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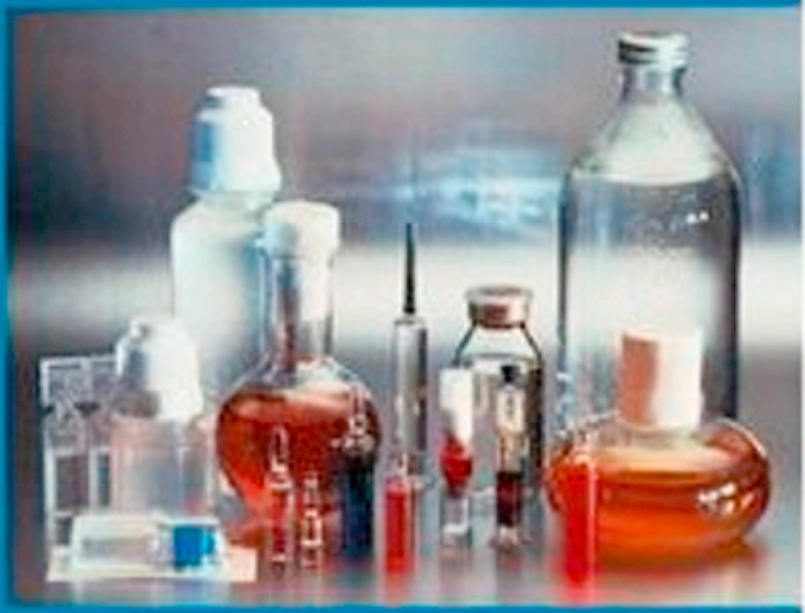
Matts Ramstorp

Contamination Control in Practice

Filtration and Sterilisation



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Contamination Control in Practice. Matts Ramstorp

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Contamination Control in Practice

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Preface

Contamination control is a modern and fast growing multidisciplinary science and development in the field is accelerating rapidly, keeping pace with both technical market and governmental demands. Every co-worker within a company is important and must have knowledge covering and overlapping several different technical areas. One demand put on co-workers is to have the ability to view sub-parts from a greater perspective, i.e. to have a holistic view of their work situation.

This book supplies such a holistic view of knowledge. The cleanliness of products or process flows is often complex in nature and very hard to define in easily understood terms. This book deals with the overall cleanliness of products and process flows and covers different techniques utilised in order to achieve and maintain certain cleanliness levels.

It not only covers cleanliness from a microbiological point of view but also in regard to other types of particle contaminants, those particles that in the literature are often described as dead particles.

The book may be read as an isolated textbook but it is also one of a series of three books covering contamination control. It supplies basic knowledge for branches of industry working with increased demands for cleanliness, for instance cleanliness of water, steam, pressurised gases and different flows in a process, together with finished products and products to be transferred into vials.

I would like to acknowledge the neverending support from my dear family. Pia, Anna and Johan, I love you and I do appreciate the way you always try to put my focus back on what I am supposed and expected to do. I also wish to express my gratitude to Mrs Camilla Dahl for all the help supplied in finishing this manuscript. I would also like to thank Mr. Alf

Gustafsson who wrote the part on sterilisation in the Swedish version of this book.

The purpose has not been to write a book that fully covers the subject but rather one that should be a good basis for both beginners and people with long experience and also for those needing a summary of all cleaning techniques used for products and process flows.

Malmö, October 2002

Matts Ramstorp Ph D,
Professor in Contamination
Control and Cleanroom
Technology

1 Contamination Control

1.1 Introduction

During the last two decades the overall concept of contamination control has gained greater interest and found increasing use within several branches of industry. The microelectronic industry and the pharmaceutical industry use the total concept of contamination control, whereas, for example, the food and beverage industry only uses one or several parts of contamination control as technical support in a certain process or for a certain product.

Contamination control is a multidisciplinary science constructed of a great number of different sub-parts that have been quite well known, from a scientific point of view, for a long period of time. The novelty is the holistic application of the different parts of the technique in association with research and development, production, control etc.

As the use of contamination control increases, the demand for knowledge will also increase. More and more individuals of different categories will work with the technology, making knowledge and most of all a thorough understanding of why it is necessary to work in a particular way, an important demand.

The first book in this series, *Introduction to Contamination Control and Cleanroom Technology* (2000), covers the holistic approach to contamination control. This second book takes the subject a step further by focusing on the cleanliness of products and other types of process fluid.

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1.2 Contamination Control – A Holistic Technique

The major purpose of contamination control is to control different, and especially critical, contaminants to prevent them negatively influencing products, processes and people. In the first book in this series the various sub-parts of contamination control are discussed: the different types of contaminants that can be observed together with how these contaminants are detected and analysed. The first book also covers clean rooms and clean zones, cleaning and decontamination, together with cleanroom clothing and personal responsibility during the handling and maintenance of a clean and hygienic production process.

In this book the different types of contaminants are only summarised, as shown in Fig. 1. This figure shows in a schematic and holistic way the interrelationship between various activities in a clean environment, i.e. how contaminants may arise, how they may be dispersed and transported away from their source and finally where they may be deposited. This second book focuses on how to protect products and processes from being contaminated by substances outside the process and also on different ways to take care of and control contaminants, either by eliminating them from a product or a process flow (through filtration), or by rendering them harmless (through sterilisation by moist heat).

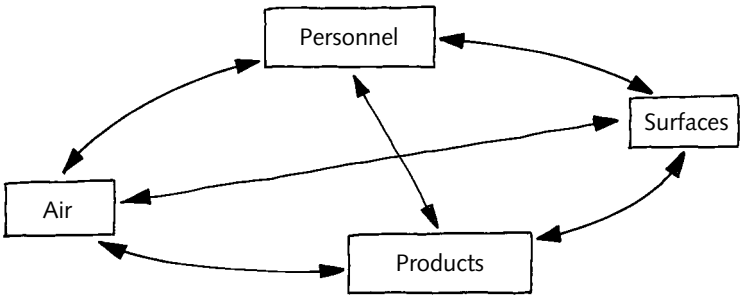

Figure 1. The interaction between the four major areas of contamination control. The figure shows schematically how the personnel can have an impact on the cleanliness of the air, the surfaces and the products. In the same manner the figure shows how the cleanliness of the air may affect the cleanliness of the personnel, the surfaces and the products handled, etc.

1.3 Source, Dispersion and Deposition of Contaminants

A contaminant is generally defined as a solid, liquid or gaseous substance, or a physical state that is found in the wrong place and (or) at the wrong time. Contaminants can be divided into solid material (particles), liquid material (often residual product or residual cleaning agents) and gaseous materials, together with different physical states, that in a negative way might influence product, process or man.

In this book the major focus is on particles, i.e. contaminants comprised of solid material. Particles can be divided into two major categories, dead particles and microorganisms, often simply called ›live particles‹.

1.3.1 Dead Particles

Dead particles are comprised of solid material and have no possibility to reproduce by themselves to increase in number. Particles can be transported by air in the form of soil, sand, particles from automobile exhaust, exhaust gases from industry or as fibers from insulating materials or textile materials. Dead particles can also be generated by humans in the form of scales of skin. All of these particles can be transported into a production area where, after suspension in the room air, they will finally end up on more or less critical surfaces. Dead particles cannot reproduce by themselves, but microorganisms attached to their surface can in many cases later become a problem. Solid contaminants are normally called particles. A particle is defined according to the US Federal Standard 209 as something solid or liquid in the size range 10^{-3} to 10^{3} µm (1 µm = 10^{-3} mm). The same standard also defines the word fiber. A fiber is defined as a particle, according to the above definition, but with a size ratio length/width ≥ 10.

Particles can be characterized in different ways. Traditionally particles are characterized according to their size. Generally, particle size is presented in one dimension only, for example particles found in a product are said to have a size of 5 µm or 50 µm. Such a characterization might be interpreted to mean that all particles have only one dimension, i.e. that they are totally spherical. This is however frequently not the case. Most particles have a three-dimensional extension, i.e. they have different *x*-, *y*- and *z*-dimensions. The reason why a particle is reported as having only one dimension is that

Table 1. Sedimentation time for different materials and particles. The table shows the time taken for the particles to fall 1 m in water.

Particle size /mm	Material	Time to fall
10	Gravel	1 s
1	Sand	10 s
0.1	Fine sand	2 min
0.01	Clay	2 h
0.001	Bacteria	8 days
0.0001	Colloidal particles	2 years
0.000001	Colloidal particles	20 years

particles within contamination control are measured with instruments or with techniques only giving the particle size in one dimension. This situation can occasionally be very difficult to handle, especially for instance when choosing filters based on tests performed by a filter manufacturer.

Larger particles are, due to their mass in conjunction with the gravity force of the earth, harder to disperse and also to transport over long distances in air. When deposition as well as accumulation of particles takes place, for instance in a product, the size as well as the mass of the particle plays a vital role. The larger the particles, the easier they are to eliminate with the aid of a filter or by simply letting the product stand for a while, allowing the particles to settle to the bottom under the influence of gravity. In order to give a better perspective on how the size of particles, or more accurately, their respective mass, will affect them in a liquid as well as in air, some data is presented in Table 1. This table shows the relationship between the size of a particle and the velocity at which the particles will sediment in water and air, respectively.

This table shows how rapidly particles suspended in a water solution will move during a sedimentation process. The larger the particles the faster they will move through the liquid under the influence of gravity. Particles that are smaller will take much longer to travel a corresponding distance in water. If a particle is small enough, the velocity will be so minute and nearly non-existent that the particle can be considered to be

floating free in the solution. The different particles mentioned in the table have been studied in a pure water solution. When studying the same situation in a viscous solution, i.e. a liquid that is ›thicker‹ and flows much less readily, e.g., concentrated sugar solution, the time required for the particle to move a corresponding distance will increase dramatically.

1.3.2 Microorganisms

Microbiology is the science of microorganisms, which is the science dealing with small living organisms (Latin: micros = small, bios = life, logi = science). Microbiology deals with organisms that are so small that they cannot be viewed with the naked eye. In order to study microorganisms they must be enlarged, for instance by the aid of a microscope, up to one thousand times or sometimes several thousand times, Table 2. Sometimes microorganisms are also called microbes.

Within contamination control, microorganisms are normally characterized as contaminants with the ability to reproduce themselves and thereby increase in number. Microorganisms are found everywhere and this is one of the primary reasons why they are considered to be a problem during clean and hygienic production. Microorganisms are often found in environments where other life forms are present, but they are also found in environments where normal life forms cannot survive. Naturally occurring microorganisms can grow

Table 2. The approximate size of some biological and non-biological materials.

Length of intestinal villi	1 mm (1000 µm)
Human egg cell	0.1 mm (100 µm)
Diameter of human hair	0.07–0.1 mm (70–100 µm)
Visibility border for human eye	0.04 mm (40 µm)
Protozoa	0.01 mm (10 µm)
White blood cells	0.01 mm (10 µm)
Red blood cells	0.007–0.008 mm (7–8 µm)
Bacteria	0.001 mm (1 µm)
Viruses	0.1 µm (100 nm, 1000 Å)
Protein molecules	0.01 µm (10 nm, 100 Å)
Amino acids	0.001 µm (1 nm, 10 Å)
Single atoms	0.0001 µm (0.1 nm, 1 Å)

1 mm = 1000 µm, 1 µm = 1000 nm, 1 nm = 10 Å

extensively and thereby increase rapidly in number in a quite short time interval. In one cubic centimeter of soil, up to 1 billion bacteria can be found. This corresponds to approximately 3% of the total weight of the soil sample. The human body is a large carrier of microorganisms. All surfaces of the body, interior as well as exterior, are more or less covered with microorganisms. As many as one million bacteria can be found on one square centimeter of the surface of the skin. Saliva contains approximately one billion microorganisms per milliliter. By far the greatest amount of microorganisms in conjunction with human beings are found in excrement, where as many as one hundred billion bacteria per gram can be found, corresponding to approximately 50% of the total dry weight of our excrement.

1.4 The World of Microorganisms

From a totally non-scientific point of view microorganisms can be divided into five characteristic subgroups. These subgroups are intended to show where, and most of all why, the various microorganisms are found in different environments and on different occasions.

The first group, called *essential microorganisms*, are responsible for all the reactions and actions taking place in nature, i.e. the microorganisms that are active in the degradation processes of organic material in nature.

The second group, *usable microorganisms*, are frequently utilised for production of different types of food products and pharmaceutical products, i.e. through different fermentation processes.

The third group, *harmless microorganisms*, consist of organisms that are normally found in the digestive tracts of our body, where they play an important role in the metabolism.

The fourth group are called *harmful microorganisms*. These organisms have the possibility to destroy different types of products, for instance products produced by the so-called useable microorganisms, making these products taste, smell and/or look bad. In some systems this degeneration can lead to the formation of different toxins, which in turn can negatively influence man.

Finally the fifth group, *dangerous microorganisms*, which are

the organisms responsible for diseases in a host organism. These organisms are often called pathogenic organisms.

It is of vital importance to acknowledge the definition stated earlier for contaminants, i.e. that contaminants are something that are in the wrong place and/or on the wrong occasion and that might negatively influence products, processes and (or) man. This is the reason why harmless organisms are only harmless when they are located where they are expected to be and at the right time, for instance in the intestinal tract.

A more scientific way to describe various microorganisms is in terms of:

- Bacteria
- Algae
- Fungi
- Protozoa
- Viruses

Even if all microorganisms can be divided into different categories they have many general properties in common. They need three overall environmental states in order to survive and also in order to be able to reproduce, namely:

- Food
- Moisture
- Correct temperature

1.4.1 Bacteria

Bacteria are a large group of microorganisms comprising several thousand species of unicellular organisms. Bacteria are found everywhere, in the air, in water and in soil and they are also present in living and dead higher organisms. In order to reproduce and develop they need moisture. Despite this fact, there are many microorganisms that can survive dryness, which generally means that the movement of air can transport them in an unaffected state for quite a long distance. Some bacteria are called aerobic, i.e. they only develop in the presence of oxygen. Other bacteria are anaerobic, which means that they only live and reproduce in an environment that is free of oxygen.

Different bacteria develop at different temperatures. There is always an optimum temperature for each type of bacteria.

The pathogenic bacteria generally develop best at a temperature similar to that of the living body, the host. At lower as well as at higher temperature, their ability to develop will decrease. Bacteria are quite sensitive to higher temperatures and many bacteria will die at 60 °C and higher. Several bacteria have the ability to form endospores, often just called spores. These bacteria often belong to a very resistant form of *Bacillus* and *Clostridium* and they can resist boiling water for several hours and also resist drying out.

Many of the disinfecting agents that are used in order to control microorganisms will leave the spores in an uninfluenced state. When kept in a dry state bacterial spores can live for several decades. When these dry bacterial spores later come into contact with a suitable environment (an environment containing food, moisture and at an optimal temperature), they will leave the spore state and return to the vegetative state, after which they can start reproducing again. When an object, process flow or a product is to be sterilised, which means that the surface, the gas or the liquid must be totally freed from all living organisms, there is a demand for techniques and methods powerful enough to eliminate and destroy not only the vegetative organisms, but also the spores.

The bacteria are the largest group by number both in the surroundings and on man. This is why they also are the largest and most common contamination sources in regard to microorganisms during clean and hygienic work.

Bacteria have different shapes. Rods or bacilli are straight and can be short or long; they can have rounded edges or square edges. Spheres or coccies are spherical and have a large ability to be collected together with other cells of the same type. The coccies exist as pairs, i.e. two and two, and are then called diplococcies. The appearance of these cell pairs is that they are slightly pulled in in the middle when they are stacked together, making them look rather like coffee beans. Another group of bacteria is the spiral shaped bacteria. The vibrios, looking like a comma sign, are part of this group. The spiral formed bacteria, the spirills, consist of long and rigid spirals whereas spirochetes and treponemer look like long spiral-shaped bacillus.

1.4.2 Algae

The algae differ from the other microorganisms mostly in their ability to transform light into chemical energy (through the biochemical reaction system called photosynthesis). This means that the algae are in general quite like plants. Certain bacteria can also perform photosynthesis. It is not fully clear where the algae belong within biology (the science of life). The algae are in many respects like plants or even more like protozoa. From a physical point of view the algae may be some micrometers in length and up to several hundred meters in length when they are found in the ocean.

Since the algae are dependent on water and have the ability to perform photosynthesis, they are nearly always found in environments with excess water and light. Algae are normally not considered as a problem in cleanroom technology but they can however become a problem, for instance in circulating water systems.

1.4.3 Fungi

Fungi exist as molds and yeasts. Molds are often observed as a green-grey network present on the surface of food products that have been left standing for a long period of time at room temperature. The organisms visible on the food product consist of a multicellular network that is characteristic for molds. The network form can spread extensively and in the outer parts spores are formed, comprised of highly specialised cells that are active in association with reproduction, i.e. development and spreading out of mold spores.

When bacteria form spores, one cell only forms one spore. A single mold cell can, during spore formation, form thousands of spores each of which can be freed into the air and transported far away from the source. When such a spore reaches a site where there is excess food and moisture, the mold spore will leave the spore form and become a vegetative cell that in turn can form a new multicellular network.

Mold spores are somewhat more resistant towards changes in the environmental system than vegetative cells in a multicellular network. Mold spores are however not as resistant as bacterial spores, which means that various methods that negatively influence bacterial spores will also negatively influence mold spores. In other words sterilisation techniques or methods capable of killing bacterial spores will also kill mold spores.

Molds are generally considered as critical contaminants in clean production. They can in many cases present a great threat, because they live on almost everything and do not always demand high levels of humidity to survive and reproduce.

Yeast is a second form of fungi. It is comprised of single cells, often spherical or oval in shape. When looked at with a microscope, yeast cells quite resemble the cells of bacteria. They are, however, larger than bacteria, normally between 5 and 8 μm in diameter. Yeast also has the ability to form spores as a part of its reproduction process. Yeast cells have many critical and specific demands in regard to nutrition and water in order to develop. That is why they are not normally defined as critical contaminants during clean and hygienic production. However, yeast can be found in certain products, mostly those containing high levels of water and carbohydrate.

1.4.4 Protozoa

More than 40,000 protozoa are known today. Most of them are found in natural water environments like lakes, other water sources and marshlands. The role of protozoa in nature is to take an active part in the degradation of organic material and they are therefore an important part of the natural evolution. These unicellular and quite complex microorganisms live on bacteria and other smaller microorganisms. They are also food for other larger and more complex organisms. Since protozoa have a very strong demand for natural water systems, they are not often considered as a problem in clean production and cleanrooms.

1.4.5 Viruses

Viruses form a special group among the microorganisms since they totally lack similarity to the other organisms. This is because they do not occur as normal cells. They are comprised of a single strain of DNA or RNA (not both) that is immobilised within a surrounding protein coat. Viruses are quite a lot smaller than other types of microorganisms.

Viruses are sometimes called obligate intracellular parasites, which is a very good way to describe their way of living. In order to reproduce, the virus particle must first find a suitable host cell to invade. The virus will fix itself to the outer surface

of the host cell, after which the virus content, DNA or RNA, is injected into the host cell. The ribonucleic acid will then start to replicate, i.e. it will start to copy in order to create more identical DNA/RNA molecules within the host cell. After this replication the outer virus components, the surrounding protein shell of the virus, will be produced within the host cell and when everything is complete and new viruses have been formed within the host cell, these will be released into the surroundings. This process can take some time, ranging from less than 20 minutes to several hours or days. Generally viruses are not considered as a great threat when working in cleanrooms.

1.5 The Growth of Microorganisms

A good example of microbiological growth is the growth of bacteria. When bacteria are placed in a suitable environment they will, after some initial time, start to reproduce. The reproduction occurs in such a way that each cell divides into two new cells, a process commonly known as mitosis. The initial cell, called the mother cell, will divide to form two totally new and identical cells, called daughter cells. The type and concentration of nutrients in the surrounding environment plays a vital role for cell division. A mother cell can form two daughter cells every 20 minutes. This means that the number of bacteria in a solution containing the right nutrients and kept at an optimal temperature can increase rapidly after just a short time period, see Tab. 3. This table shows that the growth of microorganisms is nearly like an explosion. If this growth is plotted in a traditional diagram, lin-lin (linear-linear), the growth curve will have a very special appearance, see Fig. 2A. After some time the growth can no longer be studied since the growth curve will spread outside the diagram. This is the reason why a lin-log (linear-logarithmic) diagram is used instead, see Fig. 2B.

1.5.1 Growth Systems

Bacterial growth can be studied in two different systems. The first is comprised of a limited volume of water-based growth media to which a certain amount of nutrients has been added. The other type consists of a volume that is kept constant by continuously renewing the growth media in the system and at

Table 3. Growth of microorganisms through cell division.

Time (hours)	Number of bacteria (theoretical values)
0	1
1	16
2	250
3	40964
4	65536
5	1048576
6	16777216
7	268435456
8	4294967296
9	34359738368
10	549755813888

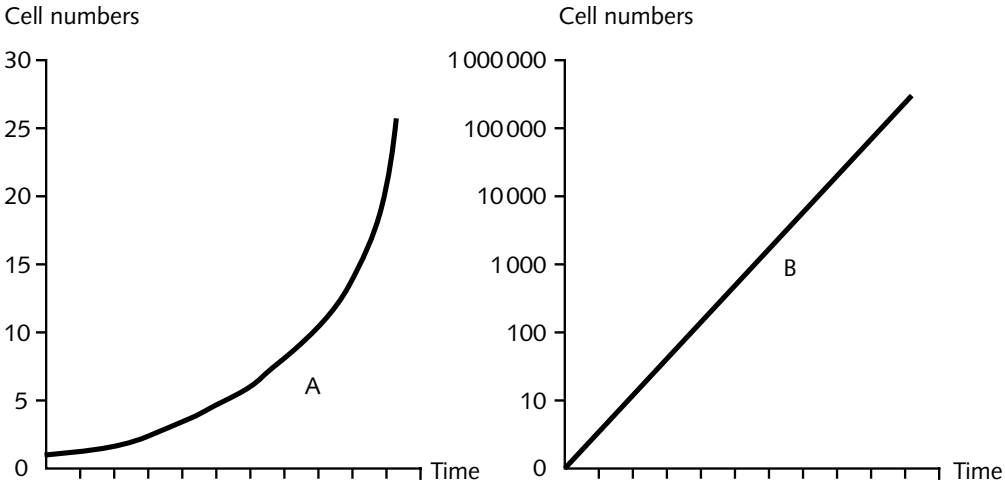

Figure 2. Two ways to describe the growth of microorganisms. The growth is illustrated in a linear – linear diagram (A) and a linear-logarithmic diagram (B).

the same time removing the same volume from the system. The first system, comprised of a limited volume of liquid is normally called a batch system. The second system, in which there is continuous addition and removal of liquid, is called a continuous system. From a practical point of view the batch system represents the situation when, for example, a product is left untouched for a long period of time: a tank filled with a

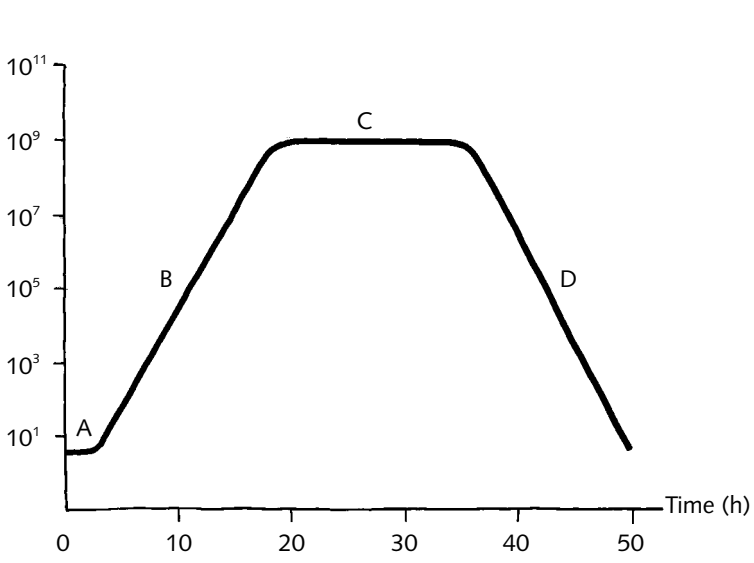

Figure 3. A schematic representation of the growth curve of bacteria. This figure shows the growth curve for bacteria in an environment containing a limited amount of water and nutrients. The curve can be divided into four distinct parts: A, the lag phase; B, the logarithmic growth phase; C, the stationary phase and D, the declination phase.

product to which microorganisms have had access. The continuous system represents a process of continuous production of a product to which microorganisms have been added (i.e. a continuous fermentation process). Let us study the batch system. When organisms are added to such a system they will follow a path consisting of four stages (Fig. 3):

- The lag phase
- The logarithmic growth phase
- The stationary phase
- The declination phase

During the lag phase (Fig. 3A) the microorganisms will come into contact with the new environment in which they are going to live and reproduce. During this initial phase hardly any new cells are formed. Despite this fact, the activity in the cells is very high since they are adjusting to their new environ-

ment. The cells are adjusting in order to cope with the environment, in regard to correct temperature, pH, ion concentration and also the availability of nutrition.

After the cells have adjusted to the existing environment, the logarithmic growth phase starts (Fig. 3B). During this second phase the cells multiply, i.e. one mother cell will divide in order to create two identical daughter cells. This means that one cell becomes two. In the next step these two cells become four, four cells become eight and so forth. During this phase the cells will extract nutrients from the surrounding liquid in order not only to create energy for reproduction, but also to produce building blocks from the nutrient in order to create the new cells. During this phase and all other life cycle phases of the cells waste products are produced. These waste products must in one way or another, exactly as for humans, leave the cells, which in practice means that the cells will continuously release different forms of waste products into the surrounding solution. This means that the solution in which the cells are living will become poorer and poorer in regard to nutrients (since nutrients are not added to the solution during the growth process) at the same time as the surrounding solution becomes richer and richer in waste products. The waste products produced by the cells are often toxic or in another way negatively affect further development of the cells.

