelque moyen que ce soit - électronique, mécanique, ou autre - sans une autorisation préalable écrite de l'OMS.

Les opinions exprimées dans les documents par des auteurs cités nommément n'engagent que lesdits auteurs.

2. TABLETS

Tablets are solid dosage forms containing one or more drug substances. They are obtained by single or multiple compression (in certain cases they are moulded) and may be uncoated or coated. They are intended for oral administration.* Different categories of tablets include soluble and effervescent tablets, tablets for use in the mouth and modified-release tablets. Unless otherwise specified in the individual monograph, tablets are normally circular in shape, their surfaces are flat or convex. Tablets may have lines or break-marks, symbols or other markings. They are sufficiently hard to withstand handling, including packaging, storage and transportation, without crumbling or breaking.

Tablets may contain pharmaceutical aids such as diluents, binders, disintegrating agents, glidants, lubricants, substances capable of modifying the behaviour of the dosage forms and the drug substance in the gastro-intestinal tract, colouring matter and flavouring substances. When such pharmaceutical aids are used, it is necessary to ensure that they do not adversely affect the stability, dissolution rate, bioavailability, safety and efficacy, of the drug substance; incompatibility between any of the components of the dosage form should be avoided.

Manufacture

The manufacturing processes for tablets should meet the general requirements on Good Manufacturing Practices (GMP),** especially with regard to cross-contamination. The following information is intended to provide very broad guidelines concerning the main steps during production with an indication of certain critical factors:

The particle size of the drug substance is primarily significant in terms of dissolution rate and extent, bioavailability and uniformity, especially for substances of low solubility in aqueous media. In order to obtain an appropriate formulation, it is usually necessary to mix the drug substance with a number of suitable pharmaceutical aids. It is essential that such mixing is carried out in a manner to ensure homogeneity. In some cases the physical characteristics of the mixture are such that it may be directly compressed. Sometimes, it is necessary to granulate before compression, e.g. by wet granulation or precompression (slugging).

The granulate and powders normally need to be mixed with lubricants and/or disintegrating agents. The use of excessive amounts of lubricants should be avoided since these will deleteriously affect the tablets. The final tablet

* Preparations intended for use other than by oral administration (implants, solution tablets for injections, irrigations or for external use, vaginal tablets etc.) may also be presented in this form. These preparations may require a special formulation, methods of manufacture or form of presentation appropriate to the particular use. For this reason they may

** Adopted in World Health Assembly WHA28.65 and text reproduced in PHARM/82.4/Rev.3 Certification Scheme on the Quality of Pharmaceutical Products moving in International Commerce (periodically updated).

mixture is volumetrically fed into the die cavity to assure tablets of uniform mass and compressed at an adequate pressure. When necessary, the tablets may be coated, e.g. in coating pans or by air suspension technique.

Throughout the manufacturing procedure critical steps should be validated and monitored by carrying out appropriate in-process controls. Such tests should be designed to provide assurance of the effectiveness of each stage of production. Important in-process controls for different stages of tablet production are: particle size of the drug substance, homogeneity of the mixture, moisture content of the mixture and/or granulate, size of granules, flowability of final mixture, dimensions (thickness, diameter), uniformity of mass, hardness,* friability, disintegration, or dissolution rate (if appropriate, such as for modified-release tablets) of the finished dosage form. Attention should be paid to the uniformity of mass of tablet cores before coating in the manufacture of coated tablets, particularly for sugar-coated tablets.

The packaging of tablets should protect them from light, moisture and damage during transportation.

GENERAL REQUIREMENTS

Visual inspection

Visually inspect at least 20 tablets. They should be without any damage, smooth and usually of uniform colour. Evidence of physical instability may consist of the following:

- presence of excessive powder and/or pieces of tablets at the bottom of the container (from abraded, crushed or broken tablets);
- cracks or capping, chipping in the tablet surfaces or coating, swelling, mottling, discoloration, fusion between tablets;
- the appearance of crystals on the container walls or on the tablets.

Labelling

Every pharmaceutical preparation must comply with the labelling requirements established in the Good Manufacturing Practices (GMP).

The label on the container should include:

- (1) the name of the drug; INN's should be used wherever possible;
 - (2) the name and amount of the drug substance(s) in each tablet and the number of tablets in the container;
 - (3) the batch (lot) number assigned by the manufacturer;
 - (4) the expiry date, and when required the manufacturing date;
 - (5) any special storage conditions or handling precautions that may be necessary;
 - (6) directions for use, warnings and precautions that may be necessary;
- and
- (7) the name and address of the manufacturer or the person responsible for placing the drug on the market.

* Hardness = crushing force

Storage

Throughout their shelf-life the tablets should maintain their integrity by storage in well-closed containers, protected from light, crushing and mechanical shock, kept at temperatures compatible with their stability, and whenever necessary in areas of low humidity. They should withstand handling, including packaging and transportation without losing their pharmaceutical integrity. Moisture sensitive forms, such as effervescent tablets should be stored in tightly closed containers or moisture proof packs and may require the use of separate packages containing water-adsorbent agents, such as silica gel.

Additional special packaging, storage and transportation recommendations are given in the individual monograph.

Uniformity of mass

Tablets comply with the test for "Uniformity of Mass for Single Dose Preparations", unless otherwise specified in the individual monograph.

Uniformity of content

A requirement for the "Uniformity of Content for Single Dose Preparations" is specified in certain individual monographs for sugar-coated or enteric-coated tablets, where the test for uniformity of mass does not apply. In addition a requirement is specified in certain individual monographs where the drug substance is 5 % or less of the total formulation. In such cases the test for uniformity of mass may not be required.

Dissolution test

Where a requirement for the "Dissolution Test" is specified in the individual monograph the "Disintegration Test for Tablets and Capsules" may not be required.

REQUIREMENTS FOR SPECIFIC TYPES OF TABLETS

1. Uncoated tablets

The majority of uncoated tablets are made in such a way that the release of drug substances is unmodified. A broken section, when examined under a lens, shows either a relatively uniform texture (single-layer tablets) or a stratified texture (multi-layer tablets) but no signs of coating.

Disintegration test

Uncoated tablets, except effervescent tablets, tablets for use in the mouth and chewable tablets comply with the "Disintegration Test for Tablets and Capsules". Operate the apparatus for 15 minutes, unless otherwise specified in the individual monograph, and examine the state of the tablets.

1.1 Soluble tablets (tablets for solutions)

Soluble tablets are uncoated tablets that dissolve in water to give a clear solution.

Disintegration test

Use water at room temperature, and operate the apparatus for 5 minutes, unless otherwise specified in the individual monograph.

1.2 Effervescent tablets

Effervescent tablets are uncoated tablets generally containing acid substances and carbonates or hydrogen carbonates which react rapidly in the presence of water to release carbon dioxide. They are intended to be dissolved or dispersed in water before administration. They should be labelled not to be swallowed directly.