After a certain time, depending on the batch volume and the concentration of nutrients, cell growth will decrease and the next phase, the stationary phase, will be entered (Fig. 3C). During this third phase the number of new cells formed corresponds to the number of cells that are dying. At the end of this phase, the concentration of nutrients is extremely low, at the same time as the concentration of waste products is extremely high, which in practice means that the cells are not feeling too good.

After some time the generation of new cells will further decrease and the declination phase or, as it is sometimes called, the logarithmic death phase (Fig. 3D) will be entered. This phase can be viewed as the reverse of the logarithmic growth phase, for instance 256 cells in a solution become 128, 128 cells become 64, 64 cells become 32 etc. It is not true that

zero (0) cells in the solution can be reached, unless any form of action is taken to sterilise the solution.

The continuous system follows, in general, the same initial path as the batch system, i.e. the cells will pass through the lag phase, and will enter the logarithmic growth phase in order to finally end up in the stationary phase. All this will happen if there is nutrition, humidity and optimal temperature available, together with correct adjustment of the flow of growth media through the system.

1.6 Detection of Contaminants

The general reason for the detection of contaminants is to prove either their existence or non-existence at various occasions. Dead particles can be detected and analysed by taking a sample from a product or a process flow. In most process systems the concentration of particles in a product or a process flow will be extremely low, making the particles difficult to study even under a microscope. A much better way is to take a sample, i.e. a certain amount of liquid or gas and filter the sample through a very fine filter, normally called an analysis membrane. After filtration the filter is viewed under a microscope. The advantage of this method is that it is quite easy to perform and especially that it not only shows whether there are particles or not, but also often indicates what type of particles are present in the fluid. It is, furthermore, possible to decide whether the particles are just particles or fibers and it is also, in most cases, possible to decide the contamination source. In this way it is possible to detect fibers released from wound filters being used too long in a process, or fibers from a filter that is not chemically and (or) physically compatible with the process. Residues of metal arising from the pipe network and particles from loosely bound filters, such as sand filters and other types of bed-type filters, can be detected.

Microorganisms are harder to detect with the same type of methodology. This is even the case when the microorganisms are present in a large amount in a sample. The reason for this is that microorganisms contain so much water that they, when viewed under a microscope, tend to become totally transparent. To facilitate the detection of microorganisms different types of staining techniques have been developed. The micro-

biological sample is treated with different types of synthetic dyes that stain the cells or part of the cells, after which they can be observed under the microscope. The most well known method for staining bacteria was developed by the Danish physician Gram. This method divides bacteria into two major groups, Gram positive and Gram negative. When performing a Gram staining para-rosaniline is added to the sample and then washed away with alcohol. Gram positive bacteria will acquire a blue color whereas the Gram negative bacteria will not be stained by the dye.

Traditional microbiological analysis utilises the enormous and also characteristic ability of microorganisms to develop and reproduce. Microbiological analysis uses exactly the same phenomenon that takes place after for instance a product has been infected. In connection with the sampling the organisms are allowed to come into contact with a growth medium, often in the form of an agar, a gel-forming material containing both humidity and nutrients. The sample may come in contact with the agar either by direct contact through a contact plate (in surface analyses) or by the transfer of the sample with a swab that has been in contact with the product or the surface to be analysed. The agar material is thereafter placed in an incubator at the correct temperature for the microorganisms in question. It is left in the incubator for a predetermined time in order to give the microorganisms sufficient time to reproduce. After incubation the agar plate is studied with the naked eye. The result is presented as the number of formed colonies, CFU (Colony Forming Unit), i.e. the small spots that can be observed on the agar surface.

1.7 Dispersion and Processes

The major purpose, associated with the various working techniques and operations within contamination control, is to keep the handled products free from materials or physical states that can be of danger for the user or the consumer of the product. When contaminants are found in a process step or in a process flow the question will always arise as to how the contaminants have entered into the process flow. Contaminants can come into contact with process flows either by being created within the process system or by being trans-

ported from the outer production environment (the production facility) into the inner production environment, where the process is taking place.

Dead particles can be formed within the internal part of a process system, for instance as a result of sealing materials degrading due to age and successively coming into contact with the product. In many industrial processes different types of axles go from the outer environment into the process environment. The axle is generally sealed at the interface, but particles can be released from the sealing material during axle rotation. Vibrations and other types of external interference with the process can be a danger to the product in the process by releasing different types of particles and allowing them to enter into the process. The list of critical parts can be very long.

One of the most frequent contamination risks occurs through dispersion of contaminants from the outside into the inner process environment. One way to reduce this type of risk is to design and construct the process in such a way that it becomes as enclosed as possible. In theory this means that nothing can either enter the process from the external environment or leave the process into the external environment. This type of solution is however very expensive and often reduces the flexibility of the operator. All processes cannot, however, be constructed and designed in such a way that the product or internal parts of the process equipment will not be in contact with the external environment. It is however possible to gain protection from external contaminants, for instance by using a buffer tank equipped with some type of safety device. Such a tank can be equipped with a filter that will ensure that air entering or leaving the tank will be freed from contaminants that might be harmful. A transfer to the process environment can take place from the outer surrounding environment, i.e. the air that is present in the production room, for example by the physical presence of humans and also by the influence of different media used in the process not having the correct cleanliness level.

In order to reduce the possibility of contaminants in the surrounding air entering the process or the product, filters can be installed at certain critical spots. Another way to reduce the

total exposure to contaminants is to work with the product in a cleaner environment surrounding the process. The production process can for instance be performed in a cleanroom or a clean zone. The interference by humans can, to a great extent, be minimised by the aid of a cleanroom. Working in a cleanroom means that the personnel are appropriately dressed (cleanroom clothing) and that the work is performed correctly in accordance with written instructions. As far as possible any direct contact with products and surfaces in contact with products must be avoided.

All types of media that are used in production, such as water, steam, pressurised gases and various cleaning solutions and disinfectants, must possess the correct cleanliness in order not to cause any problems.

Independent of how a contaminant will come into contact with a product or a critical surface, a general rule exists in connection with all forms of hygiene and clean handling. There must always be a driving force to avoid contaminants in the near proximity of the product. At the same time the eventual contact of contaminants with the production process as well as the product must be kept to an absolute minimum.

1.7.1 Hygienic Design

The hygienic design of a processor or a production facility is a powerful tool in order to reduce the possibility of negative influence on the product by contaminants. From a design perspective, the most important factor is to avoid cracks and pockets, especially on product surfaces. A crack is described as a quite narrow and relatively deep defect, whereas a pocket is characterised as having a small depth and a larger opening towards the outer environment, see Fig. 4.

It is also important to construct all parts in a process system so that the system can be easily drained. This will facilitate the possibility of product, rinsing water and also cleaning chemicals leaving the process system more effectively. So-called ›dead-legs‹ should always be avoided. A dead-leg is a dead-end pipe of a larger piping system that has no flow through it and is not easily and readily cleaned by traditional methods.

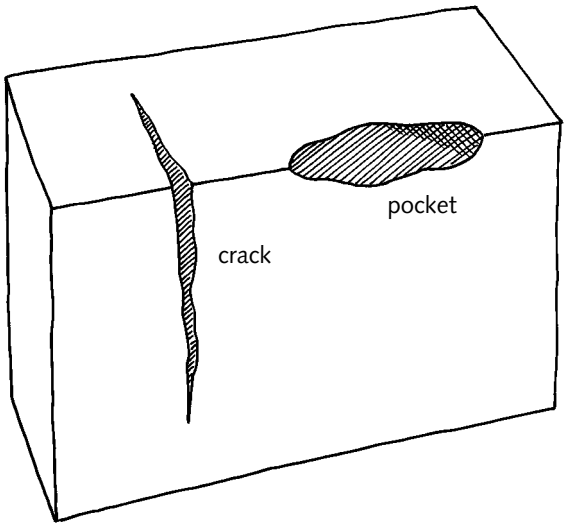

Figure 4. Hygienic design plays an important role within contamination control. This figure shows the difference between cracks and pockets.

1.8 Choice of Cleaning Technique

This book covers two important techniques utilised in order to eliminate or neutralise containants in a product or in a process flow, namely filtration and sterilisation (heat treatment).

Filtration has the advantage that a semi-permeable filter material allows the actual flow of gas or liquid to pass through at the same time as undesired contaminants will be collected in or at the filter.

A temperature dependent method, such as steam sterilisation, will influence microorganisms in such a way that they will not be able to develop and reproduce. The organisms will, after such a sterilisation process, still exist in the solution, even if in a dead state. When manufacuring pharmaceutical products especially intended for injection into the body, even small concentrations of dead microorganisms in products are not tolerated. This demand is due to the fact that some dead microorganisms will, upon degradation, release critical substances that might be harmful to the patient.

1.9 Conclusion

The overall purpose of contamination control is to control different critical contaminants that negatively influence products, processes and people. Contamination control must be looked upon as a holistic science, i.e., a science based on knowledge of the interrelation between people, processes, air, surfaces and products.

2 Separation

2.1 Introduction

The development of filtration, and also the development of different types of filtration products, has to a very great extent taken place in cooperation with industry, especially the pharmaceutical industry. The reason why this branch of industry has had such an impact on the development is particularly the result of the continuously increasing demands for cleaner products and production steps during manufacturing. New products and processes developed within the food and beverage industry as well as the packaging industry have given other branches of industry, especially those that are increasing in activity, the possibility to adopt the same cleanliness demands.

To choose, install, use and validate a filter system demands knowledge. Within the pharmaceutical industry most of the filtration knowledge at the end of the 80s and at the beginning of the 90s was obtained according to the principle of ›trial and error‹, i.e. continuous trials were made in order to obtain a functional solution to a problem. The knowledge was then inherited by co-workers within the company through experience and internal learning. Filter manufactures and filter-delivering companies have also been engaged as knowledge sources and co-partners. This cooperation has been very positive and profitable, which indicates and confirms that the development of both manufacturing processes and products is based on factors like knowledge, knowledge supply and adjustment of production processes.

2.2 Separation Techniques

Separation is the overall technical term, including for instance filtration, and in practice means that components in a mixture are separated from one another in different ways. The purpose of a separation process is either to isolate one or more of

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the components from the mixture or to obtain a cleaner fluid after separation.

Separation techniques consist of a huge area ranging from the removal of larger particles or structures (for instance algae and particles from seawater), to the isolation separation of single components in complex mixtures (for instance specific proteins from the circulatory system in man). The various separation techniques can be divided into:

- Absorption
- Adsorption
- Sedimentation
- Centrifugation
- Flocculation
- Filtration

In general separation processes are used to separate different components in a mixture from one another. Differences exist between the various techniques, for instance in the resulting accuracy, the process design, the possibility for industrial adoption and lastly in regard to safety.

2.2.1 Absorption Processes

To absorb a component from a mixture means that, in one way or another, the component to be isolated is forced into and kept in a more or less solid physical state, whereas the rest of the components in the mixture will exist in another physical state (liquid or gaseous). Figure 5A shows the principle of absorption.

2.2.1.1 Gas Absorption

A gas flow may be comprised of a mixture of different gases, a situation commonly observed within the process industry. The purpose of gas absorption is to try to separate the different gases from one another. It can for instance be that one of the gas components is of considerable value and therefore must be collected. Another reason for separation is that one gas component is harmful for the product, process or the personnel and therefore is not allowed within the gas flow. Gas adsorption can be achieved by letting the gas mixture come into contact with a liquid in which the gas component of interest is more soluble than the rest of the gases in the mixture. The gas of interest will be absorbed in a liquid state,

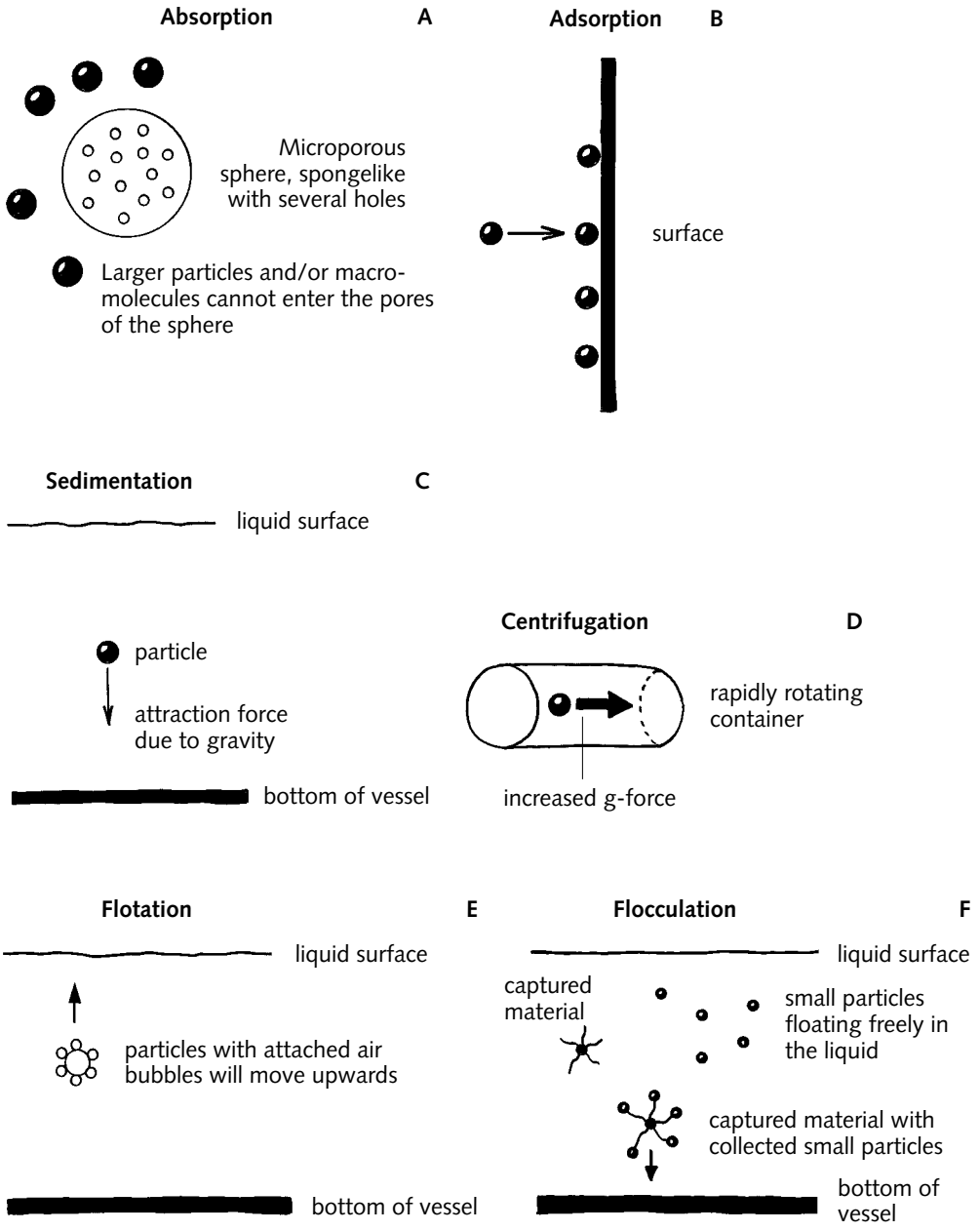

Figure 5. This figure illustrates the principles of absorption (A), adsorption (B), sedimentation (C), centrifugation (D), flotation (E) and flocculation (F).

whereas the rest of the gas components in the mixture will be kept in pure gas form and pass more or less freely through the separation system.

2.2.1.2 Liquid Absorption

In a similar way as for gas absorption, liquid absorption can be used to separate different components in a liquid phase from other components. An example is a water soluble protein solution that is very dilute. If this protein solution needs to be concentrated, absorption can be used. During the concentration (water separation) step of a dilute protein solution, a portion of dry particulate material comprised of spherical, non-water soluble particles manufactured from a microporous material, i.e. small spheres containing very fine pores (holes), is added. The micromolecules of the protein solution (comprised of water and salt) will have the ability to penetrate into the micropores of the spheres and will be kept in the spheres in an ›immobilised‹ state. The proteins in the solution are far too large to be able to penetrate into the pores. This form of absorption can be compared to a dry piece of cotton that is allowed to absorb a certain amount of water when placed in a beaker filled with water. The water will be drawn into the cotton.

By choosing an appropriate microporous material with sufficiently small cavities, the protein molecules will not be able to enter into the spheres due to their size and structure, whereas the water and the salt molecules will enter and be absorbed by the spheres. The residue protein solution outside the micropore spheres will be concentrated by the dewatering action of the spheres. Absorption can be used for separation on a small laboratory scale but is also used for large scale applications.

2.2.2 The Adsorption Process

Adsorption is most easily explained as the formation of a very thin molecular layer on the surface of a solid material. Adsorption of particles in this way can be explained by the forces, either physical or chemical in origin, that during the flow of the mixture over the adsorbing material will attract and thereby immobilise the particles by adsorption to the surface. The solid capturing material used for adsorption can, from a macroscopic point of view, be a small tube through which the

flow is passing or the pores in a porous filter material. Figure 5B shows the principle of adsorption.

2.2.3 The Sedimentation Process

In total this separation process includes sedimentation, centrifugation, flotation and flocculation, see Fig. 5C-F. Sedimentation occurs spontaneously in nature due to the attractive force of the earth that will affect particles which are suspended (mixed) in gases or liquids. The gravity force will cause the mixed and suspended solid material, if it has a large enough mass, to fall downwards and be collected on the bottom of seas and on land. Sedimentation is used within the process industry for gross cleaning of water, for instance drainage or waste water.

Centrifugation is a forced form of sedimentation that can be obtained by exposing a solution to an increased gravity force. This increased gravity force can be obtained by placing the mixture to be cleaned in a special container or in a complete apparatus that is circulated at very high speed. The centrifugal forces that are created through this procedure will cause the suspended solid material in the mixture to move faster out to the perimeter of the centrifugation vessel where it will settle and be collected. Centrifugation is used both for large-scale separations and for laboratory-scale separations. This separation can be performed either in a continuous or in a batch-wise manner.

Flotation is a technique that is totally the opposite to sedimentation. Flotation means that a particle suspended in a liquid that normally does not wet the particle will attract bubbles of gas that are naturally occurring in the liquid. The attracted gas bubbles can, if the particles in the liquid are small enough (i.e. have a small enough mass), affect the particles in such a way that they are lifted up to the surface of the liquid, like a ›balloon-effect‹. The particles are collected on the surface of the liquid in the form of a foam, which can be quite easily removed in a purely mechanical way. Particles that, on the other hand, are wetted by the liquid will not be affected by this technique since the air bubbles or gas bubbles in the liquid cannot be attracted to wet particles. These particles will therefore not rise towards the surface of the liquid. Instead, due to the sedimentation effect, they will either fall towards the

bottom of the vessel or, if small enough, stay in a suspended form in the liquid, neither moving upwards nor downwards, see Tab. 1.

Flotation can be controlled and influenced by the addition of different types of chemicals, so-called flotation aids. These flotation aids will have a major impact on the desired separation effect by rendering the particles not able to be wet by the solution in which they are suspended. In short these aids help to alter the hydrophobic and hydrophilic characteristics of the particles. With the aid of different additives it is possible to control the separation process, so that different suspended particles in a solution can be separated from one another. Flotation is most suitable for large-scale separations, for instance drainage or waste water.

Flocculation is another technique that can be used with great advantage when smaller particles are to be eliminated, for instance colloidal particles or similar. This technique is based on the use of different types of additives that have the ability to aggregate particles, i.e., to collect smaller particles together making them form larger aggregates. The additives traditionally comprise molecules or particles equipped with ›capturing arms‹ to which the smaller particles can be attached in order to form larger aggregates.

The aggregates formed can thereafter be allowed to sediment by themselves or be more easily captured in a filter. Occasionally these aggregates can disintegrate when passing through a filter. Upon disintegration, the small particles may pass through the filter and, when on the other and clean side of the filter, have the ability to form new aggregates. However, this type of phenomenon does not necessarily mean that the filter is not working according to specification.

2.2.4 The Filtration Process

Filtration is a technique based on the fact that particles will be trapped and held in the filter by means of pure mechanical or adsorptive forces when a solution or gas is forced through such a porous and semi-permeable material, normally called the filter material or the filter medium. Filtration is suitable for both large-scale and laboratory-scale separations and can be used for continuous and/or batch-wise processes.

Until approximately 15–20 years ago filtration was con-

sidered a relatively crude separation method compared to the other separation methods mentioned. Today filtration equipment and filter media are available to take care of nearly all types of contaminants and process environments.

2.3 Conclusion

Separation comprises different types of techniques that are used in order to separate different components in a mixture from one another. Different types of separation techniques exist, for instance absorption, adsorption, sedimentation (divided into sedimentation, centrifugation, flotation and flocculation) and filtration. Differences exist between these different processes, especially in regard to accuracy, performance characteristics, the ability to adapt the method to industrial use and finally also safety aspects.

3 Filtration Technology

3.1 Introduction

Filtration technology is the overall technical area in which basic definitions of filters and their behavior are discussed. It also describes filter function and the characterization of different types of filters. Different areas of application, for instance what actually takes place in a filter when it is exposed to a process flow, the difference when filtering liquids and gases together with the practical advantages and disadvantages, from a technical perspective, of using different filter types, are also included in filtration technology.

3.2 Filtration

The process of filtration has the objective of separating different components in a mixture from one another. Depending on physical as well as chemical characteristics, filtration can be explained in different ways. Figure 6 shows some of the most commonly used filtration techniques. The differences between them are chiefly based on the size and shape of the material to be separated, i.e. the physical and chemical characteristics of the contaminants. The figure also shows that components in a mixture can be of solid material (larger or smaller particles), soluble high molecular weight molecules (macromolecules) or soluble smaller molecules (micromolecules).

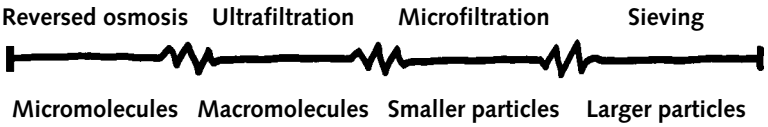

Figure 6. Schematic illustration of the various separation techniques incorporated within filtration. The borderlines between most of these techniques are not totally sharp, and some overlap may be observed.

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Filtration can be divided into the following different categories:

- Straining or crude filtration
- Microfiltration (MF)
- Ultrafiltration (UF)
- Reversed osmosis (RO)

These methods are, from a microscopic point of view, physically quite similar, since they all are pressurised membrane processes. This means that a flow of fluid is forced through a partly permeable filter material (a semi-permeable filter material or membrane). The permeability of the material depends on the possibility for a liquid or a gas to pass through the pores, the voided part, that exist in the filter medium. The size and shape of the pores in different filter materials will differ depending on which technique is used.

Straining, which is sometimes called crude-filtration, in practice means that solid material, particles such as sand, will be stopped by the strainer whereas microorganisms, other smaller particles and soluble molecules (macro- as well as micromolecules) will pass more or less unaffected through the strainer.

Microfiltration is the technique whereby microorganisms and other smaller particles can be stopped and retained in the filter material whereas soluble macromolecules, for instance antibodies and other protein materials, and micromolecules, will pass relatively freely through the microfilter material.

Using ultrafiltration high molecular weight macromolecules will be retained by the filter material whereas smaller molecules, for instance a water-based buffer system, salt solution etc., will be allowed to pass freely. Ultrafiltration plays an important role within the medical area. The technique is commonly called dialysis when used for patient treatment and also in different systems for biotechnological use.

Reversed osmosis is a technique that has the ability to separate micromolecules of equal size. The technique is mostly used for desalination of seawater. Reversed osmosis is actually not a filtration technique in the same way as the others mentioned above because it is based on other technical principles.

3.3 Dead-end and Cross-flow Filtration

The previously mentioned separation methods: sedimentation, flotation, centrifugation and flocculation, can in many cases be regarded as quite crude methods and are therefore often quite uninteresting to use. This is, however, not always correct. In many cases, especially when larger flow-rates and larger volumes are to be treated, these more traditional methods can, from both practical and economical points of view, be quite interesting to use. This is especially true when the amount of solid material to be removed is so high that a single microfiltration step or a single filtration system cannot deal with the contaminants in the process flow.

In order to cope with larger amounts of contaminants (i.e. particles) a pre-treatment step is needed to pre-clean the flow before the final microfiltration step can be performed. Contamination levels for these dead-end systems are, for economical reasons, around 1% or lower, normally 0.01%. Dead-end filtration is normally called ›one in and one out‹; a description arising from the fact that only one flow can pass through the filter after it has been cleaned, see Figure 7.

Larger contamination levels (i.e. particles) can be treated in so-called cross-flow systems (sometimes referred to as tangential flow), where the contaminated part of the process flow continuously recycles tangentially past the active filter material. This continuous recycling flow will wash clean the dirty side of the filter. Cross-flow filtration is therefore also referred to as ›one in and two out‹.

3.4 Different Types of Contaminants

When choosing a filter, and especially when a filter system is to be dimensioned, it is of vital importance to have as much knowledge as possible about the different types of contaminants, especially particles, that are to be eliminated. It is also important to have an overall understanding of the size distribution of these. Large differences can be observed for instance depending on whether it is water, steam, air or a finished product of more or less complex nature that is to be analysed.

Sample 1 in Fig. 8 schematically shows the size and size distribution of particles in an untreated plain drinking water. The sample has been taken directly from the tap and particles

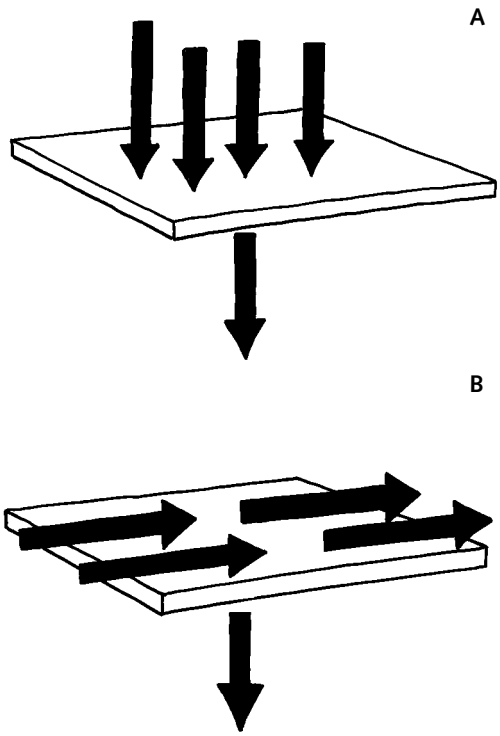

Figure 7. Filtration can be performed in two distinct ways. (A) shows the principle of dead-end filtration, sometimes referred to as one in-one out filtration. (B) shows the principle of cross-flow filtration, sometimes also called tangential flow or simply one in-two out filtration.

of different sizes are counted. The result from the particle counting is then plotted in order to give a size profile of the particle content.

Sample 2 schematically shows the differences in size and size distribution of the same water stored after filtration. The water, after filtration, is stored in a tank for a certain period of time before it is analysed again. The tank has no protection against the outside environment, giving microorganisms present in the surrounding atmosphere the possibility to come into contact with the water through the tank ventilation.

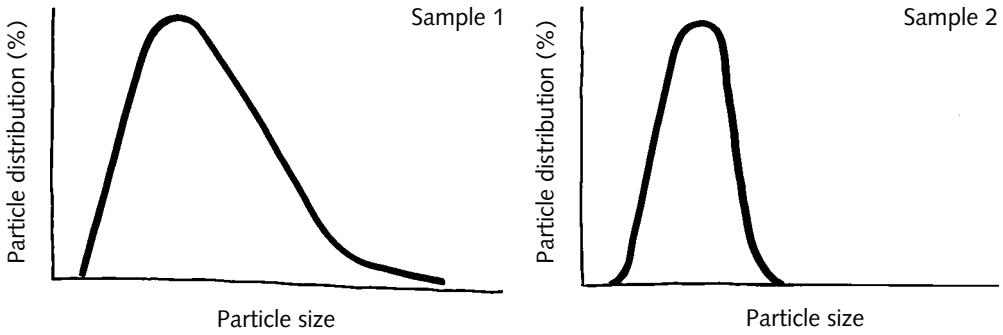

Figure 8. The figure shows a typical example of particle size distribution for particles with different origin. Sample 1 mainly contains particles of non-biological origin, whereas sample 2 mainly contains microorganisms.

Despite the fact that it is the same water in both systems studied, differences in particles can be observed in the two samples, both in regard to their number and their size distribution. Sample 1 contains mostly dead particles, particles of a non-organic nature whereas sample 2 contains mostly living particles, for instance algae, molds and other microorganisms that have come into contact with the water due to the poor protection of the ventilation of the tank. These organisms have developed and reproduced in the tank during storage and become the largest contamination in the stored water. Figure 8 shows that sample 1 has a much broader distribution of particles than sample 2. In practice and also from an economical point of view this can have a major influence on the result of the filtration.

These two schematic drawings show different types of particles with totally different particle size distributions, factors that are of importance when choosing for instance the type of filter to be used and the size of the filter system. This system is especially important when using sterile filtration or any other filter system where microorganisms are to be eliminated. The examples are however schematic but highlight the differences in particle size distribution between particles of non-organic and organic nature. They also represent a good example of the importance of having an overall more holistic

view of a system when clean products are to be manufactured.

3.5 Conclusion

The area of filtration technology incorporates the basic definition of filtration, filters and different types of contaminants.

4 Microfiltration

4.1 Introduction

Microfiltration can be divided into smaller segments depending on what type of cleanliness demands are put on the product and (or) the process. The different segments used within microfiltration (Figure 9) are:

- Coarse filtration
- Clarification
- Polishing
- Microbiological reduction
- Sterile filtration

4.2 Coarse Filtration

Coarse filtration is a very crude type of separation within the area of microfiltration. When discussing particle sizes this type of filtration is situated directly below, and sometimes even incorporated within, the area that has previously been described as straining. Coarse filtration is often used as a single

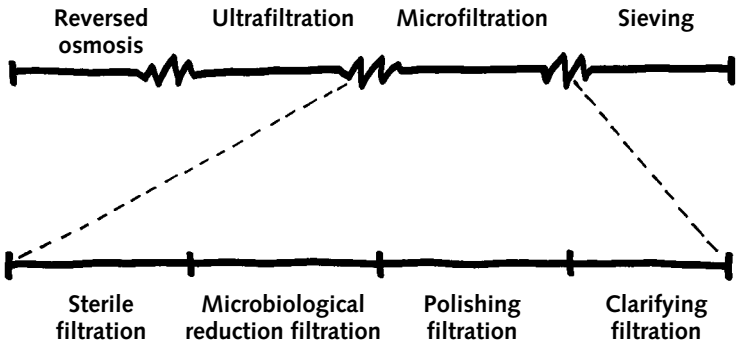

Figure 9. Schematic representation of the various subparts that together comprise the technical area of microfiltration. As shown in Fig. 6, some of these areas overlap. Only one sharp borderline exists, namely sterile filtration (0.2 or 0.22 μm respectively).

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separation step or as a first step in a larger process system in order to reduce the total amount of particles to a more appropriate level. Separation by coarse filtration eliminates larger particles and solid structures, for instance residual organic material, leaves from trees and also larger algae from cooling water and water from different sources.

Coarse filtration is not often considered as a critical filtration step because in most cases single particles of larger sizes are allowed to pass through the filter. In industrial applications different depth type filters, comprising wound filter cartridges or sheet filters constructed and manufactured from different fibrous materials, are used.

Typically, the particle size for coarse filtration lies in the region from approximately 90 μm down to 40–50 μm.

4.3 Clarification Filtration

Clarification or clarifying filtration is used for particle removal in the size range of approximately 40 μm and higher. 40 μm is called the visibility border, i.e. the size level at which the naked human eye can identify a product as clear or free of visible particles. This particle size (40 μm) should be compared to the size of a normal hair that has a diameter of approximately 70 to100 μm.

When filtering a product in order to clarify it, a filter with even lower rating is used, normally between 40 and 20 μm. Using such filters minimises the risk of letting an appreciable number of particles of size 40 μm and smaller pass the filter. Such smaller particles can be visible to the naked eye as a haze in the solution if the particle concentration is large enough.

Clarification is mostly used when cleaning for instance drinking water, different types of sodas and also sugar solutions.

4.4 Polishing Filtration

Polishing or polishing filtration is often used in processes where bio-catalysts (different types of microorganisms or other biological/biochemical material) are used in order to produce a product. Organisms are frequently used in different types of fermentation processes, for example during the manufacture of beer, cider and wine, and also increasingly during

the manufacture of different active pharmaceutical substances. The microorganism is not only used in the production of the desired product, it also lives, develops and reproduces in the solution during the fermentation process.

Interactions between the microorganisms and different components in a fermentation solution (for example product and different types of nutrients) can in some cases result in the formation of very fine particles, so-called colloidal particles. These colloidal particles are impossible to identify with the naked eye. They are often observed at high concentrations as a thin haze or shade in the solution.

Sometimes this type of contaminant is so small that it is debatable whether it is a particle or not. Sometimes these small particles have the ability to form larger aggregates. Such aggregates, i.e. the aggregated form of colloidal particles, can be observed by the naked eye to a certain extent. This means that the aggregate formed is larger than 40 µm. Products containing non-aggregated colloidal particles cannot be clarified using clarifying filters. Instead filters with much lower rating must be chosen. It is sometimes necessary to go very far down in filter ratings, even down to the level of sterile filtration and lower. The filtration rating needed to get a clean product is, as is seen from the example above, totally dependent on the origin of the contaminants in the fluid. The result of polishing filtration is therefore, to a great extent, based on what type of contaminants or particles are to be eliminated, the particle size distribution and the possibility of the particles forming more or less stable aggregates. This type of system might also be critical even if larger aggregates of smaller particles are formed. Some of these aggregates tend to be quite unstable and easily disintegrate during passage through a filter. After filtration the small particles sometimes have the possibility to re-aggregate.

4.5 Microbiological Reduction Filtration and Sterile Filtration

When looking at filtration as an overall technique there is only one clearly defined and accepted level of rating, sterile filtration. According to the FDA (Food and Drug Administration, USA) and the United States Pharmacopoeia (USP), sterile filtration is defined as a nominal rating of 0.2 µm and 0.22 µm

respectively. For a full and comprehensive definition please see page 67.

Sterile filtration is most commonly used within the pharmaceutical industry. This level of cleanliness has also been adopted by other industries, for instance the food and beverage industry.

Several branches of the food and beverage industry indicate that their products are sterile filtered before being filled into vials. From the food manufacturers point of view it is a correct statement, but in many cases a totally wrong statement in regard to the accepted definition of a sterile filter. It even turns out that the food product in many cases cannot be filtered through a 0.2/0.22 µm filter without being negatively affected, for instance changes in taste or color occur. When food and beverage manufacturers describe their products as sterile filtered, they often mean that the microorganisms destroying the product have been eliminated, but not by using filters as far down in the filter ratings as sterile filtration. A much better explanation of the system described above is filtration down to an industrial cleanliness, a form of microbiological reduction, which is totally separate from the cleanliness obtained by sterile filtration. Dangerous microorganisms (i.e. dangerous to the product) are eliminated with a microbiological reduction filtration, with ratings typically in the region 0.30 µm, 0.45 µm, 0.65 µm, 0.80 µm and, to a certain extent, even up to 1.0 µm, depending on the sensitivity of the products in relation to the microbiological contaminant present.

4.6 Conclusion

Microfiltration is a filtration technique that will retain microorganisms and smaller particles in a microfilter media, whereas soluble micromolecules, for instance antibodies and other proteins, will pass through the filter. Particle size distribution within microfiltration will vary depending on the area of use from 90 µm down to approximately 0.01 µm.

5 Filter Mechanisms

5.1 Introduction

Filtration and various other separation techniques are based on statistical grounds that in turn are influenced by several different factors. The outcome of a filtration process is mainly influenced by factors like:

- The number of pores per unit square area of the filter (the so-called porosity)
- The geometrical design of the pores
- The structural construction of the pores through the total filter media
- The total thickness of the filter media that the fluid has to pass

Other factors exists that might also play a vital role, for instance if the fluid to be filtered is a gas or a liquid, if the liquid to be filtered is hydrophilic (water attracting) or hydrophobic (water repelling) together with the type of material of which the filter media is constructed.

The physical construction of the filter will also affect the outcome of the filtration, i.e., if the filter is categorised as a depth type filter or a membrane type filter. The function of a depth-type filter is primarily based on the actual physical depth of the filter. The depth of such a filter is not really relevant, rather, a large depth of filter means in practice that a larger filter surface area will be exposed to the fluid passing through the filter, to which particles and other contaminants can be adsorbed. A membrane filter also captures particles in different ways. This type of filter looks very thin but also has a certain depth and works to a certain extent like a depth filter. A membrane filter does not, however, have the same physical depth as a depth filter, making it not as potent in regard to how many particles it can capture, the so-called dirt capacity.

Both types of filters, depth filters and membrane filters, are

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constructed of filter media that have randomised, i.e., statistically distributed, pores. In practice this means that the pores through the filter media are not fully straight and perfectly circular. However, filter media exist where the pores are formed by bombarding the filter media with ›particles‹ during production of the filter. These filter media have perfectly circular and straight pores. The porosity of the filters is, due to practical reasons, often very low, making these types of filter generally not suitable for use when handling large process flows.

Independent of which type of filter (depth filter or membrane filter) is used, filter function and the outcome of the filtration are due to two major mechanisms called mechanical and adsorptive retention.

5.2 Mechanical Retention

Mechanical retention is in many cases referred to as sieving. This separation effect is based on the fact that the particles that cannot pass through the pores of the filter will be retained and kept in an immobilised state in the filter, Fig. 10. The retained particles will be held either inside the filter media

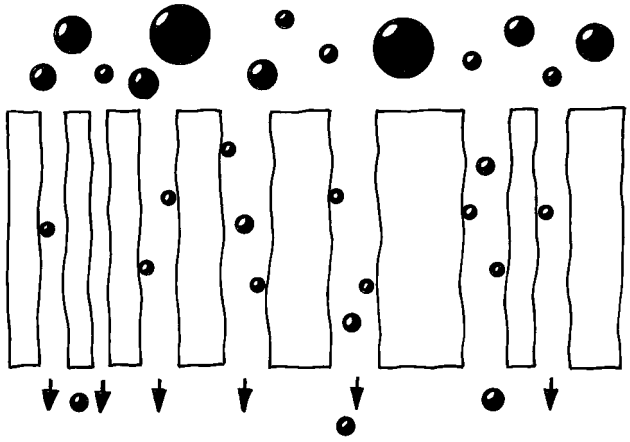

Figure 10. This figure shows the mechanisms of mechanical retention which is sometimes called sieving.

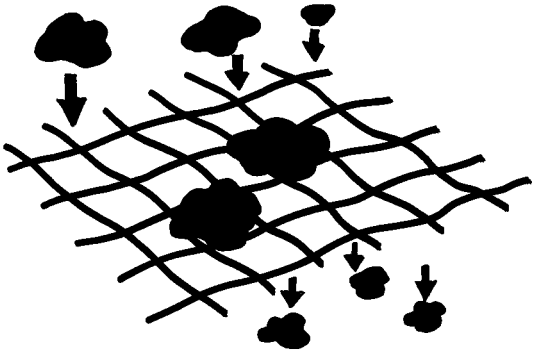

Figure 11. This figure illustrates the theoretical aspects of surface filters.

or on the surface of the filter media, depending on the size of the particles in relation to the size and structure of the pores of the filter. Mechanical retention will take place when filtering liquids as well as gases.

5.2.1 Surface Filters

The term surface filter is commonly used. In theory this type of filter will collect particles at its surface only, Fig. 11. This means that particles that are sufficiently large will be collected on the surface, whereas particles that are smaller pass through the pores of the filter in an unhindered way. Particle material collected on the surface can be removed, i.e., the surface filter may be cleaned quite easily, by back-flushing of the surface filter system. After this regeneration the filter will become totally clean and fully restored to its original state.

The surface filter is a theoretical filter, in practice it does not work according to the description above since most filters not only collect particles on their surface but also in the interior of the filter material. One exception however exists, especially if the size distribution of the particles to be eliminated is quite homogeneous. The randomised pore size distribution in a filter medium, together with the total depth of the filter medium, will play a major role since particles often have the ability not only to be retained on the surface of the filter but also inside most filter materials.

Particles that are collected on the filter surface can easily be removed by, for instance, back flushing, whereas particles that are collected inside the depth of a filter are difficult, if not totally impossible, to remove without the use of strong chemicals. Providing that the filter material is compatible and thereby can cope with the different cleaning chemicals that are used during such a regeneration, the filter can, in some cases, be cleaned. Otherwise, not only will the contaminants be dissolved by the chemicals, but also the filter medium itself. As a general rule the filter must be fully compatible with all cleaning solutions to be used during regeneration.

Surface filtration, or more accurately the theory of surface filtration, is mostly used when filtering a large flow of liquid in fully automated filtration systems. One example is the filtration of re-circulating cooling water in nuclear power plants.

5.2.2 Pre-coat Filters

The effect of surface filtration can be obtained by the action of different filtration aids. Such filtration aids will function in such a way that smaller particles will be totally stopped when trying to enter into and hence block the filter medium. Filtration with filter aids is sometimes called pre-coat filtration. A pre-coat filter is constructed of a support filter medium, a so-called septum, that in itself does not have enough retaining capacity, but if it is coated with a suspended material comprised of larger particles it can be made useful. The suspended pre-coat material has a particle size distribution that is chosen in such a manner that the pre-coat material will form a filter cake on the surface of the septum. The filter cake will, when passed by the process flow, capture particles within the cake without blocking the septum during filtration, Fig. 12.

In practice, the porous filter cake formed on the septum will act as a very fine filter medium, as compared to the septum, during the filtration process. In many cases the filtration aid used has a double function. Firstly, it can be the collector of particle contaminants and secondly, it can help to capture dissolved smaller components, such as ions, for instance if the pre-coat material also has an incorporated ion exchange property.

When the pressure drop over such a pre-coat filter system has increased to a certain predetermined level, a situation that

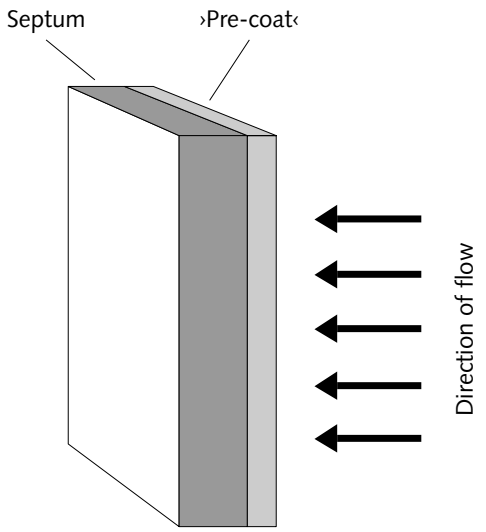

Figure 12. The figure illustrates the principle of a pre-coat filter, one of the few filters that nearly acts as a surface filter.

occurs when the number of collected particles in the pre-coat layer is large enough, the system can be regenerated by back-blowing or back-flushing. The suspensions formed during such a back-blow or back-flush not only contain the pre-coat material but also the collected particles and other contaminants. The suspension is then allowed to leave the filter system through the drain. The pre-coat filter can then be given a new suspension of filter-aid, forming a brand new pre-coat layer on the septum. The filter is then ready for a new filtration cycle.

In theory, this type of filter can result in a system where the filter septum will have an infinite life. This is, however, not the case in practice. Particles from the pre-coat material together with particles in the fluid to be cleaned can be released from the bed and enter into the septum, thereby slowly blocking the total system. This is why the septum normally needs to be changed after some time in service. The actual blocking of the septum is in most cases a very slow process and if the concentration of particles in the process flow is not constant over time it is very hard to calculate change intervals.

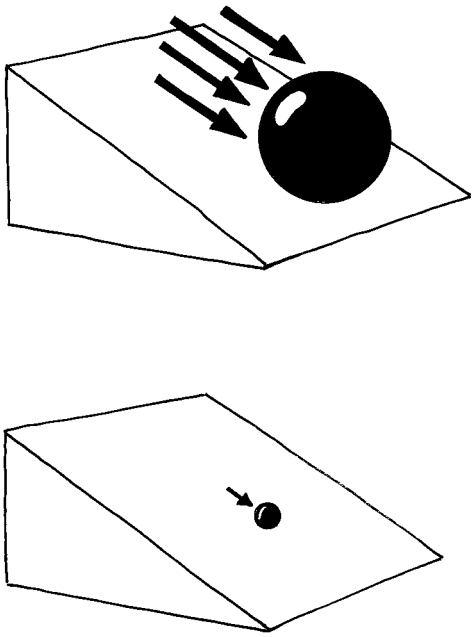

Figure 13. The impact of flow forces on particles of different sizes. The smaller the particle the harder it is to release a particle that has been adsorbed to a surface.

5.3 Adsorptive Retention

Adsorption of particles to a filter material can be explained by physical and chemical forces within a flow capturing and immobilising particles as a thin layer on the surface of a solid filter material. Adsorptive forces include ionic interactions, hydrophobic interactions and hydrogen bonding.

Each of the above-mentioned forces are very weak compared to the stronger ones, for example covalent bonding, observed in some chemically more stable compounds. The adsorptive forces, however, have the capacity to interact in order to create a very strong total binding force. Very high forces associated with passing flow are needed in the pores of a filter in order to release a particle that has previously been adsorbed, Fig. 13. Larger particles are held to a surface with much less strong forces than smaller particles are. This is because the smaller particle has a larger contact area with the surface to which it has been adsorbed in relation to its size.

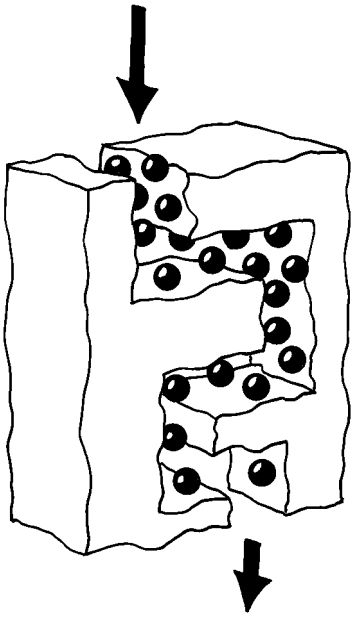

Figure 14. Smaller particles, even those small enough to pass freely through the filter pores, have a great tendency to be adsorbed to the surface of the filter pores. Adsorption can be by inertial impaction and (or) diffusion impaction.

This is why a passing flow of fluids has difficulty in releasing a previously adsorbed particle, especially if the particle in question is extremely small. The contact phase when the particle comes into close contact with the filter medium can occur in two different ways:

- Retention due to inertial impaction
- Retention due to diffusion interception

As discussed earlier, the pores in a filter medium have an uneven path when running through the filter medium and the central line of the flow will constantly change direction during passage through the filter, Fig. 14.

5.3.1 Retention Due to Inertial Impacting

Adsorptive retention can occur by inertial impaction or diffusion interception.

Adsorption due to inertial impacting means that a particle with a large enough inertia (the mass of the particle multiplied by its velocity) can easily be adsorbed to the pore wall of the filter medium. If the mass of the particle and its velocity are large enough, the particle will not be able to follow the central line of the flow, especially when the flow deviates during passage through the filter pores. Such particles will instead continue in a tangential direction out from the central line of the flow and thereby come in close contact with the pore wall where they will be adsorbed

If the adsorptive forces between the particle and the wall of the pore are strong enough the particle will be immobilised and retained bound to the pore wall, Fig. 15.

Adsorption due to inertial impaction will occur when filtering both liquids and gases.

5.3.2 Retention Due to Diffusion Interception

Diffusion is a physical effect that will affect all types of molecules. In practice diffusion is hard to observe in liquids and only visible when studying gases. Diffusion interception, i.e., the removal of particles by diffusion, is based on the fact that gas molecules surrounding particles in a flow will constantly collide with particles suspended in the gas flow due to their

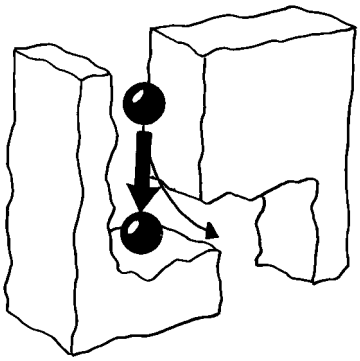

Figure 15. This figure shows the mechanism of adsorption of particles due to inertial impaction.

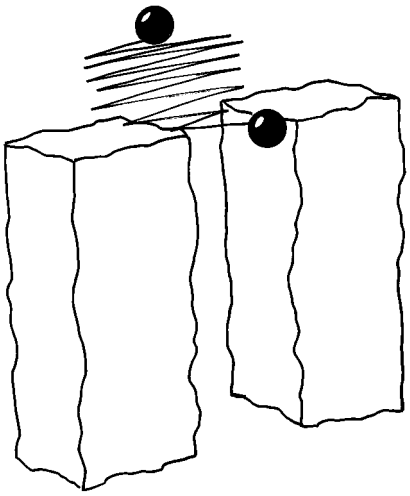

Figure 16. This figure shows the mechanism of adsorption of particles due to diffusion impaction.

irregular movement. If these particles are small enough (i.e., have a small enough mass) this constant bombardment of the solid particles with gas molecules will force the particles to move forwards and backwards in an irregular movement, the so-called Brownian Motion.

From a practical point of view the constant bombardment of particles with gas molecules will greatly affect the observed size of the particle in relation to the pore size. The particle size will, due to the Brownian motion, be observed as having a size as large as the amplitude of the oscillation (Figure 16).

The observed particle size will thus be much larger than the physical size, making particles suspended in gas come more easily into contact with the wall of the filter pores, where they can be adsorbed and retained.

5.3.3 Depth-type Filters

The effectiveness of a depth-type filter is dependent on the total thickness of the filter medium together with the adsorptive effect that the filter area will expose. Depth-type filters can have a depth from some few centimeters up to several meters. From a practical point of view, the longer the

distance that a gas or a liquid must pass, the better and more effective results will be obtained through the filter.

5.4 Conclusion

The function of a microfilter is primarily based on mechanical and adsorptive retention. Mechanical retention, in practice, means that particles that cannot physically pass a filter will be retained either inside the filter medium or on its surface. Adsorptive retention is obtained with the aid of different forces acting upon particles coming close enough to a solid surface, the solid pore wall in a filter.

6 Different Types of Microfiltration Filters

6.1 Introduction

Filter media used today and also to be used in the future are more and more tailor-made for specific filtration purposes.

Microfiltration filters can, in regard to design as well as function, be divided into two major categories, depth-type filters and membrane-type filters.

One of the first generation of industrially utilised filters, the cotton filter, was, in practice, a depth-type filter in the pure sense. The outcome from such a filter at that time corresponded to a great extent to the demands and the desires that were currently stated then. The purpose of such a filtration system when filtering liquids and (or) gases was to render these fluids free from mostly visible particles. Increasing cleanliness together with process demands have resulted in the development of new filter media that are much more effective in their retention capacity, mechanically stronger and thereby much more secure and safe during use. The development of the membrane filter and also the more recent development of depth-type filters with fixed pore structure have today led to many filters that can be exposed to higher pressure drops, variations in total pressure and also have a longer service time.