Disintegration test

Place a tablet in a 250 ml beaker containing 200 ml of water at room temperature. Numerous bubbles of gas are evolved. When the evolution of gas around the tablet or its fragments ceases, the tablet should have disintegrated, being either dissolved or dispersed in the water so that no agglomerates remain. Repeat the operation on 5 other tablets. The tablets comply with the test if each of the 6 tablets used in the test disintegrates within 5 minutes, unless otherwise specified in the individual monograph.

1.3 Tablets for use in the mouth (sublingual, buccal) and chewable tablets

Tablets for use in the mouth and chewable tablets are usually uncoated tablets. They are formulated to effect a slow release and local action of the drug substance or substances (for example, compressed lozenges) or the release and absorption of the drug substance under the tongue (sublingual tablets) or in other parts of the mouth for systemic action.

2. Coated tablets

Coated tablets are tablets covered with one or more layers of mixtures of various substances such as natural or synthetic resins, polymers, gums, fillers, sugars, plasticisers, polyols, waxes, colouring matters and flavouring substances, and drug substances. A broken section, when examined under a lens shows a core surrounded by a continuous layer of a different texture.

The tablets may be coated for a variety of pharmaceutical reasons including protection of the drug substances from air, moisture or light, masking of unpleasant tastes and odours, or improvement of appearance. The substances used for coatings are usually applied as a solution or suspension.

Three main categories of coated tablets may be distinguished: sugar-coated, film-coated and certain modified-release tablets.

2.1 Sugar-coated tablets

Sugar-coated tablets are coated tablets in which the major coating agent is sugar. If a hydrophobic subcoat is applied to the core it can diminish dissolution from the core, especially on storage.

Uniformity of mass

The test does not apply to sugar-coated tablets (see section 2, in-process controls).

Disintegration test

Operate the apparatus for 60 minutes, unless otherwise specified in the individual monograph, using water, and examine the state of the tablets. If any of the tablets has not disintegrated, repeat the test on a further 6 tablets, using hydrochloric acid (0.1 mol/l) VS.

All 6 tablets must have disintegrated.

2.2 Film-coated tablets

A film-coated tablet is covered with a thin layer of resins, polymers and plasticisers capable of forming a film.

Disintegration test

Operate the apparatus for 30 minutes, and examine the state of the tablets.

3. Modified-release tablets

3.1 Extended release tablets

Extended release tablets are coated, uncoated or matrix tablets containing pharmaceutical aids or prepared by procedures which, separately or together, are designed to modify the rate or the site at which the drug substance(s) are released in the gastro-intestinal tract.

The specialized nature of these dosage forms is such that all the requirements are given in the individual monograph.

3.2 Delayed release tablets (Enteric-coated tablets)

Enteric-coated tablets are tablets covered with one or more layers intended to resist the gastric fluid but to permit disintegration in the intestinal fluid. These properties are achieved using substances such as cellacelate (cellulose acetate phthalate), and anionic copolymers of methacrylic acid and its esters in the coating. It may be necessary to apply an additional protective coat.

Uniformity of mass

The test does not apply to enteric-coated tablets.

Disintegration test

Use hydrochloric acid (0.1 mol/l)VS as the immersion fluid. Operate the apparatus for 2 hours, unless otherwise specified in the individual monograph but in any case not less than one hour, and examine the state of the tablets. No tablet shows signs of either disintegration (apart from fragments of coating) or cracks that would allow the escape of the contents. Replace the acid by phosphate buffer solution, pH 6.8, TS. Operate the apparatus for 60 minutes and examine the state of the tablets.

3. CAPSULES

Capsules are solid dosage forms with hard or soft shells depending upon their formulation. They are of various shapes and capacities containing a single dose of one or more drug substances. They are intended for oral administration.* Different categories of capsules include hard, soft, and modified-release capsules. Their surfaces may bear symbols or other markings. They are sufficiently robust to withstand handling, including packaging, storage and transportation, without cracking or breaking. They should be protected from microbial contamination.

The capsule shells are made of gelatin or other substances, the consistency of which may be adjusted by the addition of substances such as glycerol and sorbitol. Preservatives may be added as well. The contents of capsules may be solid, liquid or of a paste-like consistency. The capsule shell and the contents of capsules may contain pharmaceutical aids such as diluents, solvents, surface-active substances, opaque fillers, antimicrobial agents, sweeteners, colouring matter and flavouring substances, disintegrating agents, glidants, lubricants, substances capable of modifying the behaviour of the drug substance in the gastro-intestinal tract. The contents should not cause deterioration of the shell. The shell, however, is attacked by the digestive fluids so that the contents are released.

When pharmaceutical aids are used, it is necessary to ensure that they do not adversely affect the stability, dissolution rate, bioavailability, safety and efficacy of the drug substances; incompatibility between any of the components of the dosage form should be avoided.

GENERAL REQUIREMENTS

Visual inspection

Visually inspect at least 20 capsules. They should be smooth and without any damage. Evidence of physical instability may consist of the following: changes in gross physical appearance, including hardening or softening, cracking, swelling, mottling or discoloration of the shell.

Labelling

Every pharmaceutical preparation must comply with the labelling requirements established in the Good Manufacturing Practices (GMP):

The label on the container should include:

- (1) the name of the drug; INN's should be used wherever possible;

* Preparations intended for use other than by oral administration (e.g., vaginally or rectally) may also be presented in the form of capsules. These may require special formulation, methods of manufacture or presentation appropriate to the particular use and may not comply with certain parts of this monograph. Starch capsules (often known as cachets) are not described in this monograph.

- (2) the name and amount of the drug substance(s) in each capsule and the number of capsules in the container;
- (3) the batch (lot) number assigned by the manufacturer;
- (4) the expiry date, and when required the manufacturing date;
- (5) any special storage conditions or handling precautions that may be necessary;
- (6) directions for use, warnings and precautions that may be necessary; and
- (7) the name and address of the manufacturer or the person responsible for placing the drug on the market.

Storage

Throughout their shelf-life the capsules should maintain their integrity by storage in well-closed containers, protected from light, excessive moisture or dryness and temperatures above 30 °C. Additional special storage recommendations or limitations are given in the individual monograph. In general, capsules should be stored in areas of low humidity.

Uniformity of mass

Capsules comply with the test for "Uniformity of Mass for Single Dose Preparations", unless otherwise specified in the individual monograph.

Uniformity of content

A requirement for the "Uniformity of Content for Single Dose Preparations" is specified in certain individual monographs where the drug substance is 5 % or less of the total formulation. In such cases the test for uniformity of mass may not be required.

Dissolution test

Where a requirement for the "Dissolution Test" is specified in an individual monograph the "Disintegration Test for Tablets and Capsules" may not be required.

REQUIREMENTS FOR SPECIFIC TYPES OF CAPSULES

1. Hard capsules

Hard capsules have shells consisting of two prefabricated cylindrical sections, one end of each of which is rounded and closed, and the other open. The two sections are fitted together. The contents of hard capsules are usually in solid form (powder or granules).