6.2 The Depth Filter

The function of a depth-type filter is mainly based on the depth of the filter medium together with the overall adsorptive effect exposed by the surface of the filter material, Figure 17. A depth filter has a certain totally visible thickness ranging from a few centimeters up to several meters and becomes, from a practical point of view, more effective the larger the depth of the filter. This means that the longer distance a liquid or gas has to travel when passing a filter in order to be cleaned, the better and more effective will the

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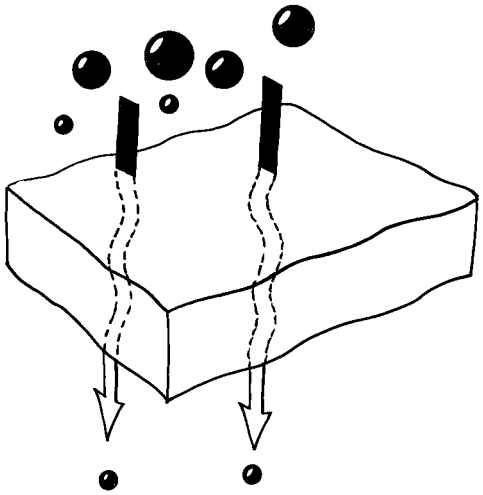

Figure 17. A depth type filter mainly utilises the mechanism of adsorption in order to capture particles.

results obtained be. Sand filters and cigarette filters are some examples of depth-type filters. Wound, compressed and even glued cartridge filters made of fibrous materials together with sheets for filter presses are other examples.

6.3 The Membrane Filter

In a membrane type filter the actual filtration takes place in a filter medium looking like a very thin piece of paper. Membrane filters can be designed, constructed and manufactured in different ways. Such filters can, for instance, be made of fibrous materials or cast as very thin membranes in a polymerisation process. Fiber-based materials are often called prefilters, and are commonly used in coarser filtration applications whereas polymer filters, often called final filters, are used for much finer and high precision filtrations, for example applications based on quantitative elimination of microorganisms from a fluid. The major difference between traditional depth-type filters and membrane filters is that the membrane filter has a much more rigid structure and total filter design, thereby rendering higher strength and durability. The

strength of a membrane filter is due to the stability of the pore structure in the filter medium.

From a practical point of view this means that a membrane filter will have the ability to retain particles collected from a fluid, and will not release these particles as long as the filter is used according to the specification of the manufacturer. Filter materials with stable pore structure will also be rigid and, if used according to specifications, not release parts of the filter medium into the clean process flow. This also means that a membrane filter does not normally show any signs of channeling (see below), i.e. enlargement of pores, as long as the recommendations of the filter manufacturer in regard to pressure, differential pressure etc. are followed.

6.4 Depth Filters with Fixed and Unfixed Pore Structure

New developments within fiber technology together with new manufacturing methods have been utilised for the development of a totally new generation of depth-type filters. Today, two distinct and totally different types of depth filters exist, the new generation that has a distinctly fixed pore structure and the older and more traditional depth filter based on quite loosely bound pore structure (often referred to as an unfixed filter).

The first generation of depth filters belongs to the group of depth filters with loosely bound pore structure. This type of filter generally suffers from severe drawbacks such as:

- Particle release
- Fiber release
- Channeling

6.4.1 Particle Release

As mentioned earlier, some particles are collected on the surface of a filter and others are collected in the interior part of the filter medium. When particles are collected on and in a filter a resistance to through flow will be built up, resulting in the creation of a pressure drop over the filter. This pressure drop is a direct response to the blockage of the pores in the filter by particles and will decrease the possibility of a flow passing the filter. The generation of a pressure drop over a filter will in turn have an impact on the filter medium, causing the filter to be exposed to quite high forces. If the filter

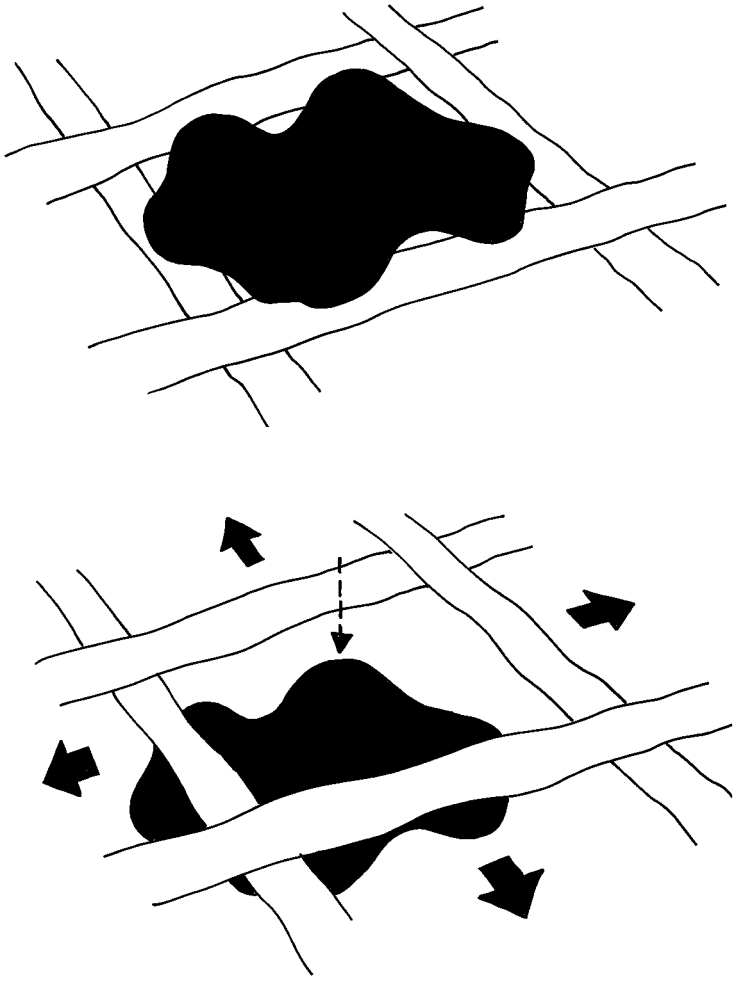

Figure 18. Particle release. Particles captured in loosely bonded filter materials have a great tendency to force their way through the filter.

medium is not firm and rigid enough, for instance if the fibers building up the filter medium are not sufficiently strongly attached to one another, the fluid flow through the filter will force these fibers to move in order to increase the flow rate through the medium. When the fibers in such a filter medium move there is a risk that previously collected particles will be released and allowed to move further down into the filter

medium. The phenomenon will continue and finally give the particles a chance to leave the system on the downstream side of the filter, Fig. 18.

Particle release is a quite commonly observed situation especially when using spun or wound or even glued filter cartridges. Unfortunately, the problem with particle release is not recognised by the filter user since the total service life of the filter is not limited. Filters demonstrating particle release are normally in service year after year. The differential pressure over such a filter can in the worst cases be constant and in some cases also decreasing, despite the fact that the filter will be in active service for a very long time.

6.4.2 Fiber Release

Fiber release is the second drawback of a loosely bound filter and occurs because fibers from the filter medium, since they are not bound together with strong forces, will loosen and be released on the downstream side of the filter, Fig. 19. This situation may be observed in connection with strong or sudden changes in the pressure and temperature of the filter.

6.4.3 Channeling

Channeling is often a final result of both particle release and fiber release. The flow through the filter is continuously trying to pass in the easiest way. This means that the flow during fiber release will erode the filter material, resulting in the

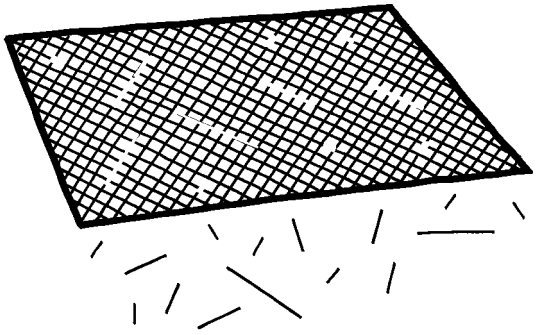

Figure 19. Fiber release. The fibers building up a loosely bonded filter material have a tendency to leave the filter and contaminate the fluid passing through the filter.

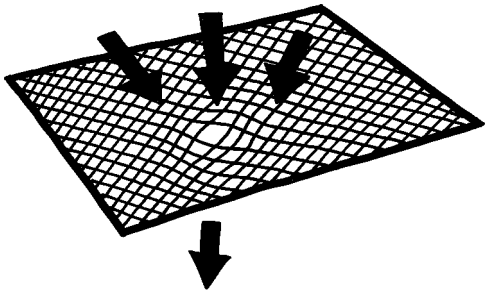

Figure 20. Channeling. The phenomenon of channeling ocurs when, for example, a pore of a loosely bonded filter material is enlarged. This situation can also arise when the filter material is not sealed well enough to the filter holder.

formation of larger pores (or channels) in the filter medium, either by just moving fibers to the side or by releasing fibers from the filter medium, Fig. 20. Channeling is also very commonly observed in connection with faulty sealing of the filter to the filter housing.

Erosion of this kind in a filter will result in increased size of the channels. This in turn will increase the possibility of erosion and further amplify the fiber release from the filter medium. Channeling is not only a problem of fiber release. Another part of this complex problem is that the majority of the total flow through the filter will choose the larger channel rather than the smaller pores for its passage. After some time, more and more of the process flows will choose the larger channels, resulting in a filtered fluid that has not been filtered to the desired level.

A short term solution for the problem with particle release and fiber release, especially for new filters, is to flush the filter prior to use. If the process flow is water, the water used for flushing can go directly into the drain. However, if the filter is to be flushed with a fluid other than water, then this solution must be returned and re-circulated back to the filter in order not to contaminate system parts further down the process.

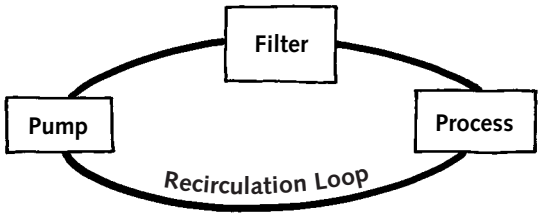

Figure 21. This figure shows a recirculating process system, a re-circulation loop.

The re-circulation is continued until the flow of liquid after the filter is clean enough. By using this type of solution the filter will in fact clean itself, Fig. 21. This type of solution could possibly be used in traditional chemical industry when the demand for cleanliness is not too high. Other industries cannot use this type of solution.

6.5 Development Due to Increased Demands

Increased demands together with development in especially fiber and polymer technology have led to a further development of the depth-type filter. Depth filters with loosely bound pore structure are however still used extensively, especially in processes where the cleanliness demands are not too high and where maintenance in connection with control and exchange of filters is performed in an excellent manner. In many cases these type of loosely bound filters are used as pre-filters, i.e., they are followed by other and more rigid filters, for instance a filter with a more fixed pore structure. Such filters with fixed pore structure will guarantee that eventually released particles and (or) fibers are collected.

6.6 Conclusion

Microfiltration filters are traditionally divided into two major groups in regard to construction as well as function: depth-type filters and membrane-type filters. The function of a depth filter is mainly dependent on the total depth of the filter medium and on the adsorptive effect exposed by the surface of the filter medium. The greater the depth (i.e., the path

length of the flow) the better the filtration effect. In membrane-type filters the filtration take place in thin paper-like layers constructed of a strongly fixed fiber and (or) polymer material.

7 Filter Rating

7.1 Introduction

The function of a filter is often expressed by the filter manufacturer as the rating or the pore size of the filter. The rating of a filter is however not fully standardised, which means that filter manufacturers and suppliers do not always follow the same theories and perform the same tests. This is a very unfortunate situation since it is not always easy or even possible to compare the ratings of one type of filter from one manufacturer with a filter from another manufacturer. In some cases it is not even possible to make comparisons between two different types of filter even if produced by the same manufacturer.

The rating of a filter should, in theory as well as in practice, be a description of the removal capacity of a filter expressed in such a way that it can be an aid to the filter user in order to find a suitable filter for an application. Let us take a practical example. A 1 µm depth filter with loosely bound pore structure, previously exemplified as a wound filter material, has a totally different removal capacity when compared to a membrane filter or even a depth filter with a fixed pore structure. This is even true despite the fact that the two filters, according to their respective manufacturers, are 1 µm in rating. This example shows that it is necessary not only to thoroughly understand the definition of the different filter ratings and how the various tests are performed, but also to have knowledge of what type of test contaminants are used and under what circumstances the various tests have been performed.

7.2 Contaminants

In order to investigate and determine the function of a microfilter different types of test particles are used. A generally used test contaminant is comprised of spherical particles, for instance glass beads or latex spheres. Spherical particles are

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mostly used to test filters for particulate rating in filter systems where microorganisms are not considered to be a problem.

The test contaminant ACFTD (Air Cleaner Fine Test Dust), is comprised of a very special dust from a desert in Arizona and has a much broader size distribution and also contains more unevenly formed particles. Spherical particles are not often observed in industrial systems and are therefore in many cases not considered as a relevant test contaminant. This is one of the reasons why many manufacturers today tend to change to AC Fine Test Dust, because these tests tend to be more accurate when compared to reality.

7.3 Removal Ratings

There are different ways of describing the function and efficiency of a filter. The various ratings within microfiltration are:

- Nominal rating
- Absolute rating
- Beta-value
- Titre-reduction

7.3.1 Nominal rating

The National Fluid Power Association (NFPA) defines nominal rating as ›*a micrometer value stated by the filter manufactured, based on the elimination of a number of percentage of all particles of a given size or larger*‹.

In practice the test is quite easy to perform since a suspension of contamination is introduced on the upstream side of the test-filter. After the fluid has passed the filter then either the filter or the flow leaving the filter is analysed, Fig. 22. Often the test-filter is weighed in a dry and clean condition before the test. The filter is then placed in a holder and the flow containing test contaminants is allowed to pass through. After filtration the filter is taken out of its holder, dried and once again weighed. The difference in weight between the dry and new filter and the dry and used filter is calculated. This difference is then divided by the total weight of the contaminants challenging the filter and the result is presented as the removal rating. A filter with a nominal rating of 98% of a certain particle size, for instance > 5 µm, will eliminate 98% of all particles greater than or equal to the given particle size, i.e.,

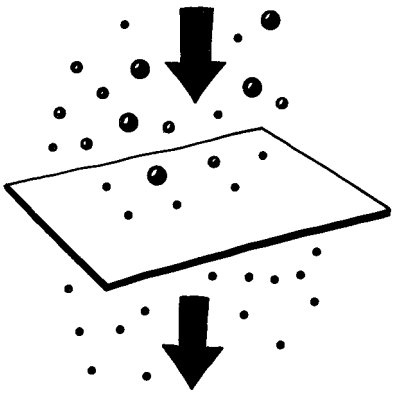

Figure 22. Schematic representation of a test used for nominal rating of a filter material.

> 5 µm. The rest of the contaminants (2%) have passed the filter during the test. The removal rating as a percentage is often expressed as percentage by weight and not as a percentage based on the number of particles (see explanation given in the legend to Fig. 23).

This test method is used quite often due to its simplicity, especially when several tests of different filters are to be performed in a standardised system and also when the filters are challenged with a known concentration of contaminants per unit time.

Another way to perform the test is to collect and weigh the particles that have passed through the filter, these particles are dried and then weighed. Nominal rating is often based on gravimetric analysis, i.e., analysis is performed by weighing before and after the test, which makes this method easy to use.

The contamination triangle shown in Fig. 23 shows the importance of differentiating between percentage by weight and percentage by numbers when the efficiency of a filter is discussed. To the left in this triangle there is a scale where the particle distribution is expressed as percentage by weight and to the right a scale where the particle distribution is expressed

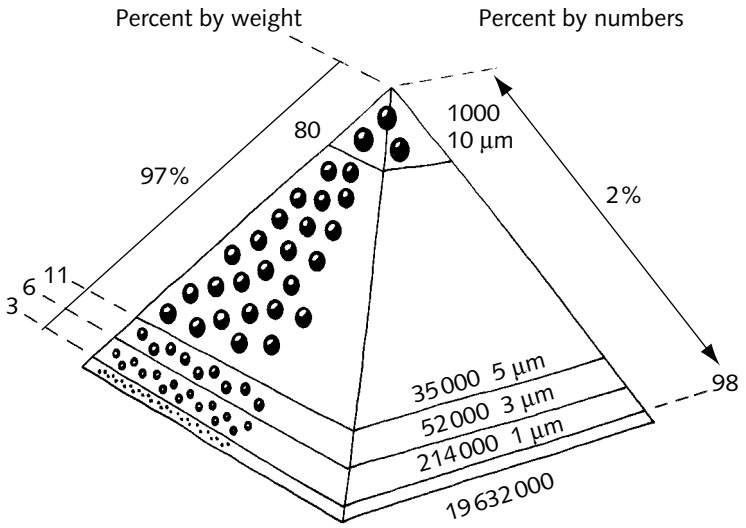

Figure 23. The pyramid of particles. This figure is used to facilitate the understanding of the difference between measurements based on percentage by weight and percentage by numbers.

by numbers. Let us presume that the filter can eliminate 97% by weight of all particles in a sample. According to the figure below this means that only 2% by numbers are eliminated. Nominal rating is only used on filters intended for the removal of dead particles.

7.3.2 Absolute Rating

NFPA defines absolute rating as ›*the diameter of the largest, hard, spherical particle that can pass through a filter under specified test conditions*‹.

Absolute rating is, for practical reasons, only possible to perform on filter media that have a structure comprised of fixed pores, like for instance membrane filters or depth-type filters with fixed pore structure. The definition includes spherical particles that are rarely observed in industrial applications. This is one of the reasons why the spherical particles in many cases are exchanged with other types of test contaminants, for instance ACFTD.

The actual test is performed in a system that looks quite like the one used for nominal rating, Fig. 24. When performing

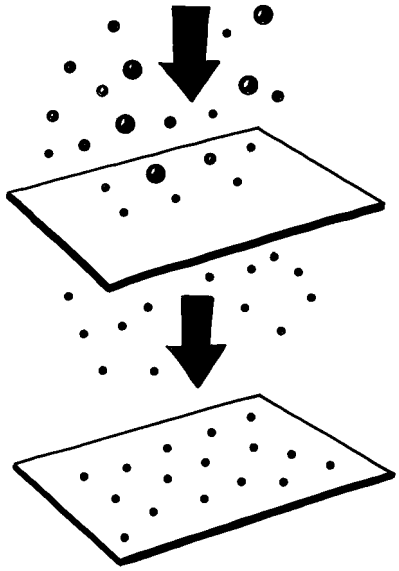

Figure 24. Schematic representation of the test used for absolute rating of a filter material.

this type of test it is not good enough to make a crude assumption of the weight of the different contaminants passing through the filter. Instead the size of the particles passing the filter must be studied. The particulate material passing the filter must therefore be collected, for instance on an analytical membrane and then studied under a microscope in order to find the largest particles that have passed through the test filter. Absolute rating is only used on filters intended for the removal of dead particles.

7.3.3 Titre Reduction

Both nominal and absolute ratings are used in order to describe the efficiency of a filter in regard to dead particles, i.e., particles lacking the ability to reproduce. Filters used to remove microorganisms are based on a totally different philosophy since the cleanliness after such a filter can be jeopardized by one single microorganism passing through the filter and entering the clean downstream side of the filter. This single microorganism can have the ability to reproduce if the envi-

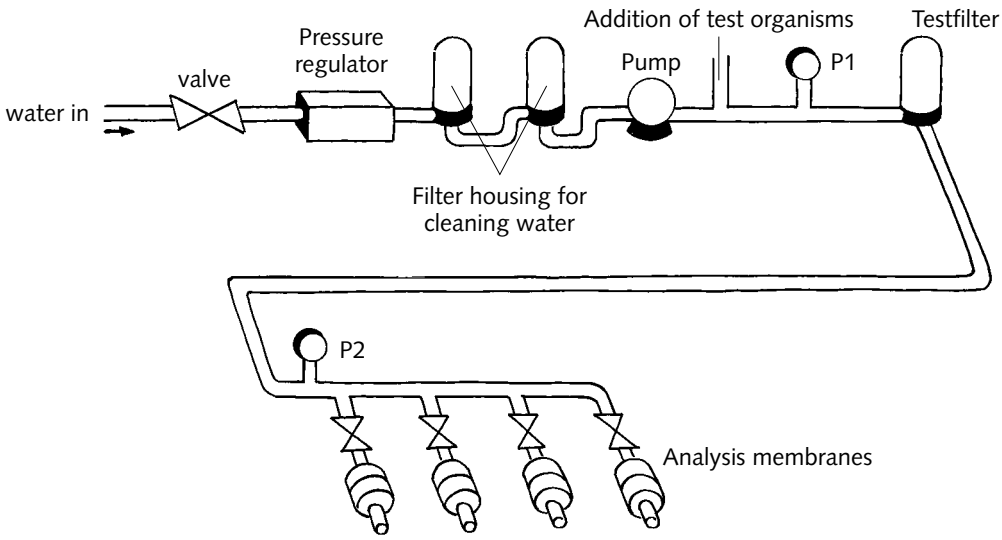

Figure 25. Schematic illustration showing the test apparatus used for evaluation of the titre reduction of a microbiologically rated filter.

ronmental conditions are right. Removal ratings expressing filter efficiency when collecting live particles such as microorganisms use titre reduction, Fig. 25.

Titre reduction is defined as ›*the number of microorganisms of a defined type that the filter is challenged with, divided by the number of microorganisms of the same type passing through the filter under defined circumstances*‹.

$$T_R = \frac{\text{number of challenging microorganisms}}{\text{number of microorganisms passing the filter}}$$

Titre reduction is often expressed as a logarithmic value, log T_R. If the titre reduction for instance is 1000 then log $T_R = 3$. If the titre reduction is 10,000 then log $T_R = 4$.

7.3.4 Beta-Value

Beta-value is, from a practical point of view, a rating method somewhere between nominal and absolute rating. Beta-value is not a new rating method since it has been used for a long time in performing tests on filters for hydraulics and lubrication oils. Beta-value can be difficult to use on a depth-type

filter with loosely bound pore structure because the function of these filters is not homogeneous and might vary due to variations in process conditions. For all filters with fixed pore structure, both depth-type filters and membrane filters used for removing dead particles, this method is extremely good.

Beta-value looks quite similar to titre reduction and is defined as ›*the relation between the number of particles of a certain size and larger that a specific filter is challenged with and the number of particles of the same size and larger that are found after the filter*‹. Beta-value is actually a filter rating method giving information on the efficiency of a filter in regard to the removal of number of particles.

$$\beta^{x} = \frac{\text{number of particles of size x and larger challenging the filter}}{\text{number of particles of the same size and larger passing through the filter}}$$

7.4 Conclusion

The removal capacity of a filter must be tested and documented in order to facilitate the discussion between the manufacturer and the user of a filter. The rating should differentiate the filter from other filters and it should also be in accordance with the results obtained by analysis and test of the filter function.

8 Choosing a Filter

8.1 Introduction

Filter media presently available, and even more so in the future, are tailor-made in order to comply with the intended use and service. This is one of the major reasons why greater knowledge and understanding of filtration technology are needed. The knowledge must incorporate the filter medium to be used, how it is constructed and designed and, most of all, knowledge of the total process and the demands of cleanliness stated by the filter user.

The most important factors to be considered when choosing a filter and/or a filter system are the dirt collecting capacity of the filter (often called just dirt capacity), the physical as well as the chemical features of the filter, together with the design and function of the process in which the filter is going to be in service.

8.2 Dirt Collecting Characteristics

One of the first and nearly always asked question when choosing a filter is: ›How long will the filter be able to be used in a system, i.e., how many litres of liquid or cubic meters of gas will be able to pass through the filter before it becomes blocked and has to be exchanged with a new filter‹?

This question is quite simple and also a natural question to ask. However, the answer is much harder to give. The total service life of a filter depends on many different factors that together will have an impact on it. Another reason why this question is hard to answer is that in practice it is nearly impossible perform small-scale trials that can be used in a scale up of the process.

The dirt collecting capacity and function of a filter depend on the design of the filter and its flow characteristics, together with the characteristics of the total fluid to be filtered. The impact of the filter depends on factors like the total available

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area, the porosity and the rating. The overall result of the filtration depends also on the size of the flow, the amount of liquid or gas that is to be filtered per unit time, together with the number of particles, the size distribution of the particles and their physical characteristics. The characteristics of the particles include the type of particles, where they come from etc. All these factors must be included and put in relation to one another and also to the different cleanliness demands desired from the filtration process.

8.3 Overall Process Demands

There are several overall factors that might have an impact on the process, the choice of filter and its dimensions. These factors comprise the total cleanliness demand of the process, the sanitation or sterilisation of the total process system together with whether the process is performed continuously or in a batch-wise manner. Sometimes the situation might be even more complex, for instance if several different batches and different products are to be filtered with the same filter system.

8.4 Cleanliness Demands

What is the meaning of clean? What demands can and must be stated in order to obtain a certain level of cleanliness? How can the correct cleanliness level in respect to certain process parameters be determined? Is it possible to control whether the correct cleanliness level is obtained? The list of questions can be very long. To define cleanliness it is necessary not only to investigate at a certain point in a process, but also to look at the total process from a holistic point of view. In order to obtain an economically as well as technically acceptable solution, consideration must be given to what has happened previously in the process and also to what will happen later in the process, i.e., after the filtration is performed.

8.4.1 Sterility

As mentioned earlier, there is only one absolute defined cleanliness level in regards to microfiltration, sterility. Sterility is an absolute level and means that the sterile process flow or sterile process equipment is totally freed from living organisms, regardless whether the organisms are pathogenic (have the ability to cause sickness) or not.