Manufacture

The manufacturing and filling process for hard capsules should meet the general requirements on Good Manufacturing Practices (GMP), especially with regard to cross-contamination. The following information is intended to provide very broad guidelines concerning the main steps during production with an indication of certain critical factors:

The particle size of the drug substance is primarily significant in terms of dissolution rate and extent, bioavailability and uniformity, especially for substances of low solubility in aqueous media. In order to obtain an appropriate formulation, it is usually necessary to mix the drug substance with a number of suitable pharmaceutical aids. It is essential that such mixing is carried out in a manner to ensure homogeneity. In some cases, the physical characteristics of the mixture are such that it may be directly filled into the shell. Occasionally, it is necessary to granulate before filling. Normally the granulates need to be mixed with lubricants and/or disintegrating agents. The use of excessive amounts of lubricants should be avoided since these may deleteriously affect the capsules. In certain cases the contents of capsules may be in the form of encapsulated powders or micropellets.

A uniform mass of the capsule mixture is volumetrically fed into the narrower lower section of the shell body which is then closed by slipping the larger section or cap over it. The security of the closure may be strengthened by suitable means.

Throughout the manufacturing procedure critical steps should be validated and monitored by carrying out appropriate in-process controls. Such tests should be designed to provide assurance of the effectiveness of each stage of production. Important in-process controls for different stages of hard capsule production are: particle size of the drug substance, homogeneity of the mixture, water content of the mixture or granules and of the shells, size of the granules, flowability of the final mixture, uniformity of mass, capsule size, integrity of the seals, disintegration or dissolution rate (if appropriate, such as for modified-release capsules) of the finished dosage form.

Disintegration test

Hard capsules comply with the "Disintegration Test for Tablets and Capsules".

Use water as the immersion fluid unless hydrochloric acid (0.1 mol/l)VS is specified in the individual monograph. Operate the apparatus for 30 minutes and examine the state of the capsules.

If capsules float use a disc as described under "Disintegration Test for Tablets and Capsules" (Disintegration apparatus).

2. Soft capsules

Soft capsules have thicker shells than those of hard capsules. Preservatives usually are added. The shells consist of one piece and are of various shapes. There may be partial migration of the constituents from the capsule contents into the shell and vice versa because of the nature of the materials and the surfaces in contact.

Manufacture

The manufacturing processes for soft capsules should meet the general requirements on GMP. The following information is intended to provide very broad guidelines concerning the main steps during production with an indication of certain critical factors:

The particle size of the drug substance is primarily significant in terms of dissolution rate and extent, bioavailability and uniformity, especially for substances of low solubility in aqueous media. In order to obtain an appropriate formulation, it is usually necessary to mix the drug substance with a number of suitable pharmaceutical aids. It is essential that such mixing is carried out in a manner to ensure homogeneity. Soft gelatin capsules are usually formed, filled and sealed in one operation but for extemporaneous use, shells may be prefabricated. Liquids may be enclosed directly. Solids are usually dissolved or dispersed in a suitable pharmaceutical aid to give a solution or dispersion of somewhat paste-like consistency.

Throughout the manufacturing procedure critical steps should be validated and monitored by carrying out appropriate in-process controls. Such tests should be designed to provide assurance of the effectiveness of each stage of production. Important in-process controls for different stages of soft capsule production are: particle size of the drug substance, homogeneity of the mixture, viscosity of the contents, uniformity of mass, capsule size, integrity of the seals, disintegration, or dissolution rate (if appropriate, such as for modified-release capsules) of the finished dosage form.

Disintegration test

Use water as the immersion fluid unless hydrochloric acid (0.1 mol/l)VS is specified in the individual monograph. Operate the apparatus for 30 minutes and examine the state of the capsules.

3. Modified-release capsules

3.1 Extended release capsules

Extended release capsules are hard or soft capsules in which the contents or the shell or both contain additives or are prepared by special procedures such as microencapsulation, which separately or together are designed to modify the rate or site at which the drug substance(s) are released in the gastro-intestinal tract.

The specialized nature of these dosage forms is such that all the requirements are given in the individual monograph.

3.2 Delayed release capsules (enteric capsules)

Enteric capsules are hard or soft capsules prepared in such a manner that either the shell or the contents resist the action of the gastric fluid but are attacked by the intestinal fluid to release the drug substance.

Manufacture

The statement for either hard or soft capsules applies as appropriate.

Disintegration test

For enteric-coated capsules use the following test:

Use hydrochloric acid (0.1 mol/l)VS as the immersion fluid. Operate the apparatus for 2 hours, unless otherwise specified in the individual monograph but in any case not less than one hour, and examine the state of the capsules. No capsule shows signs of disintegration or rupture permitting the escape of the contents. Replace the acid by phosphate buffer solution, pH 6.8, TS with added pancreatin R where specified in the individual monograph. Operate the apparatus for 60 minutes and examine the state of the capsules.

4. PARENTERAL PREPARATIONS

Parenteral preparations are sterile, pyrogen-free liquids (solutions, emulsions or suspensions) or solid dosage forms containing one or more drug substances. They are intended for administration by injection, infusion or implantation into the body.* They are packaged either as single dose or multidose preparations.

There are four main forms of parenteral preparations: injections, intravenous infusions (large volume parenterals), powders for injections and implants.

Certain injections and intravenous infusions may be presented in the form of a sterile concentrated solution which must be suitably diluted before use.

Parenteral preparations may contain pharmaceutical aids such as solvents, suspending agents, buffers, substances to make the preparation isotonic with blood, stabilizers or antimicrobial preservatives. The use of pharmaceutical aids however should be minimized, and when used it is necessary to ensure that they do not adversely affect the stability, bioavailability, safety and efficacy of the drug substances, or at the concentrations used, cause toxicity or undue local irritation. Incompatibility between any of the components of the dosage form should be avoided.

Water for injection is used as the vehicle for aqueous injections. It should be freshly distilled by the process described under "Aqua pro Injectione", free from carbon dioxide and complying with the test for pyrogens (see Vol. 1, p. 155). The sterilization at this stage may be omitted, provided that the final solution or preparation is immediately sterilized. For non-aqueous injections fixed oils of vegetable origins are used as vehicles.

Unless otherwise specified in the individual monograph, sodium chloride, or other suitable substance or substances, may be added to an aqueous solution for injection in order to make it isotonic with blood.

* Preparations, such as vaccines, human blood and products derived from human blood, peritoneal dialyzing solutions, radioactive preparations or prostheses to be implanted require special formulation, methods of manufacture or presentation appropriate to the particular use and may not comply with certain parts of this monograph.

MANUFACTURE

The manufacturing process for parenteral preparations should meet the general requirements on Good Manufacturing Practices (GMP). The following information is intended to provide very broad guidelines concerning manufacture with an indication of certain critical factors:

The quality of starting materials, the design and maintenance of the equipment and the methods of manufacture must be such as to ensure the stability of the active substance and a final product which is sterile and free of pyrogens * and particulate matter.

Methods of sterilization that may be used for parenteral preparations are given in "Sterilization Methods". For aqueous preparations, heating in an autoclave is the method of choice and therefore, whenever it is known to be suitable, this method should be used.

When a parenteral preparation is liable to deterioration due to oxidation, the operation of filling may be performed in an atmosphere of a suitable inert gas, such as nitrogen, the air in the container being replaced by the same gas.

Throughout the manufacturing procedure critical aspects should be validated and monitored by carrying out appropriate in-process controls. Such tests should be designed to provide assurance of the effectiveness of each stage of production. Important in-process controls during the manufacture of parenteral preparations are: monitoring of environmental conditions especially with respect to particulate and microbial contamination, pyrogens [use of a limulus amoebocyte lysate (LAL) test may be advantageous], pH of solution, clarity of solution, freedom from particulate matter, integrity (absence of leakage, etc.). Additionally: for dispersions - the particle size of the dispersed phase; for powders for injections - uniformity of mass, moisture content, and the ease of reconstitution of a solution or suspension; mass or volume contained; presence of preservatives or other additives as these can influence the choice of assay method.

GENERAL REQUIREMENTS

Containers

Parenteral preparations are usually supplied in glass ampoules, bottles or vials, plastic bottles or bags and prefilled syringes, which are coloured in cases of light sensitive substances.

These containers are made from materials which are sufficiently transparent to permit the visual inspection of the contents, except for implants and where indicated in the individual monograph. They should not adversely affect the quality of the preparation and should not allow diffusion into or across the material of the container or yield foreign substances to the preparation.

* From the clinical viewpoint all parenteral preparations must be pyrogen-free. However, on practical considerations certain categories of parenteral preparations may be exempted from the test for pyrogens as specified in the individual monograph.

Closures

Closures for parenteral preparations should ensure a good seal, prevent the access of microorganisms and other contaminants and usually permit the withdrawal of a part or the whole of the contents without removal of the closures. They should not contain components which react with the contents, nor yield foreign substances into the preparation. The plastic materials or elastomers of which the closure is composed are sufficiently firm and elastic to allow the passage of a needle with the least possible shedding of particles. Closures for multidose containers are sufficiently elastic to ensure the puncture is resealed when the needle is withdrawn and protect the contents from airborne contamination.

Visual inspection

Parenteral preparations in the form of solutions, reconstituted solutions and intravenous infusions (except dispersions), should be free from visible particulate matter, and clear.

Labelling

Every pharmaceutical preparation must comply with the labelling requirements established in the GMP:

The label on the container should include:

- (1) the name of the drug; INN's should be used wherever possible;
 - (2) for injections: the name and amount of the drug substance(s) in a suitable dose volume and the volume in the container; for powders for injections: the name and amount of the drug substance(s) in the container;
 - (3) the batch (lot) number assigned by the manufacturer;
 - (4) the expiry date, and when required the manufacturing date;
 - (5) any special storage conditions or handling precautions that may be necessary;
 - (6) directions for use, warnings and precautions that may be necessary;
- and
- (7) the name and address of the manufacturer or the person responsible for placing the drug on the market.

For parenteral preparations that are solutions or dispersions the concentration of the drug substance should be given in terms of mass or biological activity per volume. The label of concentrated solutions should state the composition and the dilution to be carried out before use.

Test for sterility

Parenteral preparations comply with the "Test for Sterility."

Test for pyrogens

All intravenous infusions and those injections and powders for injections where the volume to be injected in a single dose is 15 ml or more comply with the "Test for Pyrogens". In addition, where specified in the individual monograph, injections and powders for injections, irrespective of the volume to be injected in a single dose, comply with the "Test for Pyrogens" (certain preparations where the active ingredient is of 'biological origin').

For injections, the amount should relate to the volume of the dose and is specified in the individual monograph. This amount should usually be also related to the maximum dose of the active drug substance tolerated by the animal.

For powders for injections, the amount of powder and the nature and volume of the liquid in which it is to be dissolved or suspended are specified in the individual monograph.

REQUIREMENTS FOR SPECIFIC TYPES OF PARENTERAL PREPARATIONS

1. Injections

Injections are sterile pyrogen-free solutions or dispersions (emulsions or suspensions) of one or more drug substances in a suitable vehicle.

Whenever possible, an injection should be prepared using an aqueous vehicle. If necessary, suitable non-aqueous solvents are indicated in the individual monographs. Injections which are dispersions should remain sufficiently stable so that after shaking, a homogeneous dose can be withdrawn.

Whenever possible the use of single dose injections is to be preferred. When the preparation is intended for administration by routes where, for medical reasons, an antimicrobial preservative is not acceptable, e.g. intracisternal, intrathecal, the use of a single dose injection is essential.

1.1 Single dose preparations

Single dose preparations contain a sufficient quantity of the injection to permit the withdrawal and administration of the labelled volume using a normal technique.

1.2 Multidose preparations

Multidose preparations must contain a suitable antimicrobial preservative in appropriate concentrations except where the preparation itself has adequate antimicrobial properties, and the containers should be able to ensure adequate protection of the contents after partial withdrawal. In order to minimize the risk of contamination resulting from multiple penetrations, the volume of multidose preparation should normally not exceed 30 ml.

2. Intravenous infusions

Intravenous infusions are sterile, pyrogen-free aqueous solutions or emulsions with water as the continuous phase, usually made isotonic with blood. They are intended for administration in large volumes (usually 100 ml or more), and should not contain any antimicrobial preservatives.

On visual inspection emulsions for intravenous injections should show no evidence of phase separation. The particle size of the dispersed phase should be controlled by the manufacturer.

3. Powders for injections

Powders for injections are solid substances (including freeze-dried materials), distributed in their final containers and which, when shaken with the prescribed added volume of the appropriate sterile liquid, rapidly form either clear and practically particle-free solutions or uniform suspensions. Powders for injections after dissolving or suspending comply with the requirements for injections or intravenous infusions, as appropriate.

Uniformity of mass

Powders for injections when intended for single dose use, comply with the test for the "Uniformity of Mass for Single Dose Preparations", unless otherwise specified in the individual monograph.

Uniformity of content

A requirement for the "Uniformity of Content for Single Dose Preparations" is specified in certain individual monograph where the drug substance is less than 40 mg. In such cases, the test for uniformity of mass may not be required.

4. Implants

Implants are solid preparations containing one or more drug substances. They are of a size and shape suitable for parenteral implantation, and provide a release of the drug substance over an extended period of time. They are presented in individual sterile containers.

The specialized nature of these dosage forms is such that all the requirements are specified in the individual monograph.

5. DISINTEGRATION TEST FOR TABLETS AND CAPSULES

This test determines whether tablets and capsules disintegrate within a prescribed time when placed in the immersion fluid under the prescribed experimental conditions. Disintegration is defined as the state in which no residue of the tablet or capsule, except fragments of undissolved coating or capsule shell remain on the screen of the test apparatus or, if any other residue remains, it consists of a soft mass having no palpably firm, unmoistened core.