With this definition of sterility parallels can be drawn to microfilters and microfiltration, namely to the area that is called sterile filtration. It is quite easy to get the impression that a sterile filter must correspond to the definition of sterility. This is however not the case. FDA (Food and Drug Administration, USA) defines a sterile filter as ›a filter with a capability if challenged with at least 10^7 *Pseudomonas diminuta* (ATCC 19146) per cm^2 of filter area, of allowing not one single organism to pass through the filters, i.e. giving a sterile effluent‹. Filters, corresponding to this definition are, furthermore, allowed to be called a nominal 0.2 µm or a nominal 0.22 µm filter. In practice this definition means that a sterile filter can be challenged with these numbers of microorganisms without giving rise to non-sterility.

According to FDA ›Guidelines on Sterile Drug Products Produced by Aseptic Processing‹, Center for Drugs and Biologics, Rockville, MD June 1987, *Pseudomonas diminuta* has recently been reclassified to *Brevundimonas diminuta* ATCC 19146. The size range of this microorganism is 0.3–0.4 µm in diameter and 0.6–1.0 µm in length.

Many of the sterile filters used today have a much higher level of open area, porosity, than the filters available at the time when this definition was stated. In practice this means that presently available filters will be able to meet much larger challenge levels before they are blocked. The definition of a sterile filter given by FDA is still valid today despite the fact that new and much more potent filter products have been developed. Many of the sterile filter manufacturers are today still using this FDA definition. These filters however, will give sterility up to a minimum of 10^7 *Brevundimonas diminuta* per cm^2 of area since they are tested up to this challenge level. A natural question to ask is what will happen when such a filter is challenged with more organisms. In order to gain greater security all sterile filters must be tested to total blockage, i.e., when no more flow is able to pass the filter.

In order to make a correct choice of filter, knowledge of the demands from the FDA together with the possibility of discussing your demands with the filter manufacturers must be available. The results from challenge tests must be readily available for the filter user together with a thorough description on how to interpret these results.

8.4.2 Industrial Cleanliness

The term ›industrial cleanliness‹ is sometimes used in industry, indicating a slightly different cleanliness as compared to sterility. The demands of industrial cleanliness can be lower than the sterility demand but the demands can also be higher. The most difficult question to answer is what cleanliness level you need for a certain process fluid or product. To answer this question it is necessary to consider the sensitivity to the product, the overall production system and its design, the packaging and transportation of the product. Other factors that might affect the product are how it is to be stored, its intended use and also the desired shelf-life of the product.

Different branches of industry have different demands on cleanliness. Within the dairy industry yeast and molds are considered the greatest risks during production. The same risk applies to the manufacturing of soda and cider. The elimination of yeast and molds to a suitable level for these industries can be obtained in a filter with, for instance, an absolute rating of approximately 1–2 µm or lower.

Beer and wine manufacturers often state higher cleanliness demands. Not only yeasts and molds are critical, but also the presence of other microorganisms and especially microorganisms that might have a negative influence on the product. The level of industrial cleanliness for the beer and wine industry is therefore slightly higher than just eliminating yeast and mold. Depending on what type of microorganisms that might be harmful to the product, the removal ratings of filters used in these industries have absolute ratings from 0.65 µm, 0.45 µm or 0.35 µm.

The different µm values stated above are used when describing ratings of filters used for the elimination of microorganisms. The numbers 0.35 µm, 0.45 µm and 0.65 µm respectively, are actually designations of the different filters based on titre reduction for certain well defined test organisms, see Tab. 4 below. The stated µm values are not the pore size of the filter media. The term pore size should actually not be used at all. This strange situation can easily be described by looking back in history. During the 1970s a sterile filter was defined and called a 0.45 µm filter and was tested with the microorganism *Serratia marcescens*. This microorganism has a size range of approximately 0.6-1.0 µm.

Table 4. Examples of different microorganisms used for the classification of filters for microbiological removal.

Removal rating/µm	Organism
0.10	*Acholeplasma laidlawii* *Brevundimonas diminuta*
0.20	*Brevundimonas diminuta* *Burkholderia pickettii*
0.45	*Serratia marcescens*
0.65	*Leuconostoc oenos* *Lactobacillus brevis*

The pharmaceutical industry in some cases states higher levels of cleanliness than sterile filtration. Such a situation can occur when there is a need to eliminate viruses, mycoplasma and/or pyrogens. This is another form of industrial cleanliness that is far below the sterility level of 0.2 or 0.22 µm. Industrial cleanliness is, as seen from the examples given above, dependent on many different factors and can vary considerably both within and between different industrial applications.

8.5 The Process Flow

The size of the process flow does not play a vital role in regard to cleanliness when a filter with fixed pore structure is used. However, the size of the process flow has a major impact on the total service life of the filter medium. The higher the flow passing through a filter per unit time, the shorter the service life of the filter. In practice this means that the filter will be blocked faster at higher flow rates than when used at lower flow rates, Fig. 26.

This figure shows schematically how the pressure drop across a filter will vary with the flow rate over the filter. The pressure drop will increase linearly with the increase in flow rate.

From a practical point of view the observed dirt capacity of a filter (which is one way to express the life of a filter) can be related back to what is often called the flow density. Flow density is the size of the flow through a filter divided by the

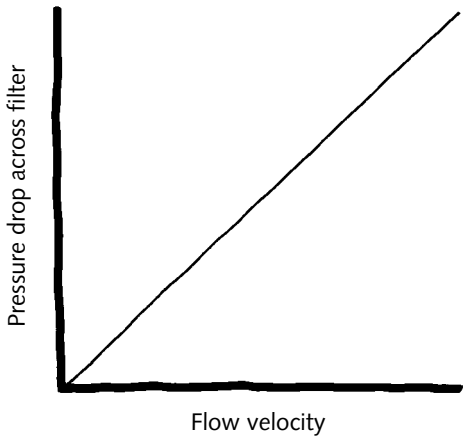

Figure 26. The pressure drop across a filter medium.

area of the filter. The following example explains the situation mentioned above. A homogeneous, porous filter medium is exposed to a certain constant flow per unit time. The life of this filter is set to 1 time unit. When the filter is used in a situation where the flow is twice as high on the same filter area, two things will happen. First, twice as many particles and other contaminants per unit time will challenge the filter. Second, the amount of contaminants or dirt due to the stronger force from the flow will be more heavily forced into the filter medium. Figure 27 explains schematically what will happen when the filter area is increased during a constant process flow.

In Figure 27A, a filter with an area of 1 unit is used for filtration. Figure 27B shows the same filter material but with 3 times higher area. When the filter with the smaller area collects particles, these are, in this theoretical example, collected on the surface of the filter. When particles have been collected up to a thickness the size of 1 unit, ΔP or the differential pressure of the filter is at its maximum. The filter is at this stage totally blocked and cannot be used to collect any more particles.

When the filter with the larger area is used to collect particles to the same thickness, i.e., 1 unit, the pressure drop over the larger filter will become 1/3 of ΔP, as compared to when

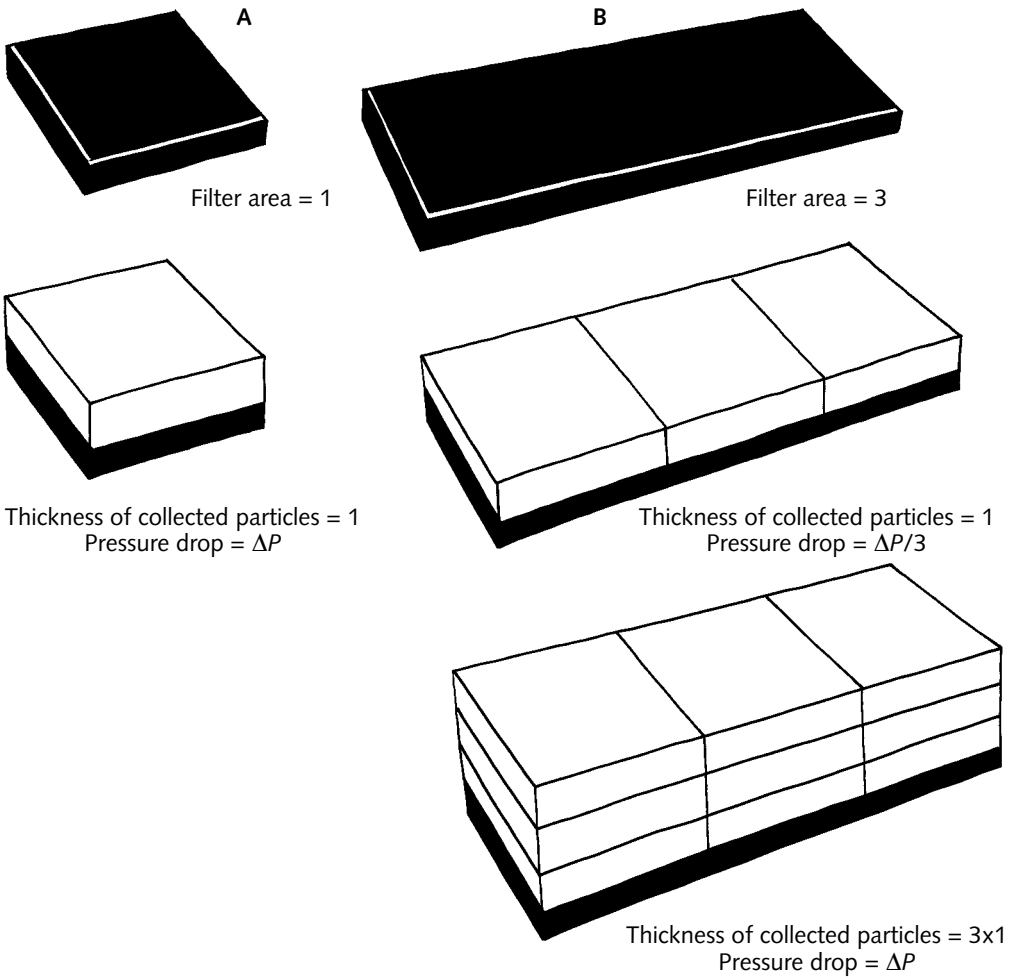

Figure 27 The influence of surface area on the life of a filter. The life of a filter can be viewed as the dirt collecting capacity of the filter.

using the smaller filter. In practice this means that the larger filter is capable of collecting three times as much dirt on its surface before the final differential pressure, ΔP, is reached. In theory this means that the collection of dirt on the larger filter is $3 \times 3 = 9$ times higher than with the smaller filter.

The example discussed above indicates that an increase in the surface area of a given filter at constant process flow and

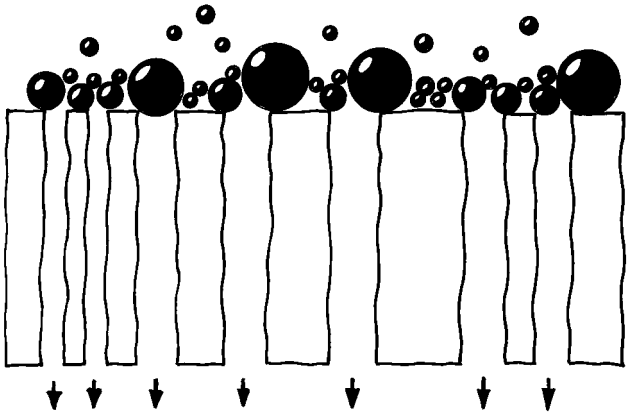

Figure 28. Larger particles collected on the surface of a filter will form a cake which in turn will act as a pre-filter for smaller particles in the flow.

process pressure will result in an increase in the dirt collecting capacity and thereby also in the theoretical service life of the filter. From a mathematical point of view the increase in service life of a filter will correspond to the square of the increase in the area of the filter. From a practical point of view, the life of the filter will increase somewhat less because the filter in the given example is assumed to work as a surface filter, i.e. a filter that only collect particles on its surface and not in the depth of the filter.

The size distribution of the various particles existing in a process flow is also of major importance for the service life of the filter. The larger the particles in the flow, the higher the possibility for the filter to work in close resemblance to a surface filter. Larger particles present in a process flow will to a very high degree be collected on the surface of the filter since these particles cannot penetrate into the pores of the filter medium. If larger particles are collected on the outer surface of a filter, these particles will act as a pre-filter by forming a porous filter cake on the surface of the filter. The porous cake thus formed will act as a pre-filter for all the smaller particles existing in the flow, Fig. 28.

8.6 The Pressure of the Process Flow

The pressure of the process flow and its impact on the choice of the filter is divided into the total pressures in the flow and the differential pressure across the filter.

8.6.1 The Total Pressure

The filter housing, i.e., the solid container in which the filter membranes or the filter cartridges are mounted, is in most cases identified as a pressure vessel. Many national as well as international standards and references will affect the choice of the filter housing or filter container in regard to the total pressure in the filter system. The total pressure does not influence the choice of filter at all. The differential pressure, however, is of utmost importance.

8.6.2 Differential Pressure

When filter cartridges or other types of filter material are placed in a process flow, the total pressure will have a minor impact. The impact of pressure on a filter is mostly dependent on the differential pressure across the filter, Fig. 29. The differential pressure (often called just delta-*P*) is defined as the difference between the pressure on one side of the filter, the pressurised side or upstream side of the filter (pressure measured by manometer P_1), and the pressure on the other side, the downstream side of the filter (pressure measured by manometer P_2). The pressure on the downstream side of the

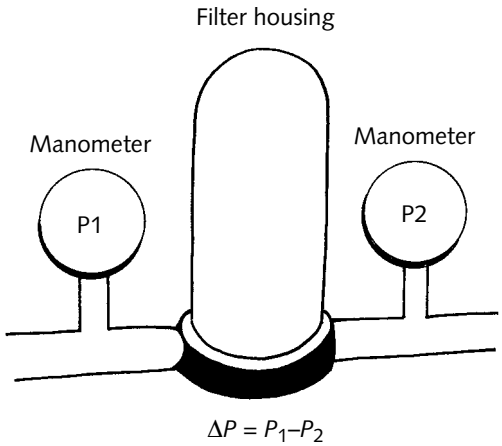

Figure 29. Schematic representation of the pressure drop across a filter system.

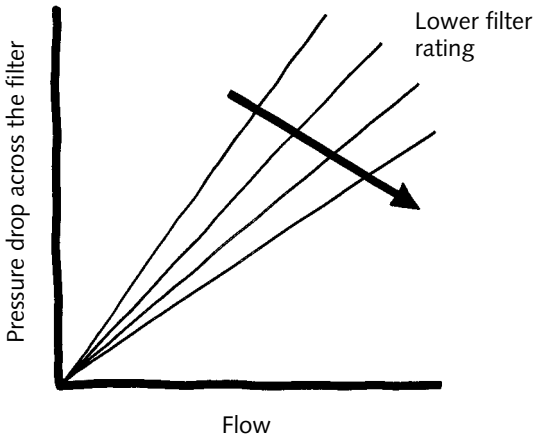

Figure 30. The pressure drop across a certain filter material available with different removal ratings. When observing a filter system having the same filter area it turns out that the better the rating of the filter, the higher the pressure drop .

filter is either atmospheric pressure or a higher pressure, depending on the function and design of the process.

When making a comparison between different filter media the initial differential pressure (delta P_{init}) is often used, i.e., the pressure drop over a clean and totally new filter at the start of the first filtration cycle. The initial differential pressure depends on the flow velocity of the fluid, the viscosity of the fluid and the temperature, the available surface area of the filter together with the porosity and removal rating of the filter material.

To choose a filter with respect to the removal rating is an excellent way to begin after the cleanliness demand of the process has been defined, Fig. 30. If the filter of choice shows a too high pressure drop, the total system can be altered in various ways in order to obtain the desired cleanliness of the filtered liquid or gas while the lifetime of the filter increases. This increase in the filter life can be achieved either by increasing the filter area or decreasing the total flow rate through the filter (i.e., decreasing the flow density across the filter). In either of these cases the flow density, i.e., the total flow

through the filter divided by the area of the filter, will increase.

8.7 Viscosity of the Process Flow

The viscosity of a liquid can easily be explained as the internal resistance in the liquid to flow. A liquid with high viscosity will be harder to pump and harder to flow through a pipe. This internal resistance will make it difficult to filter high viscosity liquids. The differential pressure across a filter will increase dramatically when filtering viscous solutions. In order to get a functioning filter system in practice, the total filter area must often be severely increased compared to when considering the dimensions of the filter from a traditional point of view.

Another way to cope with a highly viscous solution is by heating the liquid which will decrease the viscosity. By decreasing the viscosity the liquid will flow more easily through a filter medium. An industrial example of such a situation is the filtration of concentrated sugar solutions. Highly concentrated sugar solution can show high viscosity levels at room temperature. By performing the filtration at 60 °C instead of at 4–10 °C, highly concentrated sugar solutions can be filtered without using large filter areas and filter systems.

The viscosity will have an impact on the differential pressure in the filter system. Generally one can state that the higher the viscosity, the higher the pressure drop across the filter. A direct linear relation exists between the viscosity of a flow and the pressure drop over a filter.

The following example shows differences in differential pressure when comparing the filtration of water with the filtration of a sugar solution in the same filter system.

Water of a predetermined flow and pressure is to be filtered through a filter having a clean pressure drop (delta *P* init) of 200 mbar at a viscosity of 1 cP (centipoise). A sugar solution with a higher viscosity, 20 cP, is to be filtered using the same filter system. The pressure drop when filtering the sugar solution will be 20 times higher than for the filtration of water, i.e. 20×200 mbar $= 4000$ mbar $= 4$ bar. This filter system dimensioning is not acceptable from an economic point of view. If the filter system used in the example has a maximal pressure differential of 5 bar, the difference between

the maximum ΔP and the initial ΔP will be too small. The total dirt collecting capacity in such a filter is in terms of pressure 5 bar – 4 bar = 1 bar.

This, and corresponding problems, are however sometimes quite easy to solve. One way is to decrease the flow through the filter in order to decrease the total differential pressure over the filter. If the overall process demands that the higher flow must be retained, the flow rate through the filter cannot be decreased. In this case either the filter area must be increased or the temperature of the liquid to be filtered must be increased, or both.

By choosing a filter in the above example that is 8 times larger, the initial differential pressure during filtration of the sugar solution will decrease from 4 bar to 4000/8 mbar = 500 mbar. By increasing the area of the filter surface it will be possible to filter the sugar solution at the desired flow rate without altering the temperature of the sugar solution.

If it is possible to increase the temperature of the sugar solution to a higher level the viscosity will decrease, making it possible to filter it through the same filter system that has previously been used for water. By choosing a correct temperature for the liquid to be filtered it is possible to use a smaller filter system. The differential pressure will decrease at the same time as the flow through the filter will be at a more acceptable level. The discussion above is schematically shown in Fig. 31. In total all of the factors discussed will lead to filter systems that are smaller in size, which in turn leads to the fact that the number of filter cartridges in the system can be decreased, resulting in a lower filtration cost.

8.8 Compatibility and Temperature of the Process Flow

Compatibility from a general point of view is often defined as how different materials and substances interact when they come together, i.e. how a process flow will interact with the material in a process system. If a process flow is not compatible with a process component, for instance a filter, more or less of the filter material can be dissolved and extracted out into the fluid.

A very good example of the influence of temperature on compatibility is shown when a filter medium manufactured

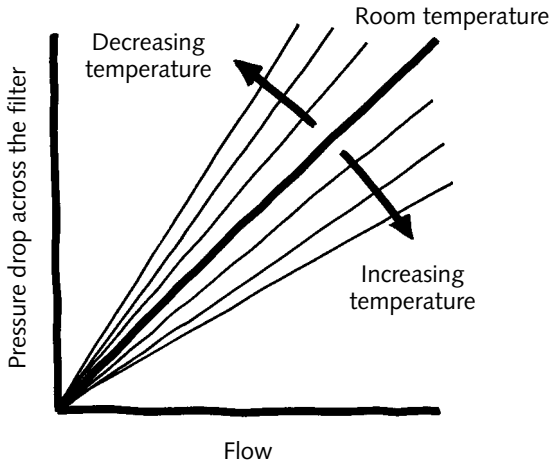

Figure 31. The impact of viscosity on the pressure drop across a filter in relation to the temperature of the fluid to be filtered.

from nylon is placed in a process flow of re-circulating hot water (> 85 °C). The hot water will, especially if it contains dissolved oxygen that is strongly oxidising, dissolve the nylon filter medium. The dissolved nylon will be added to the solution of hot water, not only as particles but also as dissolved filter material. The stated example illustrates the effect of very poor compatibility since the dissolving process is extremely rapid. Several filter manufacturers publish compatibility charts, publications containing tables showing the compatibility of their different filter materials to various single chemicals. Compatibility data can be reported in various ways. Some years ago it was not unusual to report this data in more or less subjective terms as, for example, excellent compatibility, good compatibility and bad compatibility, Table 5. Using such subjective terms of compatibility is not satisfactory for the filter user when choosing filters. Most of the industries using filters today have become more and more sensitive in regard to compatibility and demand exact numbers of what can be extracted from the filter material, how large this extraction is and also under what conditions the stated compatibility tests have been performed. It turns out that most filter media are fully acceptable when the compati-

Table 5. A poor example of expressing the compatibility of different filter materials. R = recommended, L = limited use, N = not recommended

	Acetic acid (5%)	**Boric acid**	**Hydrogen chloride**	**Sulfuric acid**
Filter material A	R	R	L	L
Filter material B	R	R	R	L
Filter material C	R	R	L	N

bility test is performed at 10 °C. After increasing the temperature some 10 °C and even more, this material can release large levels of extractable material, making the filter unacceptable for use in the desired processes.

Some manufacturers test their filters in regard to compatibility by using a static process. Such a static test is performed by taking a known amount of filter material and placing it in a beaker filled with a solution to be examined. After a certain time period the tested filter material is removed. The solution together with the different substances that have been extracted from the filter medium is then analysed.

A dynamic compatibility test is performed in such a way that either the filter material is placed in a test rig, forcing the filter to move up and down in the solution, or a test rig is used comprising a filter placed in a filter housing through which the process flow is continuously re-circulated. After a certain predetermined time the filter test is aborted. The filter is removed and the test solution, together with the extracted substances, is further analyzed.

8.9 Depth Filter or Membrane Filter

Filters are not only grouped in regard to their physical appearance and the bonding of the pore structure of the filter material (depth-type filter and membrane filter), but also depending on how they are constructed, i.e., if they have fiber-based or polymer-based filter media.

Filter media constructed from fibers are mostly used for coarser types of filtration. The reason for this is that it is practically difficult to manufacture a filter medium comprised of fibers with removal ratings below 0.5 µm. Correspondingly, filter media constructed of polymeric materials have

ratings from approximately 1 µm and downwards. This differentiation is due to the fact that it is hard to manufacture polymeric filter media with larger pores. Filters with lower removal ratings are in general more expensive to buy than filters with higher removal ratings.

8.9.1 Choice of Filter Type

It is often totally wrong, for reasons of quality, to choose a filter constructed of fibers when intended for the removal of microorganisms from a process flow, especially if quantitative removal is needed. In the same way it is wrong, but in this case for reasons of economy, to choose a polymeric filter material in order to remove coarser or larger particles. The polymeric filter, which is more expensive, will become blocked too quickly. A good filter system, from both an economic point of view and a practical qualitative point of view, comprises for instance a fiber-based filter material which can be used as a pre-filter before a polymer based final filter (for instance a sterile filter).

The differences between depth filters and membrane filters are not only the physical appearance of the filter and the differences in removal efficiency. The large internal volume of a depth filter will result in higher initial pressure drops as compared to a membrane filter. This phenomenon will be independent of whether the pore structure of the depth filter is bonded or unbonded.

8.9.2 Hardware and Software

A filter system in general is comprised of two parts, a hardware part represented by the filter housing or the filter holder in which the filter medium is mounted, and a software part comprised of the actual filter material (either a cartridge or a filter disc). The filter material is responsible for the actual cleaning of the fluid passing through the filter.

Depth filters will in general show higher initial pressure drops than membrane filters and for this reason will often be larger, i.e., when using depth filters there is often a need for much larger filter housings. This larger filter housing is vital in optimising the service life of the total system. From an economic point of view the investment costs in filter housings will in general be higher when depth filters are used.

When using membrane filters it is often possible to use

smaller filter housings. In general there are no official guidelines to follow when making a choice between depth filters and membrane filters. Many different factors will have an impact and must be taken into account for each unique filtration application.

8.9.3 Liquid and Gas Filtration

Filtration of liquids and gases is performed with the aid of mechanical and adsorptive retention as discussed previously. When liquids are filtered only mechanical retention and adsorptive retention by impaction are involved. When filtration of gases is performed, both mechanical retention and adsorptive retention through impaction as well as diffusion interception are involved. Diffusion interception will only affect extremely small particles, which is the reason why removal of particles will become greatly increased when filtering gases as compared to liquids.

Adsorption due to diffusion interception only occurs, from a practical point of view, if and when the filter is in a dry state. If the filter is wetted by some kind of liquid the diffusion interception action of particles will disappear. The action of such a filter that has been wet will instead correspond to the removal efficiency of a filter that is used for filtration of liquids.

Since the wetting liquid also will block some of the pores in the filter medium, an increased risk of higher-pressure drops over the filter will exist. This is why filters for gas nowadays in most cases are manufactured from hydrophobic (i.e., non-water-wettable) filter media.

Another way to retain a filter in a dry state is by heating the filter system above the dew point. This can be performed by, for instance, heating the filter housing or the filter holder with an external electrical heating coil. Another way is to use a mantled filter house where steam is allowed to continuously pass through the mantling.

8.10 Conclusion

The most important factors having an impact on the choice of filter and filter systems are the dirt capacity of the filter, the physical and chemical compatibility of the filter together with the design and function of the process.

9 Sanitation and Sterilisation

9.1 Introduction

Sanitation and sterilisation are processes that are used to clean a filter system, either by removing dead contaminants, microorganisms or by killing the microorganisms. Moreover sanitation and sterilisation processes will affect all biological and temperature sensitive material, for instance coagulated protein and dead cells, disintegrating them and, to a certain extent, transporting them out of the filter medium in a dissolved state. This type of external effect leads to, from a microbiological point of view, a cleaner filter system and also to a certain degree of reconditioning of the filter. Such reconditioning can in certain cases result in a decrease in the pressure drop of the filter, in practice meaning that the filter can be used for a longer service time.