Disintegration apparatus

The apparatus consists of a basket-rack assembly (Fig.), a suitable vessel for the immersion fluid (such as a 1 litre beaker), a thermostatic arrangement for maintaining the fluid at the required temperature, normally at 37 ± 2 °C and a device for raising and lowering the basket-rack in the immersion fluid at a constant frequency of 28-32 cycles/min through a distance of 50-60 mm. The volume of the fluid in the immersion vessel is such that at the highest point of the upward stroke the wire mesh which forms the bottom of the basket

remains at least 25 mm below the surface of the fluid, while, at the lowest point of the downward stroke, it descends to not less than 25 mm from the bottom of the vessel. The time required for the upward stroke should be equal to the time required for the downward stroke, and the change in stroke direction should be a smooth transition rather than an abrupt reversal of motion. The basket-rack assembly consists of 6 open-ended cylindrical glass tubes and a rack for holding them in a vertical position. The tubes are 75-80 mm long, have an inside diameter of about 21.5 mm and a wall about 2 mm thick. The tubes are held vertically by 2 superimposed plates, circular in shape and made of transparent plastic material, each about 90 mm in diameter and 6 mm thick, perforated by 6 holes of a diameter that allows the tubes to be inserted. The holes are equidistant from the centre of the plate and equally spaced one from another. A piece of woven gauze made of stainless steel wire about 0.635 mm in diameter with a mesh aperture of 2.0 mm is attached to the under side of the lower plate. The upper plastic plate is covered with a stainless steel plate, about 1 mm in thickness, of a diameter similar to that of the plastic plates. The steel plate is perforated by 6 holes about 22 mm in diameter positioned to coincide with those of the upper plastic plate and the upper open ends of the glass tubes. The steel plate fits over the tubes and holds them between the plastic plates. The plates are held rigidly 75-80 mm apart by vertical stainless steel rods at the periphery. A metal rod is fixed to the centre of the upper plate. This enables the assembly to be attached to a mechanical device and to be suitably lowered and raised.

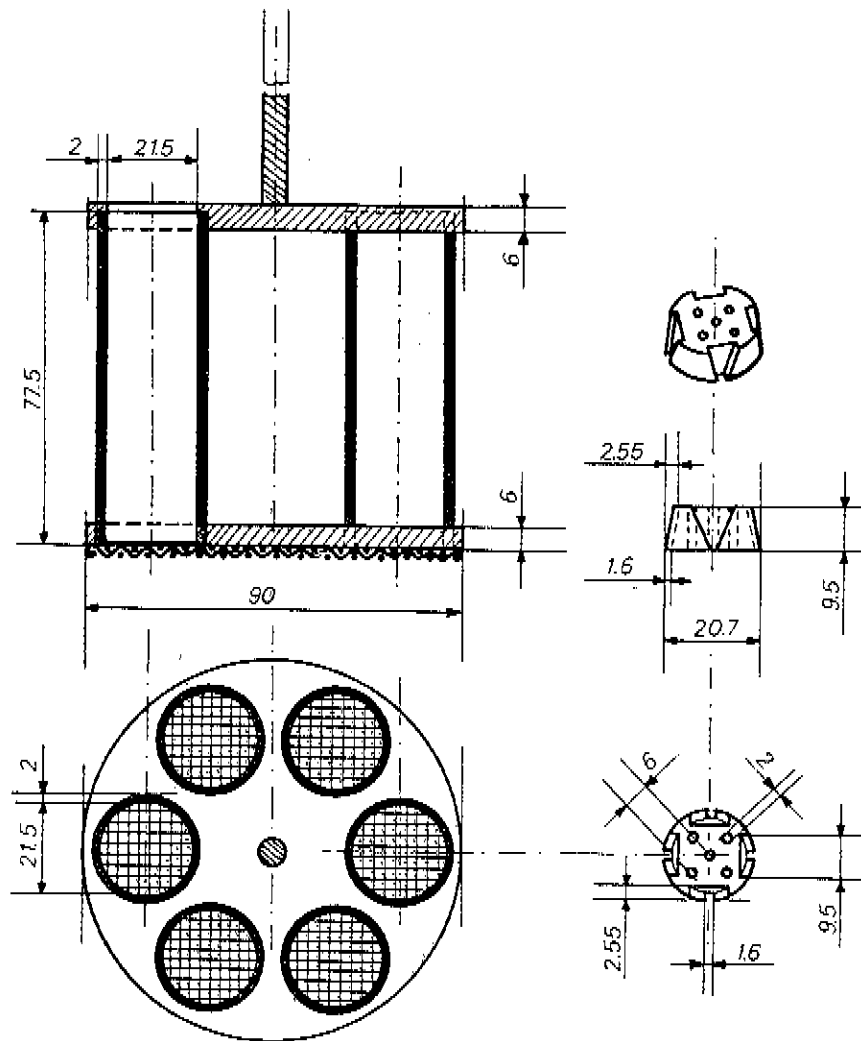
Where a disc is prescribed the following configuration and dimensions apply: a cylindrical disc 20.7 ± 0.15 mm in diameter and 9.5 ± 0.15 mm thick, made of transparent plastic with a relative density of 1.18 to 1.20. Each disc is pierced by five holes 2 mm in diameter, one in the centre and the other four spaced equally on a circle of radius 6 mm from the centre of the disc. On the lateral surface of the disc, four equally spaced grooves are cut in such a way that at the upper surface of the disc they are 9.5 mm wide and 2.55 mm deep and at the lower surface 1.6 mm square.

RECOMMENDED PROCEDURE
(except for effervescent tablets)

Unless otherwise specified in the individual monograph, use water maintained at 37 ± 2 °C as the immersion fluid. Place one tablet or capsule in each of the 6 tubes, and where prescribed in individual monographs add a disc to each tube. Operate the apparatus for the prescribed period, then withdraw the assembly and examine the state of the tablets or capsules. To pass the test all 6 tablets or capsules must have disintegrated.

FIGURE

BASKET-RACK ASSEMBLY OF THE
DISINTEGRATION APPARATUS



Dimensions in millimetres

6. UNIFORMITY OF CONTENT FOR SINGLE DOSE PREPARATIONS

This test should be applied only where the declared quantity of drug substance in tablets or capsules is 5 % or less of the total formulation, or in the case of sugar-coated and enteric-coated tablets, where the test for "Uniformity of Mass for Single Dose Preparations" does not apply, or for certain powders for injections when specified in the individual monograph.

RECOMMENDED PROCEDURE

Individually, determine the amount of drug substance in each of 10 units using the analytical method specified in the individual monograph or, where no method is stated specifically for Uniformity of Content, use the analytical method described under Assay.

For tablets and powders for injections

Each single unit contains an amount of drug substance within ± 15 % of the average amount. If one individual unit contains an amount of drug substance outside ± 15 % but within ± 25 % of the average amount, examine a further 20 units drawn from the same population as the original 10 units. The preparation being examined complies with the test if not more than one unit from the total of 30 units examined contains an amount of drug substance outside ± 15 % and none contains an amount outside of ± 25 % of the average amount.

For capsules

Not more than one unit contains an amount of drug substance outside ± 15 % and none contains an amount outside of ± 25 % of the average amount. If 2 or 3 individual units contain an amount of drug substance outside ± 15 % but within ± 25 % of the average amount, examine a further 20 units drawn from the same population as the original 10 units. The preparation being examined complies with the test if not more than 3 units from the total of 30 units examined contain an amount of drug substance outside ± 15 % and none contains an amount outside ± 25 % of the average amount.

7. UNIFORMITY OF MASS FOR SINGLE DOSE PREPARATIONS

Tablets

Uncoated tablets and film-coated tablets formulated to contain 5 % or more of the drug substance comply with the following test:

RECOMMENDED PROCEDURE

Weigh 20 tablets and calculate the average mass. When weighed singly, the deviation of individual masses from the average mass do not exceed the limits given below.

Average mass of tablet	Number of tablets	Deviation %
80 mg or less	not less than 18	±10.0
	not more than 2	±20.0
More than 80 mg and less than 250 mg	not less than 18	± 7.5
	not more than 2	±15.0
250 mg and over	not less than 18	± 5.0
	not more than 2	±10.0

If film-coated tablets fail this test it may be because of variability in the thickness (mass) of the coatings. In such a case a test for uniformity of content should be applied, and if the tablets meet the requirement of this test, they can be considered acceptable.

Capsules

RECOMMENDED PROCEDURE

Weigh 20 intact capsules individually, and calculate the average mass. The mass of each capsule should be within ±10 % of the average mass. If not all the capsules fall within the aforementioned limits, weigh the 20 capsules individually, taking care to preserve the identity of each capsule, and remove the contents as completely as possible.*

Weigh the emptied shells individually and calculate for each capsule the net mass of its contents by subtracting the mass of the shell from the respective gross mass. Determine the average net content from the sum of the individual net mass. Then determine the difference between each individual net content and the average net content. Deviation of individual net masses from the average net mass do not exceed the limits given below.

Net mass of capsule contents	Number of capsules	Deviation %
Less than 300 mg	not less than 18	±10.0
	not more than 2	±20.0
300 mg and over	not less than 18	± 7.5
	not more than 2	±15.0

* For soft gelatin capsules, wash the shell with ether or some other suitable solvent and allow it to stand until the odour of the solvent is no longer perceptible. Other means such as a jet of compressed air may be used.

Powders for injections

The test applies to powders for injections where the content is more than 40 mg. The recommended procedure is the same as described for capsules. The deviation of individual net masses from the average net mass do not exceed the limit given below.

Number of containers	Deviation %
not less than 18	±10
not more than 2	±20

For preparations with a content of less than 40 mg the test for "Uniformity of Content for Single Dose Preparations" applies instead.

8. TESTS FOR STERILITY*

Sterility testing should be carried out under conditions designed to avoid contamination of the material under test (e.g. by using laminar air flow technique), and any antimicrobial action on possible contaminating organisms. In order to assure adequate performance, it is advisable periodically to include controls in the sterility testing. The testing conditions should meet similar requirements as for manufacturing operations.

The test is applied to substances, preparations or articles which, according to a pharmacopoeia, are required to be sterile. However, a satisfactory test result only indicates that no microbial contaminants have been found in the sample examined in the conditions of the test. The extension of this result to the whole of a batch of a product requires the assurance that every unit in the batch has been prepared in such a manner that there is a high degree of probability that it would also have passed the test. This assurance depends on compliance with Good Manufacturing Practices (GMP), including sterility assurance, throughout the entire manufacturing operation.

Sampling

For the purposes of the test, a batch is defined as a homogeneous collection of sealed containers prepared in such a manner that the risk of microbial contamination is the same for each of the units (e.g. one sterilizer load, or a quantity of containers filled under aseptic conditions in one working session at one work station).

* The tests described are only applicable to parenteral preparations for the International Pharmacopoeia. For implants, ophthalmic and other non-injectable preparations, surgical dressings, catgut and other surgical sutures, other test conditions must be chosen.

<u>Number of items in the batch</u>	<u>Minimum number of items to be tested</u>
Not more than 100 containers	10 % or 4 containers whichever is greater
More than 100 but not more than 500 containers	10 containers
More than 500 containers	2 % or 20 containers whichever is less.

Culture media (see Vol. 1, p. 175-177).

The following culture media have been found to be suitable for the sterility test: Fluid sodium mercaptoacetate and soya-bean casein digest media.

Fluid sodium mercaptoacetate (sodium thioglycolate) medium (culture medium Cm4) is primarily intended for the culture of anaerobic bacteria but will also detect aerobic bacteria. Soya-bean casein digest medium (culture medium Cm5) is primarily intended for the culture of aerobic bacteria but is also suitable for fungi. Other media may be used provided that they have been shown to sustain the growth of a wide range of microorganisms and that they comply with the test for effectiveness of the medium in the presence of the preparation to be tested.

The sterility of each lot of medium should be confirmed by incubation of representative portions of it at the temperature and for the length of time specified in the test.

The growth promoting quality of each new batch of medium should be tested by adding inocula, each containing approximately 100 viable microorganisms, of strains of appropriate microorganisms e.g. Staphylococcus aureus ATCC 6538 P or 6548 (NCIB 8625, CIP 53.156), Bacillus subtilis ATCC 6633, Clostridium sporogenes ATCC 19404, Candida albicans ATCC 2091 or ATCC 10231 to separate representative samples of the medium.

The media thus inoculated with the reference strains, should then be incubated according to the conditions specified in the test. The test media are satisfactory if clear evidence of growth appears in all inoculated media within 7 days.

Detection and suppression of antibacterial effects

Prior to testing, possible antibacterial effects of the substance or material under test should be determined by comparing the nutritive properties of the media in the presence and in the absence of the product under examination. This comparison should be carried out under test conditions using reference strains, as described above in the section "culture media". If the growth of the reference strains is delayed or weakened in the presence of the product examined by direct inoculation, the antibacterial effect should be suppressed by means of filtration, dilution or neutralization before or during the test. The effectiveness of the process should be tested periodically by inoculating the media with reference strains in the final stage of the test.

RECOMMENDED PROCEDURE

The test may be carried out by membrane filtration or by direct inoculation of the media with the preparation under examination. The technique of membrane filtration is to be preferred whenever the nature of the product permits, that is, for filtrable aqueous preparations especially in large volumes, for alcoholic or oily preparations and for preparations miscible or soluble in aqueous or oily solvents which do not themselves have an antimicrobial effect in the conditions of the test. Whichever test is used the media are observed at intervals throughout the specified incubation period.

1. Membrane filtration

Membrane filters with a nominal pore size not greater than 0.45 μm , of assured quality preferably certified by the manufacturer, should be used. Cellulose nitrate filters, for example, are used for aqueous, oily and weakly alcoholic liquids and acetate filters are used, for example, for strongly alcoholic liquids. The technique described assumes that filter discs of about 50 mm in the diameter will be used, the filter assembly having been previously sterilized. If filters of a different diameter are used the volumes of the dilutions and the washings should be adjusted accordingly.