The sanitation and sterilisation of filter systems is of extreme importance. All filters must be cleaned at some time interval, even filters for domestic use, such as the so-called home-water filters used for cleaning tap water. The domestic market has for some years become of increasing interest to companies working with filtration. The producers of tap water state that their product is of sufficiently good quality. However, newspaper articles on the poor quality of drinking water have caused many people to feel the need to purchase water filtration equipment for private use in homes, caravans or boats. One problem with this type of equipment is that it can create a false safety. The water quality after passing such water treatment systems can sometimes even become worse when using a filter than that of the tap water without filtration.

Filters for domestic use are often sold at a lower price than those used in industry. This low price reflects that these filters are much more primitive in their design and construction. In many cases they can be compared to filters with unfixed pore

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structure. The primitive design of these filters brings no danger if the filter is maintained and controlled on a regular basis, for instance by changing the filter according to the manufacturer's recommendations. The danger may arise when the filter is mounted without any type of sanitation before or after a certain time of use. In many cases the user instructions are not easily understood. The risk that such domestic filters that are not exchanged, sanitised or sterilised will actually function as a fermenter of microbiological contaminants is high. This is the underlying reason why it is as important to sanitise and clean domestic filters as it is for filters used for industrial use.

From a general point of view sanitising and sterilisation methods can be divided into the following:

- Chemical treatment
- Hot water sanitation
- Sterilisation using steam, radiation or gas

Steam sterilisation can be performed both by the user and the manufacturer of the filter, whereas radiation or gas sterilisation in most cases is not performed by the user. In this book only sterilisation using steam is discussed.

9.2 Chemical Treatment

Chemical treatment means that the filter and the filter system are exposed to different types of cleaning chemicals. Chemical treatment will, to a certain extent, kill the microorganisms and at the same time they will be dissolved and disintegrated. One way to perform chemical treatment is to remove the filter cartridges from the filter system and place them in a chemical solution overnight, a principle often referred to as a static chemical treatment. This method is desirable from a practical point of view, since in this way the filters are not exposed to the different contaminants that might be present in the chemical solution.

CIP, Cleaning in Place, is a chemical treatment method in which the filter cartridges do not have to be removed from the process system during the treatment. The cleaning solution is instead continuously recirculated through the entire process system at high speed, Figure 32.

Many different types of cleaning solutions are available that are potent enough to sanitise filters and filter systems. It is however very important to ensure that the filter media as well as all the other components of the process are compatible with the solution. When using CIP some of the particles that the cleaning solution is removing during its cleaning action will be collected in the filter. From a practical point of view this means that the CIP solution will clean the process system and the filter will capture many of these contaminants, a situation generally resulting in a shortening of the service life of the filter. Thus, during a CIP cleaning of a process system, it is quite common to remove the filters during the CIP action. Furthermore the top of the filter housing is often removed and replaced with a small lid in order to maintain the speed of the CIP solution through the system. The removed top of the filter housing can thereafter be washed, for instance by hand, before it is replaced.

9.3 Hot Water Sanitation

In order to avoid problems such as that described above during chemical treatment, hot water sanitation may be used in order to sanitise filters and filter systems. Compatibility is again extremely important in this type of sanitation since the filters and the rest of the process components must be able to be subjected to hot water. The hot water is allowed to pass through the filter in exactly the same way as during the CIP wash but it is much easier to keep and hold the particulate control of hot water as compared to CIP solutions. One of the reasons for this is that the CIP solution is often not discarded after one wash, but used over and over again. Hot water on the other hand is normally used once, which means that hot water will not accumulate contaminants in the same way as the CIP solution. The treatment time for hot water sanitation is in many cases also shorter than that of chemical treatment.

In general hot water at around 85–90 °C is used. The total contact time may vary from 10–30 min, depending on the cleanliness demands as well as the degree of contamination on the filters and in the system. Since filters may be present at different positions in a process the temperature as well as the

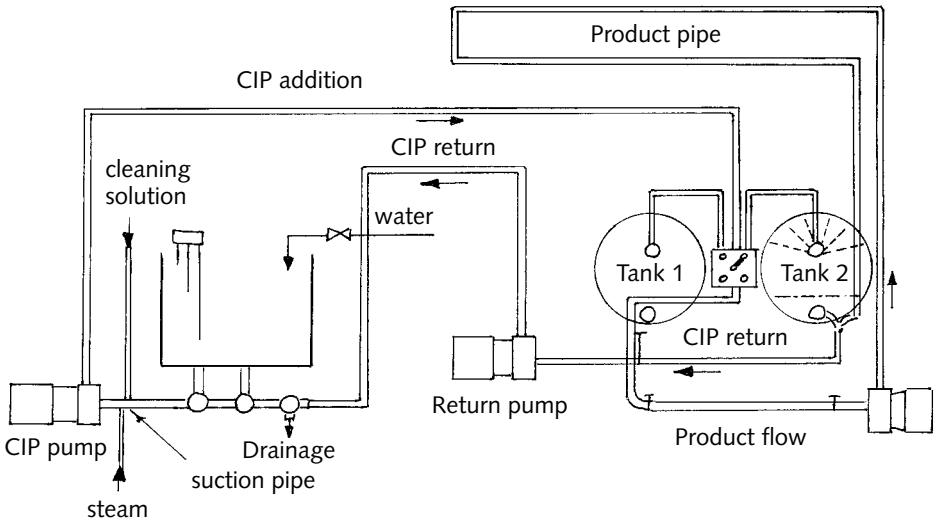

Figure 32. A schematic representation of a CIP (cleaning in place) system.

contact time at the actual temperature may vary. A filter placed close to a filling machine will therefore be affected more than a filter that has no components after it (final filter). This situation arises because the counting of the exposure time will not start until the last and the coldest component in the process chain has reached the determined sanitation temperature.

9.4 Steam Sterilisation

During hot water sanitation the total amount of energy stored in the water is used to clean the filter and the filter system. Steam stores and transfers energy much better than hot water. This is one of the reasons why steam sterilisation is a much better choice. Steam sterilisation is mostly used when there is a demand for sterility in a process system.

Figure 33 shows that the energy exchange during steam formation at a certain temperature will take place in three different steps. In the first step a certain amount of energy is added in order to raise the water temperature to 100 °C. During the second step, a certain amount of energy is added,

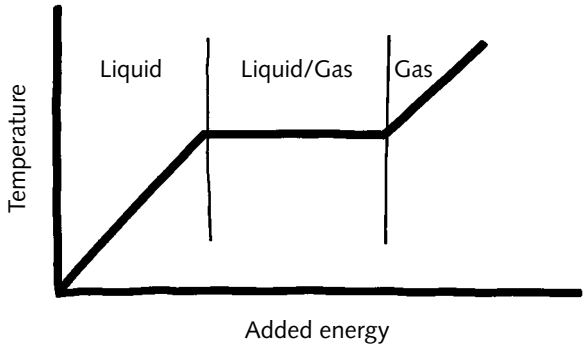

Figure 33. Water is a fantastic medium in which to store energy in the form of heat. This figure demonstrates the energy levels when heating water to produce steam. In the first step cold water is given energy in order to produce liquid water at 100 °C. The second step is the addition of energy in order to transfer liquid water at 100 °C to steam at 100 °C. The third and final step concerns the addition of energy in order to produce steam at the desired temperature.

in order to transform water in a liquid state at 100 °C to steam at 100 °C. In the third step, a certain amount of energy is added in order to raise the temperature of the steam from 100 °C to the desired temperature.

A very common but incorrect opinion amongst many users of steam is that steam itself is self-cleaning. This is in many cases not true. The steam that is mostly freed from particulate material when leaving the steam generator will become more and more dirty during its transportation to the point of use. It is quite easy to study the quality of steam in such systems by taking samples of the condensate that is collected in the pipes when the steam is not in use. In many cases such samples show that the condensate (and thereby also the steam) is dirty. Steam for sensitive products and process equipment must therefore be freed from contaminants such as particulate material. Many different quality norms exist. One is presented by the organisation in the USA, ›3 A Accepted Practices in the Dairy Industry‹, stating that steam must be filtered to at least 5 µm. In industry, a 1 µm filter has become an industrial standard. Finer removal ratings exist, even down to 0.1 µm for very critical applications.

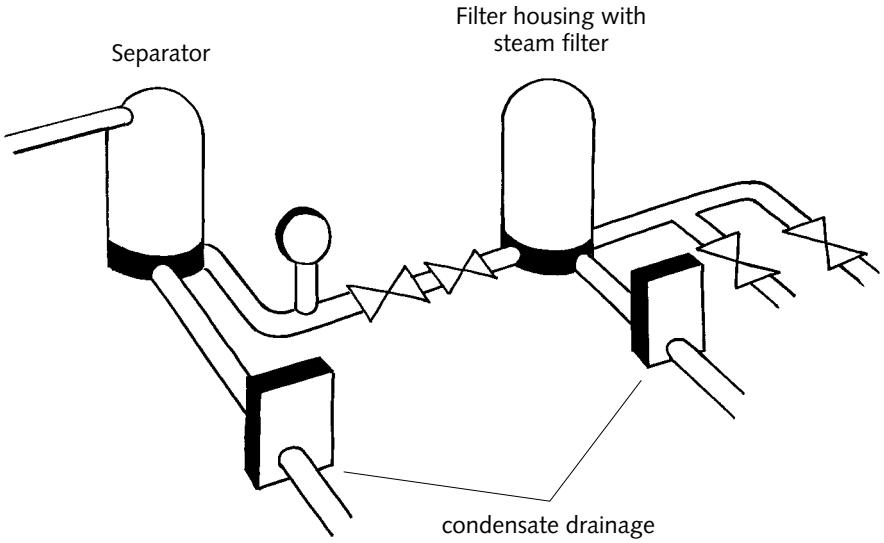

Figure 34. Schematic representation of a steam system according to 3 A.

Figure 34 shows a filter system for production of steam of Culinary Quality, i.e., steam for food and beverage applications. This steam quality, where steam is produced from a water corresponding to normal drinking water, will be suitable not only in many traditional sterilisation productions of food and beverages but also within the pharmaceutical industry. First, the steam is allowed to pass through a system eliminating small water drops and also, to a certain extent, particles transported with the steam. Then a filter is used to eliminate particles greater than or equal to 5 µm (today the industrial standard is 1 µm). All the equipment, placed after the filter is made of stainless steel, either mirror polished or electro-polished. All valves are also approved for aseptic production. More information about other and finer cleanliness levels of steam are to be found on page 137.

Steam sterilisation can actually be carried out with the filter placed in the filter system, i.e., sterilisation in place, SIP, see Section 9.4.2, or with only the filter or individual filter cartridges being autoclaved. In either case, the sterilisation cycle

itself must give a maximum possibility for non-sterility corresponding to 1 to 1,000,000.

9.4.1 Autoclaving

Most of the filters that are used in order to eliminate microorganisms are steam sterilised in some way to at least 125 °C. In some cases the filter function may vary or change, or the amount of extractable substances will increase, if the temperature is increased. It is possible to increase the temperature if it is clear that the filter will not be changed by this action.

When performing autoclaving the actions taking before sterilisation are of major importance. The goods to be sterilised must be covered in a suitable microbiological barrier that must be chosen so that the steam may pass through the barrier in order to come into contact with the filter of the filter system. In order not to create a differential pressure within the filter during steam sterilisation, the inlet as well as the outlet of the filter must be totally open to allow steam to pass.

Air present in the system will decrease the sterilisation effect. Hence air is removed from the autoclave at the start of the sterilisation cycle or the entire autoclave chamber is flushed with steam.

9.4.2 Sterilisation in Place

Sterilisation in Place or Steam in Place (SIP) is performed during overpressure and with a filter placed in its correct location in the system during the process. This sterilisation method demands that the filter housing in which the filter is placed, together with all other equipment in the process system, for instance piping, vents etc. are able to resist and also are approved for the steam pressure (15–30 psig) that the sterilisation demands. Also, the filter must not be exposed to higher differential pressures than stated by the manufacturer.

Since the steam temperature will decrease when steam is introduced into the cold process system the steam will condense, i.e. go from the gas phase to the liquid phase. In order to allow steam to flow through the system continuously this condensate must be removed and so the process system must be thoroughly drained. It is also of vital importance that the flow of steam always travels in the correct direction, since many filter cartridges are constructed for flow in only one

direction and have a much lower stability if the flow is directed in the reverse direction.

The sterilisation temperature is reached when the steam pressure and the flow, together with the fact that the steam is allowed to hiss out from the system, is regulated at the same time as a differential pressure over the filter is kept within the limits stated by the manufacturer. The entire system must however be flooded with steam.

When the steam sterilisation is complete a positive pressure must be retained within the filter system. This is accomplished by allowing pressurised air or other suitable gas to flow through the system during the entire cooling step. This is especially important when steam sterilisation is used on hydrophobic filters used for gas filtration or as breathing filters on, for instance, tanks, when there is a risk that steam will condense on the filter. If steam condenses in a filter, the pressure differential in the system will increase, which can lead to damage of the filter material.

Many different systems for steam sterilisation exist, for instance a single filter system with one filter house and mounted filter cartridges, or a process system where the filter system itself is followed by a system with, for instance, a buffer tank before a filling machine.

9.4.3 Single Filter System

In a single filter system the filter housing is connected to pipes containing product-flow, steam and pressurised air, Figure 35. The steaming of the filter is accomplished by the successive addition of steam in order to increase the pressure in the system. At the same time all the vents are kept cracked open, first in order to create a system where steam is always allowed to pass through the system, and secondly to expel the air that normally exists in the system and replace it with steam. Valves placed in lower parts of the process system should also be cracked open in order to facilitate the removal of condensate formed when the hot steam is added to a cold process system. All the air must be replaced with steam in order to reach the desired level of sterilisation.

When the system has reached the predetermined steam pressure and the correct sterilisation temperature is obtained in the coldest part of the process system, the sterilisation is

continued for the time period stated by the filter manufacturer. At the same time the differential pressure across the filter must not increase to a level greater than the value recommended by the manufacturer. After reaching the sterilisation temperature, normally in the region of 121–125 °C, sterilisation often requires a total time of approximately 20 min. After this time the steam is disconnected and the total pressure of the system is maintained by the addition of compressed air. The addition of pressurised air has two functions, to maintain the differential pressure across the filter system at a safe level, and to lower the total temperature of the filter and the filter system more rapidly.

Sometimes it is possible to flush the filter with water after the temperature has reached a lower and more suitable level. If the steam-sterilised filter is too hot the filter must not be flushed with cold water or even cold product since such treatment might be harmful to the filter.

After flushing the filter it must be integrity tested in order to ensure that the filter medium or its mounting device has not been damaged during the steam sterilisation. If the filter is manufactured from a hydrophilic filter medium, i.e., a filter medium used for filtering aqueous solutions, the bubble point

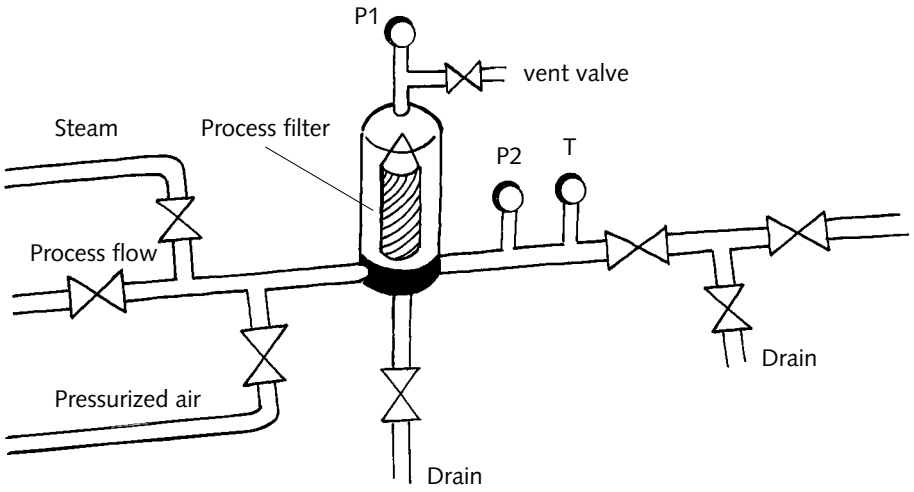

Figure 35. Steam sterilisation of a single filter system.

test or the diffusion test is the normal and traditional integrity test method of choice. After steam sterilisation of filters intended for gas filtration the diffusion test might be used after wetting the filter with for instance isopropyl alcohol or by using the water intrusion test.

After passing the integrity test the fluid to be filtered is added. If the product cannot be exposed to a filter in a wet state the filter must be dried prior to use. A wet filter can be dried quite easily by allowing pressurised air to pass through the filter for some time before use.

9.4.4 Steam Sterilisation of a Larger Process System

A process system has in fact the same basic features as the single filter system discussed above, however the total system is made somewhat larger, for instance by connecting the filter system to a tank and other process components, Figure 36. The tank itself may be equipped with some kind of ventilation system, for instance a vent filter, used in order to protect the interior of the system from contaminants (i.e. particles) in the surrounding air.

Such a process system demands not only that the product filter is sterilised with steam, but also all other components, including the vent filter on the tank. The same basic principle applies for this type of system as for the single filter system mentioned above. Steam is successively added to the total system, at the same time as strict control is kept on the differential pressure across the product filter and the vent filter on the tank. All valves placed in the system must be cracked open, allowing both air and condensate formed during the process to be drained out.

In many cases it is not sufficient to just crack open valves in order to eliminate the condensate formed. In such cases so-called condensate traps must be installed. A condensate trap is generally placed at locations in the process system where condensate is formed and held. A condensate trap works in such a way that it is closed when no condensate is present, i.e., when only dry steam exists in the pipes and other process locations. In the lower parts of the system the steam trap will allow liquid water, i.e., condensate, to leave the system. After all condensate has left the traps they are closed and will be retained in a closed state as long as no condensate is present.

When steam sterilisation is performed on a larger process system it is of the utmost importance to decide where in the system the sterilisation temperature will be obtained last. This point in the system will decide when the sterilisation time is to start. After running the sterilisation process for the predetermined time, i.e., when the required temperature in all parts of the system has been reached and held for a pre-determined time, the steam is successively shut off. In practice this means that the flow of steam is decreased at the same time as pressurised air is added to the system in order to retain the total pressure in the process system. The added pressurised air will keep the differential pressure over the filter at a safe level during the entire cooling phase of the system. A larger process system takes longer to reach a suitable temperature than a smaller system.

After final flushing with water or with product a filter integrity test is performed. An integrity test of the vent filter may be performed using the diffusion test (after wetting the filter with a suitable alcohol-based wetting liquid) or the water intrusion test (where water is used). When using the diffusion test it is important to remember that the product to be processed later in the process system will be mixed with the alcohol. If this is unacceptable the tank must be thoroughly drained and dried before use. In this case the water intrusion test is often much more suitable.

Independent of whether CIP washing, hot water sanitation or steam sterilisation is used, it is important that all parts of the entire process system are exposed to treatment by the method used. Air pockets must always be avoided. The filter housing and other large process parts must therefore have some type of venting device, for instance a valve, at the highest location of the system and this must be cracked open in order to allow the cleaning and (or) sterilisation medium to have access to all surfaces of the system.

When sterilising a filter and/or a filter system, much care must be taken that the filter material and all the different components that together build up the entire filter cartridge, can be exposed to different types of shocks, including thermal, mechanical, chemical and physical. These types of interferences can be obtained due to too high temperature or too

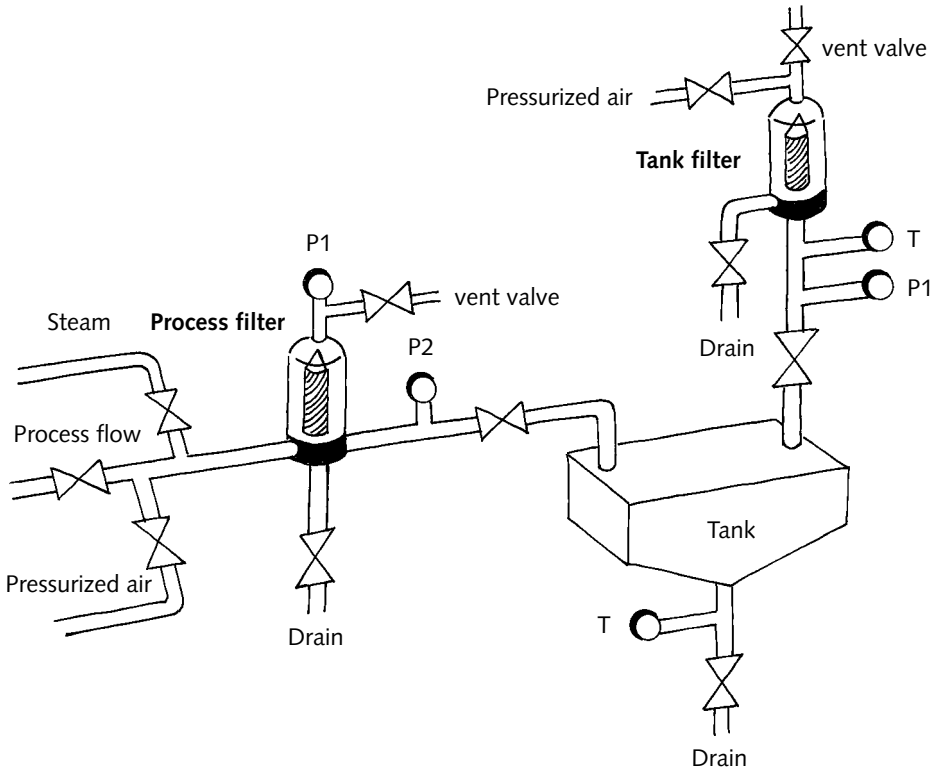

Figure 36. Steam sterilisation of a larger process system, including a filter used for the ventilation of a vessel.

high differential pressure and also due to the fact that the construction materials of the filter may be degraded or disintegrated. It is important to follow the guidelines presented by the filter manufacturer.

9.5 Continuous or Batch Processes

The basis principle when choosing a filter or filter system may differ quite a lot depending on whether the process to be performed is a continuous or a batch process. When utilising a continuous process system the major desire is generally to filter the same process fluid, often at a continuous flow rate and pressure for an extended period of time, for instance up to some 6 to 12 months. The interest when choosing a filter

for this type of application is focused on the longest service life possible for the filter cartridges. From a practical point of view this means that the filtration process does not have to be stopped in order to change, or even in some cases clean, the filter cartridges other than on planned occasions.

The uninterrupted service life of such a continuously running filter system may be maximised by either choosing an over-dimensioning of the filter area of the system or by using what is traditionally called a duplex filter system.

9.5.1 Over-dimensioning a Filter System

When choosing an over-dimensioned surface area in a filter system the most important and critical factor to take into account is the overall compatibility of the filter media and the various sanitising and sterilisation methods to be used.

9.5.2 Duplex System

A duplex system comprises two or more filter housings connected in parallel, in order to gain a larger total surface area of the filter together with the possibility of exchanging filter cartridges or cleaning the various filters while a flow is still maintained through one filter housing in the system.

One way to use the duplex system is to dimension it in such a way that both filter housings together can deal with the total flow needed, see Fig. 37.

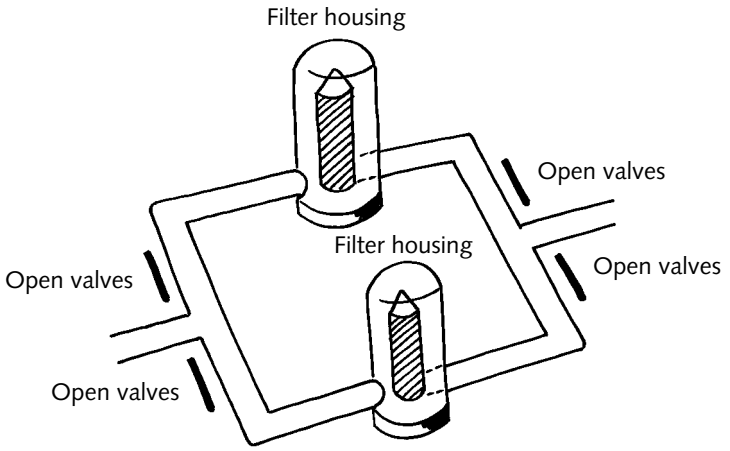

Figure 37. A duplex system in which both filters together will be able to deal with the entire flow.

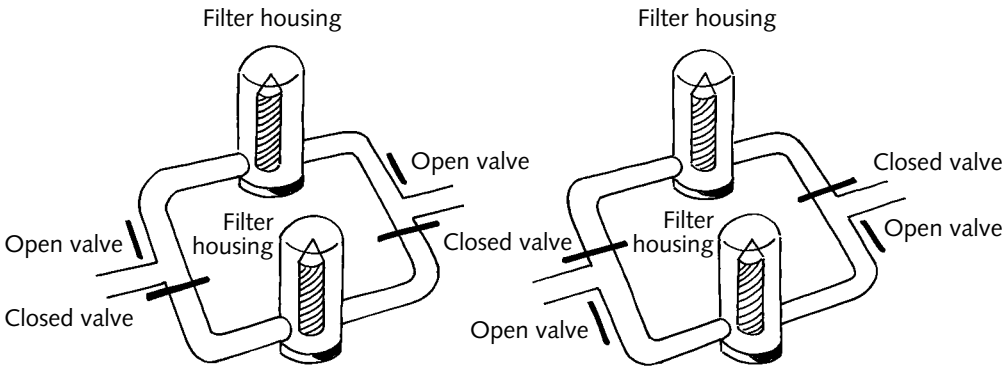

Figure 38. A duplex system in which each of the filters will be able to deal with the entire flow.