Prior to the test, a small quantity of a suitable sterile diluent such as a 1 g/l neutral solution of meat or casein peptone (culture media Cm1 or Cm5) should be filtered through the prepared filtration apparatus. The content of the container(s) to be examined should be transferred to the prepared apparatus. If possible, the entire content of the containers should be taken or the minimum quantity laid down for the test by direct inoculation. If necessary, the product to be tested may be diluted with the chosen sterile diluent. Filtration should be carried out immediately.

If the product to be tested has antimicrobial properties, the membrane should be washed by filtering through it not less than 3 successive quantities, each of approximately 100 ml, of the chosen sterile diluent, with the addition of a suitable neutralizing substance. Oily liquids may be diluted before filtration with a suitable sterile solvent, such as isopropyl myristate, which has been shown to have minimal antimicrobial effects.

Preferably, one membrane should be transferred to each of the media used, or where this is not possible the one membrane should be cut aseptically into two equal parts, which are then transferred to different media. Culture media with the membranes should be incubated for not less than 7* days at 30-35 °C in the test intended mainly to detect bacteria and at 20-25 °C in the test intended specifically to detect fungi.

2. Direct inoculation

A suitable quantity (see the following tables) of the product under examination, taken from a sealed container, should be directly transferred to culture media intended for the detection of aerobic and anaerobic bacteria and media for the detection of fungi.

* If the nature of the product or the treatment to which it has been subjected is suspected to leave microorganisms of impaired viability, a longer incubation time than the minimum of 7 days may be necessary.

LIQUIDS

Quantity in the container

less than 1 ml
1 ml or more/but less than 4 ml
4 ml or more/but less than 20 ml
20 ml or more (including large
volume parenterals)

Quantity taken and examined in each culture medium for the bacterial and fungal tests.

entire contents of container
half contents of container
2 ml
10 % of contents

SOLIDS

Quantity in container

less than 50 mg
50 mg or more but less than 200 mg
200 mg or more

Quantity for examination

entire contents of container
half contents of container
100 mg

There should be a sufficient quantity of culture medium to ensure that the addition of the product under examination does not affect its nutritive properties. Unless otherwise indicated in the monograph, the product under examination should be diluted approximately ten-fold for liquids and hundred-fold for solids, in order to assure homogeneous distribution and to eliminate antibacterial activity.

In the case of an oily liquid, an emulsifying agent may be added to the culture medium such as 0.5 - 1 per cent m/V of polysorbate 80 or 0.1 per cent m/V of (p-tert-octylphenoxy)polyoxyethanol or other emulsifying agent in appropriate concentration shown not to have any antimicrobial action in the conditions of the test.

The inoculated media should be incubated for not less than 14 days. Media intended mainly for the detection of bacteria should be maintained at a temperature of 30-35 °C and media intended specifically for the detection of fungi should be maintained at a temperature of 20-25 °C. Media containing an oily liquid should be shaken gently at regular intervals. If one medium is used to detect both aerobic and anaerobic bacteria, shaking should be kept to a minimum in order to maintain anaerobic conditions.

Interpretation of results

If no microbial growth appears in any of the culture media by the end of the incubation period, the product complies with the test for sterility.

If microbial growth appears in the initial test cultures and/or in subcultures, the organisms should be isolated and identified. If there is evidence - from the material used, the kind of organisms detected, the monitoring or the control tests - that the test was carried out under inadequate aseptic conditions, the test can be declared invalid and should be repeated with the same corresponding number of items and quantities. In the absence of such evidence, the test may be repeated on twice the number of containers used for the original test. If no microbial growth is detected, the product examined complies with the test for sterility. If microbial growth is detected in the second test, the product examined does not comply with the test for sterility.

9. STERILIZATION METHODS

The term "sterilization" means the complete destruction or removal of all living organisms (including sporing and non-sporing bacteria, viruses, fungi and protozoa). However, statistical considerations preclude the possibility of complete and assured achievement of this absolute state in all cases. For practical purposes, the assurance of sterility is therefore considered in terms of probability; i.e. an article may be regarded as free from contamination at an acceptable level of safety. Methods using saturated steam under pressure or hot air are the classical sterilizing techniques. They are the most reliable and should be used whenever possible. Other sterilization methods include filtration, ionizing radiation (gamma and electron-beam radiation), and gas (ethylene oxide, formaldehyde).

Aseptic processing, although not truly a sterilization method, is widely used for products which cannot be sterilized in the final containers. Materials and products that have been sterilized by one of the above processes, are transferred to pre-sterilized containers and sealed, both operations being carried out under controlled aseptic conditions.

Whatever method is chosen the procedure (process) must be validated for each type of load both with respect to the assurance of sterility and to the integrity of the product.* Failure to follow precisely a defined, validated process could result in a non-sterile or deteriorated product. A typical validation programme for steam or dry heat sterilization requires the correlation of temperature measurements made with sensing devices used to demonstrate heat penetration and heat distribution, with the killing of biological indicators, i.e. preparations of specific microorganisms known to have high resistance to the particular sterilization process. Biological indicators are also used to validate other sterilization methods (see specific methods), and sometimes for routine control of individual cycles. Periodic revalidation is recommended.

The efficacy of any sterilization process depends on the nature of the product, the extent and nature of any contamination present, and the conditions under which the product has been prepared. Throughout all stages of manufacture and sterilization the general requirements for Good Manufacturing Practice (GMP) must be observed.

1. Heating in an autoclave (Steam sterilization)

Under conditions of moist heat, for example saturated steam under pressure, destruction of microorganisms is achieved by the irreversible denaturation of enzymes and structural proteins. The temperature at which denaturation occurs varies inversely with the amount of water present. The sterilization is carried out in saturated steam under pressure (in an autoclave) whilst precisely controlled conditions of time, temperature and pressure are maintained. When displacement of the air by steam is unlikely to be readily achieved, the air should be evacuated from the autoclave before admission of steam. This method should be used whenever possible for aqueous preparations and for surgical dressings and related materials.

* The final product must comply with the requirements of the relevant specific monograph in The International Pharmacopoeia.

The preferred conditions for sterilization in an autoclave are 15 minutes at 121-124 °C (200 kPa)*. Alternative conditions, with different combinations of time and temperature, are given below.

<u>time</u>	<u>Temperature</u> in °C	<u>Approximately</u> <u>corresponding</u> <u>pressure (in kPa)</u>	<u>Minimum sterilization</u> <u>(in min)</u>
	126-129	250 (~2.5 atm)	10
	134-138	300 (~3.0 atm)	5

Minimum sterilization time should be counted from the moment when the required temperature reaches the coolest part of the load, and penetrates throughout the articles to be sterilized (e.g. for aqueous solutions filled into glass containers, the heat equilibration corresponds to 10 minutes for volumes up to 100 ml and 20 minutes for volumes up to 1000 ml). Monitoring of the physical conditions within the autoclave during the sterilization procedure at ± 2 °C and ± 10 kPa (0.1 atm) is essential. To this end, temperature-sensing probes inserted into representative containers, together with additional probes placed at the potentially coolest parts of the loaded chamber (as established in the course of the validation programme) should be used to provide and record the required information. Each cycle should be recorded on a time-temperature chart or by other suitable means.

Porous loads such as surgical dressings and related products should be processed in an equipment suitable for assuring steam penetration. Most dressings are sterilized by maintaining a temperature of 134 - 138 °C for 5 minutes. Some articles made of glass, porcelain or metal are sterilized at 121-124 °C for 20 minutes. Fats and oils may be sterilized at 121 °C for two hours. However, whenever possible fats and oils should be sterilized by dry heat.

In certain cases, e.g. thermolabile substances, sterilization may be carried out at temperatures below 121 °C, provided the chosen combination of time and temperature has been validated. Although lower temperatures offer a lower margin of safety, they may be satisfactory, depending on the nature and extent of the pre-sterilization bio-burden. Special conditions of temperature and time for certain preparations are stated in individual monographs.

The following bioindicator strains are proposed for the validation of the process: spores of Bacillus stearothermophilus (e.g. ATCC 7953, or CIP 52.81 presenting a D-value* of 1.5-2 minutes at 121 °C using about 10^6 spores per indicator).

2. Dry heat sterilization

In dry heat processes the low moisture content is a limiting factor and the primary lethal process is considered to be oxidation of cell constituents. Dry heat sterilization requires a higher temperature and

* 1 atm = 101 325 Pa

The pressure is used solely to obtain the required steam temperature. Therefore, the temperature and not the pressure must be used to control and monitor the process.

a longer exposure time than is required for moist heat. The method should therefore be used only for heat-stable, non-aqueous materials which cannot be sterilized by steam because of deleterious effects or failure to penetrate. Such materials include oils and some oily injections, powders and glassware.

Preparations to be sterilized by dry heat are distributed in containers which are then either finally sealed or temporarily closed to prevent recontamination. The entire content of each container is maintained in the oven for an effective combination of time and temperature. The following conditions may be used, but other combinations may be necessary for other preparations:

<u>Temperature</u> (in °C)	<u>Minimum sterilization time</u> (in min)
160	180
170	60
180	30

Special conditions of temperature and time for certain preparations are stated in individual monographs.

The oven should normally be equipped with forced air circulation to assure even distribution of heat, also within the articles processed. This should be monitored by temperature measurements. Containers which have been temporarily closed during the sterilization procedure are finally sealed using aseptic techniques so as to prevent microbial recontamination.

The following bioindicator strains are proposed for the validation of the process: Spores of Bacillus subtilis (e.g. var. niger ATCC 9372 or CIP 77.18 presenting a D-value of 5-10 minutes at 160 °C using about 10⁶ spores per indicator).

3. Filtration

Sterilization by filtration is widely used for thermolabile solutions. These may be sterilized by passage through sterile bacteria-retaining filters, e.g. membrane filters, adsorptive filters (cellulose derivatives etc.), plastic, porous ceramic or suitable sintered glass filters, or combinations of these. Asbestos containing filters should not be used.

Appropriate measures should be taken to avoid loss of solute by adsorption onto the filter and to avoid the release of contaminants from the filter. Suitable filters will prevent the passage of microorganisms, but the filtration must be followed by an aseptic transfer of the sterilized solution to the final containers which are then immediately sealed to exclude microorganisms. Great care must be taken to avoid recontamination at this stage.

* The D-value is the parameter for a 90 % reduction of the microbial population.

Usually, membranes of not greater than 0.22 μm pore size should be used. The effectiveness of the filtration method must be validated if larger pore sizes are used.

To confirm the integrity of filters both before and after filtration a bubble point or similar test should be used, in accordance with the filter manufacturer's instructions. (By means of this method, it is determined that a prescribed pressure is necessary to force air bubbles through the intact membrane wetted with either product, water or a hydrocarbon liquid.)

All filters and all pipelines and equipment "downstream" used must be sterile. Filters that withstand heat may be sterilized in the assembly in an autoclave before use at 121 °C for 15 to 45 minutes depending on the size of the filter assembly. The effectiveness of this sterilization should be validated. For filtration of a liquid in which microbial growth is possible, the same filter should not be used for procedures lasting for more than 1 working day.

4. Exposure to ionizing radiation*

Sterilization of some drug substances and drug products in their final container or package, as well as medical devices, may be achieved by exposure to ionising radiation in the form of gamma radiation from a suitable radioisotopic source such as ^{60}Co (cobalt 60) or of electrons by a suitable electron accelerator.

Gamma and electron-beams effect ionization of molecules in the organisms. Mutations are thus formed in the DNA. These reactions disturb the natural replication. Only persons well-trained and experienced in the field should decide upon the need, carry out and monitor the radiation sterilization processes, using specially designed and purpose-built plant and equipment.

Twenty-five kGy** (2.5 Mrad)*** of absorbed radiation is usually selected, although other level of radiation may be employed provided that they have been validated.

The radiation doses should be monitored during the whole process with specific dosimeters. The radiation system should be reviewed and validated whenever the source material is changed and, in any case, at least once a year.

The efficiency of sterilization can be proven by bioindicators (proposed are: Spores of Bacillus pumilus (e.g. ATCC 14884 or CIP 3.83 if 25 kGy (2.5 Mrad) are used, presenting a D-value of about 3 kGy (0.3 Mrad) using 10^7 to 10^8 spores per indicator) and spores of Bacillus cereus (e.g. SSI C 1/1) or Bacillus sphaericus (e.g. SSI C₁A) if higher doses are used.

* Laws or regulations for protection against radiation have to be respected in most countries.

** kiloGray

*** Megarad

5. Gas sterilization

The active agent of this sterilization process can be a gas or a highly volatile substance. The substance generally employed in gaseous sterilization is ethylene oxide. Among the disadvantages of this agent are its highly flammable and potentially explosive nature, unless mixed with suitable inert gases, its highly toxic properties and the possibility of toxic residues in treated materials. The whole process is difficult to control and should be only considered if it is not possible to use another sterilization procedure. It must be carried out only under the supervision of staff skilled in the process.

The sterilizing efficiency of ethylene oxide is related to the concentration of the gas, the humidity, the time of exposure, the temperature and the nature of the load. In particular it is necessary to ensure that the nature of the packaging is such that the gas exchange can take place. Records of these parameters should be made for each cycle. Appropriate sterilization conditions must be determined experimentally for each type of load.

After sterilization, time should be allowed for the dispersal of residual sterilizing agent and other volatile residues. This should be confirmed by determining the residual agents by specific tests.

As the process is difficult to control and to monitor, the efficiency should be controlled each time using bioindicators (proposed are: Spores of Bacillus subtilis, e.g. var. niger ATCC 9372 or CIP 77.18, or spores of Bacillus stearothermophilus, e.g. ATCC 7953 or CIP 52.81. The same amount of spores is needed as for other methods - see section 1 and 2.I It is also important that the humidification is sufficient for the sterilization process).

* * *