When contaminants present in the flow partially block the filter, a pressure drop builds up across the filter necessitating either exchange or cleaning of the filter cartridges. One of the filter housings in the duplex system is isolated and the total process flow is thereby forced to pass through the second and only filter housing then available in the filter system. This filter housing is however not dimensioned in order to deal with the total process flow alone, but normally this does not have a critical impact during the quite short time while the filter cartridges are exchanged or cleaned. The filter housing that is isolated and not in use is opened, the cartridges are removed and exchanged by fresh ones or regenerated, the filter housing is then sealed and brought into operation again. The same procedure is repeated on the second filter housing and when this part of the total system is reconditioned also, both filters are brought into use.

Another way to use a duplex filter system is to dimension each of the two filter housings in such a manner that each filter alone has the capacity of handling the entire flow, see Fig. 38.

This type of duplex system can be utilised in two ways. Sometimes, one of the filter housings is always kept in a stand-by mode, i.e., not in service. When enough contaminants have been collected by the filter in active service, the valves are shifted in order to let the filter housing previously kept on

stand-by be taken into active service. The part of the filter system taken out of service is then regenerated, either by exchanging the used cartridges for new ones or by cleaning the used cartridges. After the reconditioning of this isolated filter housing, it is placed on stand-by.

Both of the presented duplex systems have advantages and drawbacks. When using a duplex filter system in order to filter fluids containing varying amounts of contaminants, it may be difficult to decide when one of the filter housings is to be isolated and the other filter is to take care of the entire flow. The most critical conditions may arise when the filter taking care of the entire flow is blocked prematurely due to an extremely high concentration of particles, i.e., before the second filter is regenerated and put back into active service life. This fact must always be balanced against the cost of a larger filter housing, that normally is very much higher for a filter system in which one filter housing is always kept in a stand-by mode.

9.6 Conclusion

Sanitation and sterilisation is used in order to kill present and collected organisms. Since this type of treatment in most cases also affects all other biological and temperature sensitive material collected and trapped by the filter, the treated filter system will become microbiologically cleaner. The traditional methods used for sanitation and sterilisation are chemical treatment, hot water sanitation and steam sterilisation.

10 Filter Testing

10.1 Introduction

Two general methods exist for the evaluation of filters and filter media, destructive and non-destructive. As the name implies, a destructive test method or challenge test is one in which the filter will be challenged with contaminants. In practice, this means that the filter after such a test is unusable. This is one reason why filter manufacturers and filter users also require a non-destructive test method, in order not to negatively affect a filter prior to use. The filter user must also, for safety reasons, have the possibility to decide whether a filter used in active service is working according to the specifications stated by the filter manufacturer. This is the underlying reason why non-destructive test methods have been developed and are used. Non-destructive test methods are normally called integrity-test methods or just integrity test.

The demands of an integrity-test method are very strict. The first demand is also an absolute one that the non-destructive test must be correlated to a destructive test which is traditionally performed by the manufacturer. If such a correlation between the destructive and the non-destructive tests is not available, the result and use of the integrity test are totally worthless!

10.2 Integrity Testing

First it must be stated that it is, from a practical point of view, only possible to test the integrity of true membrane-type filters and moreover only critical filters, i.e., membrane filters intended for the removal of microorganisms (filters for sterile applications or other related applications, for example filters used for microbiological reduction). In general only filters with removing ratings below 1 µm and finer can be integrity tested and also filters based on polymeric filter media. Generally, the following principal demands are required of an integrity test-method:

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- The integrity test method must be totally non-destructive, i.e., the filter must not be influenced, negatively or positively, during the test, making it possible for the tested filter to be used in active service after testing.
- The method must be relatively rapid and quite simple to perform.
- The result from the test must be extremely well defined, i.e. there should not be a need for a more or less subjective judgement of the analytical result in order to determine whether the filter has retained its integrity or not.
- The integrity test must always be closely correlated to a destructive test in which the function of the filter is studied in its correct environment, i.e. during challenge with microorganisms.
- The integrity test method itself must be performed in such a way that the sterility of the total process system, obtained for instance during a steam sterilisation, must not be interfered with in any way in order to perform the integrity test.

In branches of industry with high staff fluctuation, problems of performing such integrity tests might occur. Such problems might exist due to the lack of time to fully train new personnel to perform integrity testing of various filter systems. This is one of the reasons why filter users in some cases also require integrity test equipment that is easy, secure and reliable to use, even by untrained personnel. Much of the filter testing equipment available has been developed in order to eliminate or minimise problems of this kind.

Different types of equipment for integrity testing exist, ranging from totally manual systems to fully automatic ones. Manual integrity test equipment puts a higher demand on the operator, both in respect of performing the test and in the translation of the test result. That level of demand does not exist to the same extent for automatic integrity test equipment. Automatic equipment is often intended to perform the same test time after time, without changing the various test parameters. In many cases these automatic tests also have the advantage of producing a printout of both test parameters

and test results. These printouts can be added to the documentation collected together in the batch protocols.

Much of the equipment used for more or less automatic testing of integrity of filters must however be checked and calibrated on a regular basis (generally once every year) in order to ensure its reliability.

10.3 Principles of Integrity Testing

When testing critical membrane filters the bubble point test, the diffusion test and the water intrusion test are normally used. The bubble point test was the first method to be developed and has been the basis for the development of other and more modern integrity test methods.

The bubble point test and the diffusion test have much in common since they are both based on the principle that the filter medium to be tested must first be wetted with a suitable wetting liquid before being placed in a filter holder in a test rig. The test rig can either be a separate filter housing or the actual filter housing in which the filter is in active service. The filter medium itself can also be located in the filter housing and then be wetted in situ. A pre-determined gas pressure is then applied on the upstream side of the filter. After applying this gas pressure, either the occurrence of pure gas flow (the bubble point test method) or the measurement of how much gas is leaving the filter system by diffusion through the wetted filter material (the diffusion method) is studied.

Figure 39 shows schematically what takes place in a wetted filter medium that is exposed to a continuously increasing gas pressure. On the *y*-axis of this diagram the amount of gas leaving the filter system at its downstream side devided by the pressure is plotted in relation to the increase in gas pressure that occurs on the upstream side of the filter.

When the pressure applied to the wet filter is increased, the diffusion flow across the filter will increase linearly. By dividing the diffusion flow by the pressure a straight line nearly parallel to the *x*-axis is obtained.

The diffusion gas flow Q divided by the gas pressure P (i.e., Q/P) changes when the gas pressure on one side of the filter medium increases. At very low gas pressures, position 1, the pressurised gas will diffuse quite slowly through the wetted

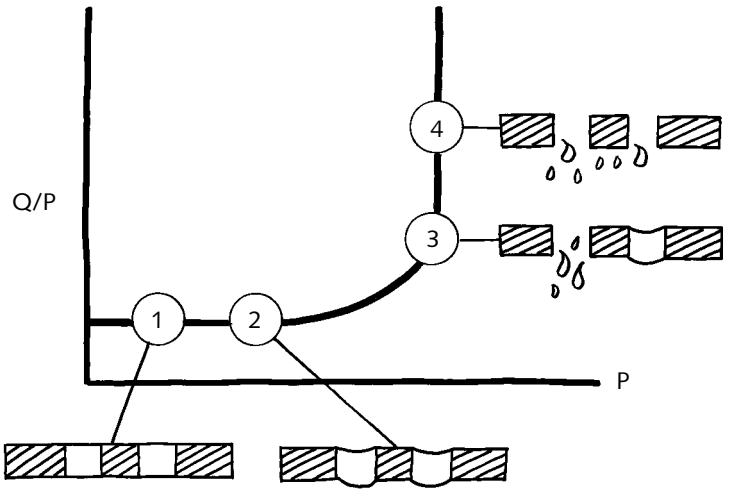

Figure 39. The traditional way of showing what is happening when exposing a wetted filter medium to a pressurised gas. The figure shows the flow of gas leaving a filter system divided by the pressure of the gas as a function of the gas pressure that is used for the pressurization of the filter.

filter medium. The gas diffusion will increase linearly as the gas pressure on the upstream side of the filter increases (follow the parallel straight line to position 2). At a certain gas pressure the force affecting the wetting liquid in the pores will become greater and greater and finally the liquid content of the largest pore will be forced out of the filter.

The force from the pressurised gas applied to the wetting liquid in the pores is dependent on several factors, for instance the type of gas used to pressurise the wet filter, the material from which the filter is manufactured, the liquid that is used to wet the pores of the filter medium and finally also the temperature at which the integrity test is performed.

In position 3 the forces applied by the pressurised gas have become large enough to force the liquid content of the largest pore to be expelled out of the pore and thereby open up the pore for a massive gas flow. The pressure applied by the gas has thereby forced the liquid content out of the largest pore and replaced it with gas. The pressure at position 3 is often called the bubble point of the filter medium. The term bubble

point comes from the fact that bubbles are formed when this test is performed, with the tested filter placed in a container containing the wetting liquid.

If the pressure across the filter is further increased, the second largest pore will be emptied of its liquid content, i.e., the largest pore and thereafter the next largest pore etc. will be consecutively emptied of their liquid content. This is shown by the dramatic change in the appearance of the curve that continues straight upwards. The gas flow at position 4 has a totally different character than the gas flow at positions 1 and 2. Whereas at positions 1 and 2 the flow of gas through the filter is due to diffusion, the gas flow at position 4 is due to a pure and massive gas flow through all the opened pores in the previously wet filter medium.

10.3.1 The Bubble Point Test

The purpose of using the bubble point test is to study and investigate at what pressure the wetting liquid content of the largest pore in a filter medium is forced out and replaced by gas. The bubble point test is schematically illustrated in Fig. 40.

The filter to be tested is wetted with a suitable wetting liquid and placed in the filter holder. A pressurised gas is then applied to the system and the gas pressure is successively increased until a steady stream of bubbles is observed on the downstream and clean side of the filter. The bubble point pressure is registered at this point for each individual filter tested. The same filters are thereafter challenged with microorganisms in a destructive test where the ability of the filters for quantitative removal of the predetermined microorganisms is studied. The two tests, i.e., the bubble point test and the microbiological challenge test, are then correlated and the minimum bubble point pressure corresponding to the desired microbiological cleanliness is used as a guideline value for the test parameters supplied to the filter user, to be used in integrity tests prior to the use of the filter. In many cases the filter manufacturer has incorporated some safety margin in these supplied test values.

The filter user applies the bubble point method in a similar way. The filter to be tested before active service is wetted with a suitable liquid and then placed in its holder. A certain pre-

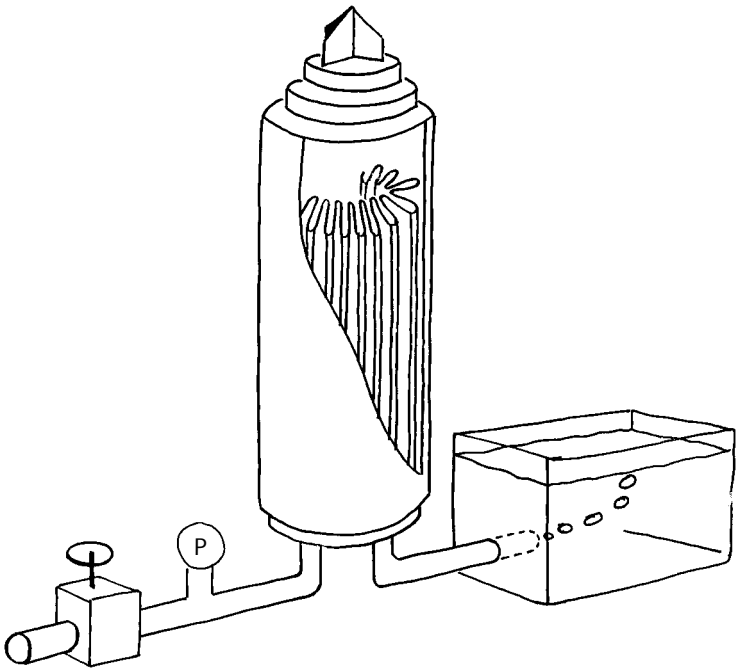

Figure 40. The bubble point test.

determined gas pressure supplied by the manufacturer is applied on the upstream side of the filter. This pressure has been established by the filter manufacturer and is based on a correlation between challenge tests and bubble point tests performed during the manufacture of the filter. If a steady stream of bubbles is observed at this test pressure, the bubble point of the filter has been exceeded and the integrity test shows a negative result. If, however, a steady stream of bubbles is not observed, the integrity test indicates a positive result, i. e., the tested filter has retained its integrity and thereby its function to eliminate the desired microorganisms.

The original bubble point test is in fact a subjective test method due to the fact that a judgement must be made as to whether the eventual gas flow through the filter is ›a steady stream of bubbles‹. More modern test equipment has been developed for performing a bubble point test that is not based on a subjective judgement, equipped with software for auto-

matic testing. Such an integrity test unit results in safer testing and also the possibility of performing the bubble point test totally independent of the skill of the operator.

10.3.2 Diffusion Test

As previously mentioned, the second integrity test method, the diffusion test, is a further development based on the general principle of the bubble point test. One of the major reasons for the development of the diffusion test was that the theoretical background of the bubble point test does not fully correspond to reality. Furthermore, the bubble point test is in some cases quite hard to correlate with the results obtained in the following destructive test, i.e., the test in which the filter is subjected to microbiological challenge. From a practical point of view the diffusion test can be performed in two distinct ways, either based on the volumetric measurement of the gas diffusing through the filter, the so-called volumetric test, or based on the indirect measurement of the diffusion flow of gas through the filter by analysing the pressure decrease on the isolated and pressurised upstream side of the filter, the so-called pressure hold test.

Both the volumetric test and the pressure hold test are based on the total concept of the diffusion test and are both carried out in the same way. The filter medium to be tested is wetted with a suitable wetting liquid after which it is placed in its holder, exactly as is done in a bubble point test. The filter manufacturer supplies a certain test pressure that is much lower than the bubble point pressure of the filter medium thereby incorporating a safety margin. This lower pressure is used in order to obtain only a diffusive flow of gas through the wetted filter medium. The diffusive flow of gas through the wetted filter medium is then measured at the applied test pressure, after which the filters are analysed further by means of destructive challenge tests. The manufacturer thereafter analyses the diffusion flow of the various filters in order to state the maximum diffusion of a filter that can achieve the desired cleanliness. This diffusion value is finally given a safety margin before it is supplied to the filter user in order to be used in an integrity test before active service of the filter.

The filter user utilises the diffusion test by applying the stated gas pressure on the upstream side of a wet filter. As

stated earlier this gas pressure is quite far below the bubble-point pressure of the filter. The pressurised gas will, during the test, diffuse from the upstream and pressurised side of the filter to the downstream side. The amount of gas diffusing through the wet filter can be measured.

10.3.3 The Volumetric Test

The differences between the volumetric version and the pressure hold version of the diffusion test lie in the way in which the volume of gas diffusion is measured. In the volumetric test, often called the Forward Flow Test, the amount of gas passing through the wet filter per unit time is physically collected in a measuring device and measured in a physical way, Fig. 41.

The collection and measurement of the diffused gas is achieved by transporting the gas in a tube into a measuring cylinder that is turned upside down and filled with a liquid, for

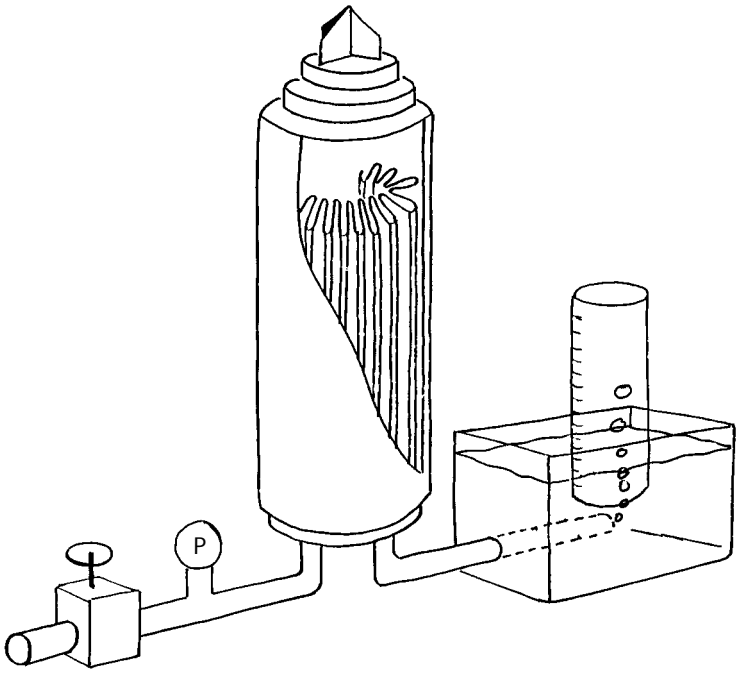

Figure 41. The diffusion test method performed as a volumetric test.

instance the wetting liquid or water. The gas entering the measuring cylinder will replace the liquid and the gas volume can be registered directly on the glass.

This method in its original version demands that measurement is performed on the downstream and clean side of the filter, making the method not totally compliant with the previously stated demands of an ideal integrity test. This is because the sterility obtained by autoclaving prior to the test cannot be retained.

10.3.4 The Pressure-hold Test

In many industrial applications there is an absolute need to test the filter after sanitising and steam sterilisation, which means that it is not useful to destroy the integrity of the process system in order to perform the integrity test. If the integrity of the process system after sterilisation is required, it is more practical to use the second version of the diffusion test, the pressure-hold test or pressure-decay test. The test equipment for this version is quite similar to that for the volumetric version. The only difference between these two tests is that an extra valve is incorporated together with a very sensitive pressure gauge installed prior to the filter to be tested. In this way a certain volume of pressurised gas can be isolated between the wet filter medium and the valve, Fig. 42.

The pressure-hold test is performed by applying a predetermined gas pressure on the upstream side of the wetted filter medium placed in its holder. After some time, in order to reach equilibrium, the main valve is turned off, thereby isolating a certain amount of pressurised gas between the valve and the wet filter medium. The isolated gas has only one possible way to leave the system, unless the shut-off valve is not good enough, namely by diffusion through the wet filter medium. This diffusion pattern is analysed and registered by observing the successively decreasing pressure of the isolated gas volume with time. The pressure decrease registered represents an indirect result of the amount of gas diffusing through the wetted filter.

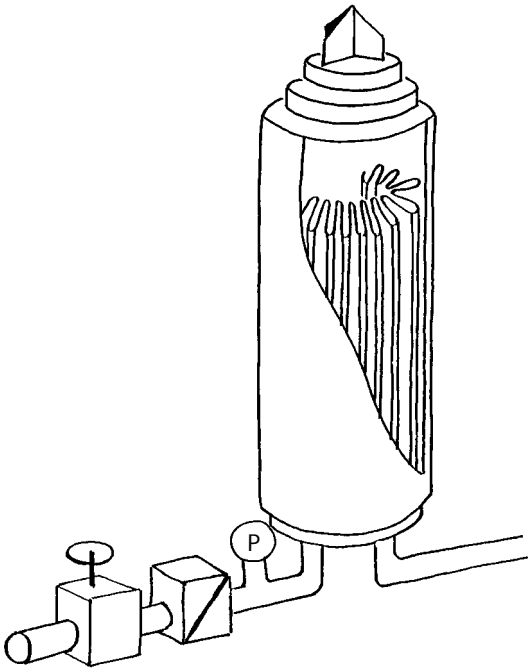

Figure 42. The diffusion test method performed at a pressure hold test.

10.3.5 Which Method to Use?

When integrity testing a disk-filter, i.e., a small circular filter medium, the disk is placed in a filter holder. This holder is in many cases very difficult to seal in a totally gas tight way. If the disk-filter is not totally sealed, it is impossible to use the pressure-hold test since the integrity of the filter cannot be retained due to the continuous leakage of gas through the seal. When integrity testing of disk-filters is to be performed the bubble point test is often better and is the method of choice.

Large surface area filter systems, for instance comprised of a large number of filter cartridges, however, can be difficult to integrity test with the bubble point method. Membrane filters today often have a very high porosity, i.e., large open area, compared to filters developed 10 to 15 years ago. When studying the gas diffusion through wet filters from such filter systems, in some cases the diffusive flow is hard to separate

from the massive flow obtained after reaching the bubble point. In practice this means that a filter system with a large total filter area, for instance a filter cartridge with approximately 1 m^2 filter area and/or larger, will have a diffusion of gas large enough to be observed as a steady stream of bubbles and even more. In this type of system it is very hard to decide if the flow of gas through the wet filter is a result of a bubble point flow or is due to diffusion. This is one of the reasons why the two diffusion test methods are most commonly used for testing filter systems with high surface areas.

Most of the filter manufacturers and suppliers of filters give detailed information and recommendations on the methods to be used for integrity testing of their products. Some manufacturers state that filter cartridges of size 5 inches or larger should be testing according to the diffusion test method, whereas smaller filters should be tested with the bubble point method.

10.3.6 The Water Intrusion Test

As stated earlier, filters can be either hydrophilic (water attracting) or hydrophobic (water repelling). When filtering aqueous solutions it is natural to use a hydrophilic filter, whereas when filtering gases either hydrophilic or hydrophobic filters can be used. When a gas is filtered not only mechanical retention and adsorption due to inertial impaction is used but also when gas passes through a dry filter adsorption due to diffusion occurs, which increases the efficiency of the filter by up to ten-fold. In order to fully obtain this extremely high filtration efficiency, the filter medium must always be totally dry, i.e., it must not be wetted by any liquid. This is the underlying reason why hydrophobic filters in most cases are chosen when gases are to be filtered.

When a hydrophobic filter is to be wetted prior to integrity testing of the filter, water cannot be used. Instead some type of hydrophobic liquid, such as for instance ethyl alcohol or propyl alcohol or a mixture of these alcohols with water, must be used. However, there is a great risk that the wetting liquid will come into contact with surfaces of the process equipment and thereby the product in the process equipment during this procedure. This is why another method for integrity testing of hydrophobic filters has been developed, the so-called Wa-

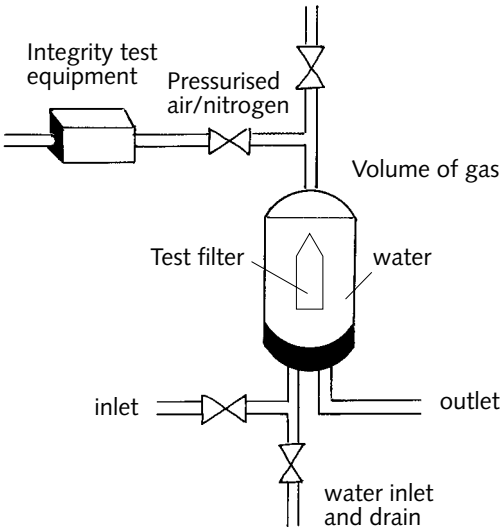

Figure 43. The water intrusion test.

ter Intrusion Test. The water intrusion test is based on the fact that a hydrophobic filter will not be wetted by water.

The water intrusion test is based on the fact that the hydrophobic filter to be tested will not allow passage of water when water is added to the upstream side of a dry hydrophobic filter in a filter system filled with water, Fig. 43. Even when the water added to the upstream side of the filter is subjected to pressure, the hydrophobic filter will hinder the water from entering the pores until the pressure of the water surrounding the filter has increased to the level that it can enter the pores. If the pressure is below the intrusion pressure however, a small but still measurable flow of water will enter into the pores of the filter. If any larger pores are present in the filter medium, they will affect the flow of liquid through the filter in such a way that the larger the pores the larger the water flow through the filter.

The water intrusion test is performed by filling the upstream side of the filter system and pressurising the upstream side (the test pressure is supplied by the manufacturer). The applied pressure will lower the water level in the filter system since the air in the outer perimeter of the filter pores is forced

out and replaced with water. The change in the upstream volume obtained during a stabilising period, in general 10 min, is needed in order to measure the very small changes of flow later when the water passes into the filter material.

After the stabilisation period, during which the temperature also is stabilised, has ended, the water flow into the filter can be measured. This flow is registered indirectly as the amount of gas needed to be added in order to retain the pressure in the filter system at a constant level. After the test has been completed the water is drained away from the filter system and the filter is ready for use. Sometimes it may be necessary to dry the filter system before use by flushing the filter with pressurised air.

The water intrusion test has many advantages. The test is suitable for in-situ testing, i.e., testing a filter cartridge placed in a process system where it is to be used for service later, and in process systems that have been sterilised, since no manipulation of the system is needed on the downstream side of the filter. Other advantages are that alcohol solutions are not used during the test and that the system generally does not need to be dried before going into active service.

10.4 General Aspects of Integrity Testing of Membrane Filters

The bubble point test is a relatively rapid test method requiring short stabilising times. The test can be quite misleading when used on larger filter systems (i.e. filters with larger surface areas) since it may be difficult to decide if the gas flow leaving the filter system is due to diffusion through the wet filter or due to bubble point flow. Performing a manual bubble point test requires that the downstream side of the filter system is opened up, which in turn means that eventually the sterility is lost.

For the manual bubble point test the following is valid:

- The pressure on the wet filter should be increased slowly in a stepwise manner.
- The pressure should be allowed to stabilise after each increase. If the pressure is increased too rapidly, there is a possibility that the bubble point- pressure will be exceeded. If this situation occurs there is no possibility of decreasing the pressure in order to start again since the filter must be re-wetted before performing a new test.

- Connections downstream of the filter should be kept to a minimum.
- The entire filter system is controlled in order to observe eventual leakage.
- It must be clearly defined what is meant by the term bulk-flow of gas.

The most commonly found error when performing a manual bubble point test is that the operator believes that he or she is observing a bulk-flow of gas when in fact the flow through the wet filter is the result of diffusion. In practice this means that it is very hard to identify the bubble point itself. In addition this means that there is a risk that a filter might be discarded in spite of the fact that it retains its integrity. Traditionally the bubble point is to be indicated as a steady stream of bubbles and not by the first bubble observed.

- The bubble point test should be performed by trained and skilled operators, especially if it is performed manually.
- The upstream volume of a test system should be kept as small as possible in order to avoid a too-long stabilisation time during the stepwise increase of the pressure on the filter.
- The test temperature should be kept within pre-determined levels.

The diffusion test has been developed in order to offer increased sensitivity for filter systems with large filter areas. The test is based on the objective quantitative measurement of the gas passing the wet filter due to diffusion. The method is however quite sensitive to fluctuations in temperature. When the diffusion test is to be performed as a manual test it is necessary for the filter system to be opened downstream of the filter, which in practice means that the sterility will eventually be broken.

The following are requirements for the diffusion test:

- The number of downstream connections (valves etc.) and the volume of the process system connected to the downstream side of the filter should be minimised.
- The entire system should, during the test, be controlled for leakage.

- The volume of the upstream side of the filter must be kept to a minimum. It can be difficult to obtain a stable system if the volume is too large.
- The test temperature must be stable during the entire test.
- The stabilisation time after applying the pressure to the filter should be long enough, generally the system must be stabilised for 10 min.
- The test should be performed at a validated pressure, stated by the filter manufacturer and at a sufficiently safe distance from the bubble point pressure of the filter material.

The pressure-hold test is relatively simple to perform and no manipulation of the downstream side of the filter is needed. The sensitivity of the test will decrease the higher the upstream volume of the test system. The accuracy of the test is limited by the accuracy of the manometer used. The state at which the test is performed, i.e., temperature and the gas used for pressurisation of the filter, should be clearly defined in order to be able to use the integrity test values supplied by the filter manufacturer.

The following are requirements for the pressure-hold test:

- There must not be any leakage in the test system.
- The upstream volume of the filter system must be measured or taken into account in order to get a correct maximum pressure drop, the upstream volume should be kept as low as possible.
- The downstream side of the filter should be ventilated towards the atmosphere.
- The stabilisation time before starting the test should be long enough. The larger the upstream volume, the longer the stabilisation time.
- A pressure-gauge (manometer) with sufficient sensitivity should be used in order to record the pressure decrease during the test.
- If the pressure decreases relatively slowly the test time must be extended. This situation is especially valid if a pressure gauge with too low sensitivity has been chosen.
- The test should be performed at a specified temperature and this temperature must be maintained during the entire test time. The smaller the upstream volume, the more sensitive the test will be to fluctuations in temperature.

The water intrusion test has been developed in order to offer a simple, validated and practical method to be used for in-situ testing of hydrophobic sterile filters for air and other gases. The method has been developed mainly to replace the diffusion test method in which hydrophobic filters must be wet with a suitable liquid, often alcohol or alcohol water mixtures, which is not always practical for all applications. The parameters during the water intrusion test, especially gas pressure (often compressed air), should be well-defined in order to obtain useful integrity test values.

The following are requirements for the water intrusion test:

- There must not be any leakage in the system.
- The water temperature should be equal to the temperature of the surrounding air.
- The surrounding temperature should not be changed or altered during the test and strong air movements in the neighborhood of the test system should be limited.
- The stabilisation time after pressurising of a filter should be sufficiently long, approximately 10–20 min.

10.5 Conclusion

The filter manufacturer defines the function of the filter products by performing tests on the filters. Destructive as well as non-destructive tests are available. A filter that has passed the destructive test can naturally not be used in a process. The demands on a non-destructive test are that it must be correlated to a destructive test.

11 Validation of Filters

11.1 Introduction

Validation of a filter briefly means that parameters measured and given as a proof of the desired function of the filter must be correlated to how the filter works in reality. To state that the sterile filter is validated therefore means that the function of the filter has been checked with different and correlated challenge tests and that these parameters are available for both the filter manufacturer and the filter user in order to perform an integrity test.

Validation has two general purposes. Firstly, it should be a tool for the manufacturer to gain control of the products to be delivered to the user. Secondly, validation aims to give the filter user a tool to ensure that the filter, when used in active service, will fulfil the demands of cleanliness stated by the manufacturer.

Validation only aims towards confirmation of the possibility of the filter giving a certain cleanliness level in regard to microorganisms used during the challenge test. Basic validation does not incorporate anything else. In the written documentation supplied by the filter manufacturer, however, other test results might be present, for instance control of extractable material and also control of pyrogens.

11.2 Testing and Validation

A microfiltration filter is generally intended for the elimination of harmful particles from a fluid. At the same time the filter system must be inert, i.e., the filter system should not add to or remove anything from the fluid, even though at first glance it may not be regarded as a contaminant. This is why all the different parts in a filter system in contact with the process flow must be tested. The filter manufacturer performs such tests and sometimes these tests are also performed by the filter user. Particle release, extractable substances, physical as

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well as chemical compatibility, together with absorption of critical substances are some of the factors that are normally investigated. Toxicity tests, bacterial challenge tests and physical integrity tests are other examples of important tests that are performed.

11.2.1 Particle Release

All filter materials release particles, some filters more and others less. Particle release is strongly dependent on external influence on the filter during use, and therefore particle release is analysed for instance at different flow rates, differential pressures and temperatures. From the filter user's point of view it is important that these tests are performed under conditions similar to the real process.

11.2.2 Extractable Substances

Extractable substances, often called extractables for short, can be extracted from a filter and (or) a filter system. USP (United States Pharmacopoeia) publishes a number of tests, guidelines and demands in regard to extractables, mostly from plastic materials. The source of extractable substances is often different surfactants or wetting agents that are added to the filter material during manufacture. Other sources might be the support material of a filter, for instance different materials used in the manufacture of cages and other hardware.

As a rule of thumb the filter manufacturer must supply data on the amount and type of extractable substance, which can be used for the identification of the eventual presence of these substances in a finished product or in a process flow. Extractable substances can be classified as toxic or non-toxic. If the filter studied is to be used within the pharmaceutical industry all extractables must be non-toxic. It is the responsibility of the filter user to show that the product only contains acceptable levels of extractable substances.

In the same way as for particle release discussed above, flow rate, temperature and differential pressure play a vital role in the release of extractable substances.

11.2.3 Chemical Compatibility

In order to evaluate the chemical compatibility of a filter, it is important not only to analyse the filter material itself, but also to incorporate all other parts that together build up the entire filter system. In practice this means that the filter material, the

outer and inner cages, together with o-ring seals and other sealing material that are used when installing a filter in a filter housing, are to be taken into account during this investigation.

In many cases the filter manufacturer has studied the impact of individual chemical substances on the filter, whereas the filter user often has a mixture of several chemical components to be filtered. This means that it is not always possible to draw a simple conclusion from the tables often supplied by the filter manufacturer. The best way is to perform compatibility tests on the product or the process flow to be filtered.

11.2.4 Adsorption

Adsorption in this context means that different components in a process flow will adhere to the surface of the filter material. This might result in a situation in which the quality of the filtered product does not correspond to the quality of the product before filtration. Adsorption is, amongst other things, dependent on the flow rate of the fluid through the filter, the concentration of the component that might be adsorbed, the temperature, pH-value and ionic strength of the fluid.

11.2.5 Physical Compatibility

Physical compatibility includes both thermal compatibility and compatibility due to variations in the pressure of the process system.

Thermal compatibility is mainly due to the overall sensitive of the filter to cope with, variations in temperature. Differences in temperature not only incorporate the differences between a filter at room temperature and a filter exposed to steam at 121 °C (sterilisation temperature) but also incorporate fluctuations and repeated changes in temperature, i.e., if the filter is for example first heated, cooled down and then heated again. Such variations of temperature can have an impact on a filter from a chemical as well as a physical point of view.

Variations in the pressure applied to a filter (i.e., the differential pressure), especially at elevated temperatures can have a major impact on the integrity of a filter and in some cases even totally destroy the filter during use.

11.2.6 Toxicity Test

Filters should not be manufactured or designed from materials that in one way or another might be toxic. This is why filter manufacturers perform toxicity tests and also give extensive details on the test results. Toxicity tests are especially vital within the pharmaceutical industry, but are also important in the food and beverage industry due to the possible influence on man.

11.2.7 Bacterial Challenge Tests

Filters that are subjected to a bacterial challenge test (a destructive test) cannot be used in active service after testing. When testing sterile filters the test organism *Brevundimonas diminuta* ATCC no 19146 (previously named *Pseudomonas diminuta*) is used. Bacterial challenge tests can also be used in order to validate a filtration process for a given product.

11.2.8 Physical Integrity Tests

Physical integrity tests are also known as non-destructive tests. In order to ensure the safety and reliability of a filter product there must be a correlation between the physical integrity tests and the destructive bacterial challenge tests.

11.2.9 Practical Validation

In practice a validation is performed in three steps: Firstly, a non-destructive test is performed (integrity testing), secondly these filters are subjected to destructive testing (challenge tests) with microorganisms and, finally, a correlation of these two tests must be performed.

After performing the non-destructive test (either the bubble point, diffusion or water intrusion test) the filter is exposed to a destructive test (the so-called challenge test) using a suitable microorganism. This challenge test answers the question of whether the filter is supplying the desired removal capacity. Finally these two tests are linked together, resulting in a correlation.

An example of validation of a filter can have the following form: the filter manufacturer is interested in validation of a newly developed filter material which is intended for sterile filtration. A statistical number of filter cartridges are manufactured and each filter is individually labelled in order to be followed through the validation process. The diffusion test is chosen as the non-destructive integrity test. In order to gain a reliable safety that only diffusive flow through the wetted

filter is measured, the test pressure for the integrity test is traditionally chosen as approximately 80% of the bubble point pressure.

The first step in the validation process includes the measurement of the diffusive gas flow through each individual wetted filter, studied at a predetermined test pressure. In the following step each individual filter is placed in a test equipment and subjected to a microbiological challenge test. The challenging microorganisms, in this case *Brevundimonas diminuta*, due to the fact that the filters are to be used for sterile filtration, are dispersed in a suitable buffer and then allowed to come into contact with the filter during a filtration cycle. During this challenge test, samples from the fluid passing through the filters are collected and thereafter analysed for the presence of test organisms.

Finally the data from both tests are combined and the diffusion value for each individual filter is plotted in a diagram that also shows whether or not the filter is giving a sterile result, Fig. 44.

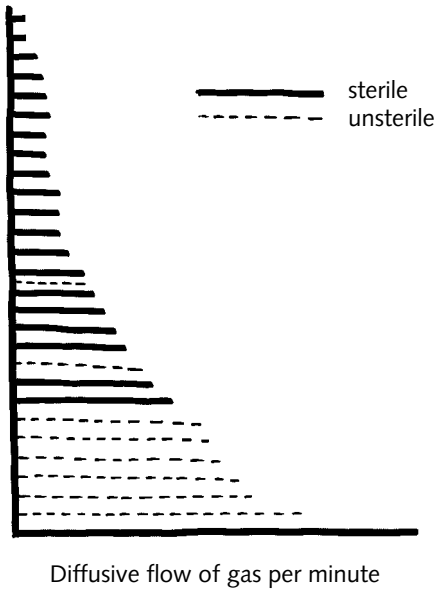

Figure 44. The correlation between the destructive and the non-destructive tests performed on a filter.

The example given in this figure shows that some filter cartridges give non-sterility if the diffusion value exceeds a certain limit. With the analytical result obtained in these tests, it is also easy to decide at which maximum gas diffusion non-sterility will be observed. If the tests are performed on a large enough number of filter cartridges of the same type and size, the results from the integrity test (the diffusion value) will become statistically safer. In practice the values presented by the filter manufacturer and to be used by the filter user often incorporate some sort of safety margin.

The example shown in Fig. 45 shows that there are filters that might result in sterility despite the fact that the diffusion value far exceeds the maximum value stated. This is why it cannot be stated that a filter with a gas diffusion value exceeding the given sterility level must always result in a non-sterile product. The correct way to express this situation is to explain that the use of such a filter has a limited possibility of giving a sterile product.

11.3 Conclusion

Validation can, in simple terms, be explained as an activity used in order to control that a process really performs in the way intended.

12 Sterilisation with Heat

12.1 Introduction

Sterilisation is a process used in order to create a sterile state. The term sterile is defined as the absolute absence of organisms, either by elimination or destruction. Previous chapters in this book discuss the process of microfiltration, which is one method of obtaining a product or process flow free from living organisms. In the following chapters thermal methods will be discussed, methods that cause much damage to the organisms, rendering them unable to survive and develop.

The definition of sterility according to the European Pharmacopoeia is as follows: Sterility is the absence of viable microorganisms. According to the EC GMP, the corresponding definition is: Sterility is the absence of living organisms. The difference between these two definitions might be minute, but there is actually a large difference between microorganisms and organisms.

12.2 Sterilisation

All sterilisation processes, for instance sterilisation with saturated water vapor, follow the principle of probability and thereby also follow an exponential curve. This is why it can never be stated with 100% safety that the product being sterilised is guaranteed to have become sterile. Furthermore, there is also no absolute possibility of controlling this statement, i.e., that sterility has been obtained. This is why most of the Pharmacopoeias press manufacturers of pharmaceutical products to perform and act in such a way that at least in theory, there must not be more than one surviving microorganism per one million sterilised units.

In order to deal with this demand from a practical point of view, the term Sterility Assurance Level (SAL) was introduced, which expresses to what degree of probability a process can give rise to sterility. An SAL value of 10^{-6}, i.e. not

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more than one surviving microorganism per one million sterilised units, is demanded for products that are sterilised in the final and closed container.

12.3 Steam Sterilisation

Proteins are responsible for the genetic information and the enzyme activity in biological cells. The three-dimensional structure of a protein molecule is of vital importance in regard to its function. The action of a protein can be seen as that of a key that fits into other molecules that are to be influenced by the protein. The three-dimensional structure of a protein is in turn dependent on relatively weak hydrogen bonding which holds the molecule together, thereby creating the characteristic three-dimensional appearance of the protein, Figure 45.

When a protein is gently heated the hydrogen bonding is broken, resulting in a change in the protein structure. If this reaction is not continued too far, i.e., if the protein is not heated too much and then cooled down again, the protein molecule can regain its original shape and also its original catalytic function. This type of reaction is called a reversible reaction. However, if more energy in the form of heat is added to the protein, more and more of the hydrogen bonds are broken. Finally the number of hydrogen bonds broken is so large that the protein cannot spontaneously regain its original shape when the temperature is lowered. This reaction is called an irreversible reaction. The irreversible reaction results in an enormous change in the three-dimensional struc-

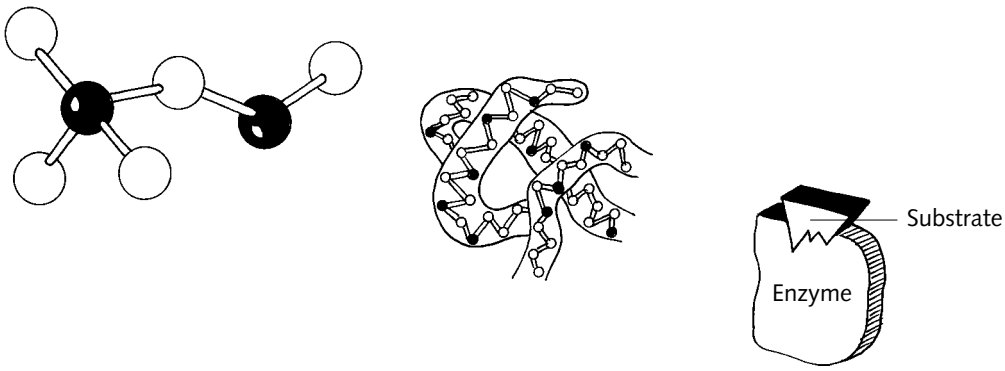

Figure 45. Change in the three-dimensional structure of a protein.

ture of the protein. The protein is said to have denatured and can no longer function as an enzyme or as a carrier of genetic information.

When biological cells have been dehydrated they become even more resistant to heat. This is why spores are even more difficult to affect by addition of heat than vegetative cells.

12.4 Definitions

Several different factors that have an impact on the result during sterilisation with heat exist. The three most important are the *D*-value, the *Z*-value and the F_0-value.

12.4.1 The *D*-Value

The *D*-value is defined as the time needed, at a certain temperature, to reduce the number of microorganisms in a microbiological population by one logarithmic unit, for instance reducing the number of organisms from 100 to 10, Fig. 46. In practice one logarithmic unit corresponds to a 90% reduction in the number of microorganisms. The *D*-value varies for different microorganisms and also for one and the same organism depending on the temperature and on the nature of the surrounding growth medium.

Mathematically the *D*-value can be expressed as:

$$D = \frac{U}{\log N_0 - \log N_U}$$

Where U is the exposure time at a given temperature, N_0 is the number of microorganisms at the start of the exposure time and N_U is the number of microorganisms at the end of the exposure time.

Example: If a 5 min exposure at a temperature of 121 °C has reduced the number of microorganisms from 2×10^5 down to 6×10^3, the *D*-value will be:

$$D = \frac{5}{\log (2 \times 10^5) - \log (6 \times 10^3)}$$

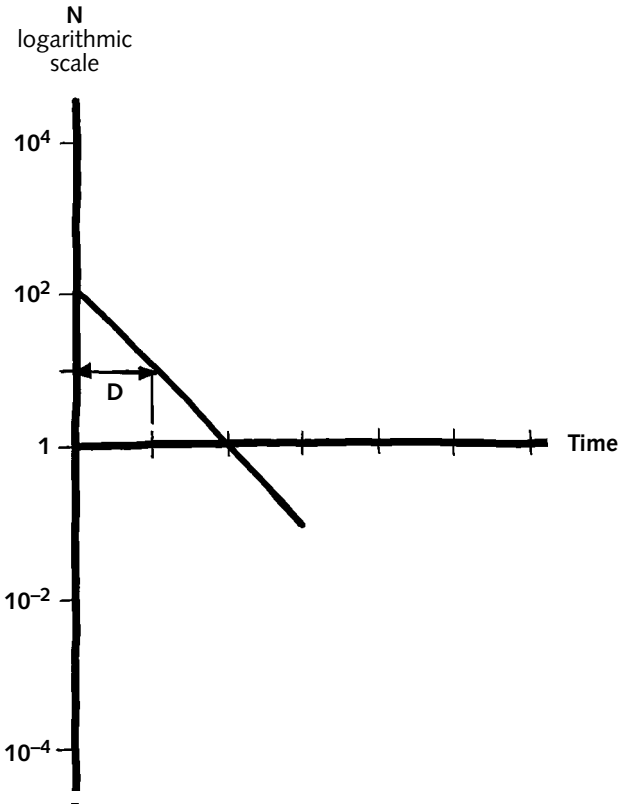

Figure 46. *D*-value.

12.4.2 The *Z*-Value

The *Z*-value is defined as the temperature needed in order to change the *D*-value by one logarithmic unit for a microbiological population, Fig. 47. For practical reasons it is established that heat resistant bacterial spores have a D_{121}-value of 1 min and a *Z*-value of 10 °C when sterilisation is performed with saturated water vapor.

12.4.3 The F_0-Value

The F_0-value describes the killing of microorganisms as the equivalent time in minutes at a temperature of 121 °C for microorganisms having a *Z*-value of 10 °C. This means that sterilisation at 121 °C for 15 min will result in an F_0-value of 15 min.

In order to obtain an approved sterilisation of goods in

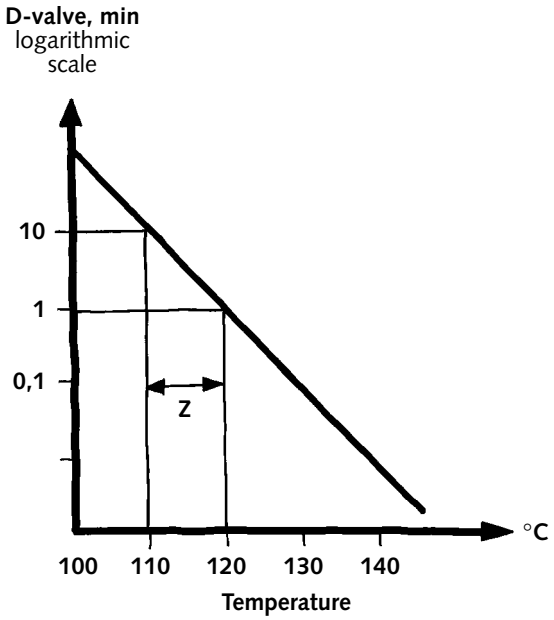

Figure 47. *Z*-value.

their final container an SAL-value below 10^{-6} is desired. An established way to describe an approved sterilisation process is that it has been performed at 121 °C with an exposure time at this temperature of 15 min, i.e. F_0 equals 15 min.

The result from a steam sterilisation process in general depends on the heat resistance of the microorganisms (i.e., their *D*-value), the number of microorganisms present in the population at the start of the process (N_0), often referred to as the bioburden, together with heat exposure (F), i.e., a combination of time and temperature.

When the logarithmic value of the number of living microorganisms (*y*-axis) is plotted as a function of sterilisation time (*x*-axis) a microbiological death curve is obtained, Fig. 48.

This figure shows that the number of microorganisms declines exponentially with time. Since the scale on the *y*-axis is logarithmic, a straight line is obtained. The speed with which the reduction of the microbiological population takes place, is dependent on the *D*-value of the microorganisms. The lower the *D*-value the faster the reduction will take place and the

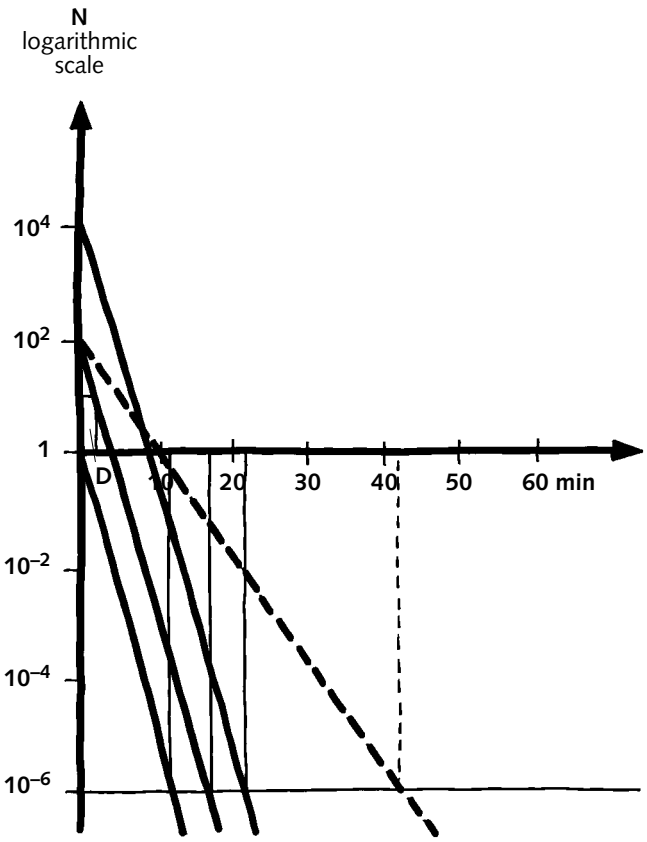

Figure 48. Microbiological death curve.

faster the acceptable SAL-value will be obtained. The *D*-value will vary between different types of microbiological populations. In the same microbiological population the *D*-value will decline with increasing temperature.

This figure also shows that at 100 (log 2) microorganisms per container, the SAL-value of 10^{-6} will be reached after approximately 15 min at the temperature used. If instead 10,000 (log 4) microorganisms are present at the start of the sterilisation, the SAL-value of 10^{-6} will be reached after 254 min exposure time. This example shows how important it is always to start the sterilisation process with a low bio-

burden, i.e., a low number of microorganisms, in order to increase the safety of the process.

The dotted line in the figure shows how the *D*-value affects the sterilisation process. With 100 microorganisms (log 2) per vial, but with a higher *D*-value, the SAL-value of 10^{-6} will be obtained after approximately 40 min.

The F_0-value describes, as previously mentioned, the killing of microorganisms as the equivalent time in minutes at a temperature of 121 °C for microorganisms having a *Z*-value of 10 °C. According to practice, heat resistant bacterial spores have a D_{121}-value of 1 min and a *Z*-value of 10 °C when sterilisation is performed with saturated water vapor. Below some examples illustrating the discussion above are shown.

Example: A microbiological population comprised of 100 microorganisms (log 2) per vessel is to be sterilised at 121 °C. How long is needed for the sterilisation process in order to obtain a SAL value of 10^{-6}?
The following equation is used:
$F_0 = D_{121} \times (\log N_0 - \log N)$,
Where
N = the number of microorganisms after sterilisation
N_0 = the number of microorganisms at the start of the sterilisation (100)
$D_{121} = 1$ (according to practice for heat resistant bacterial spores)
The equation will be as follows:
$F_0 = 1 \times (\log 100 - \log (10^{-6}))$
$F_0 = 1 \times (2 - (-6))$
$F_0 = (2 + 6) = 8$ min
In order to obtain an SAL-value of 10^{-6} the sterilisation process should be performed for a period of 8 min at 121 °C.

12.4.4 Equivalent Time

Products that cannot be exposed to too high temperatures can, in spite of their temperature sensitivity, be sterilised with heat. The sterilisation process is in this case performed at a lower temperature, but as a conclusion of the discussion above, for a longer time period.

Example: The same system is used as in the former example with the exception that the product to be sterilised can only tolerate a temperature of 115 °C. Calculate the sterilisation time that is needed at this decreased sterilisation temperature in order to obtain an F_0-value of 8 min, which is the time needed to obtain an SAL-value of 10^{-6}.
The following equation is used:

$F_0 = F \times 10^{(T\text{-}121)/z}$,
where
F_0 = the time, in minutes, needed at 121 °C (previously calculated to be 8 min)
F = the time, in minutes, at the lower sterilisation temperature
T = the lower sterilisation temperature (115 °C)
Z = 10 °C (according to the practice previously stated)
$8 = F \times 10^{(115-121)/10}$
$F = 32$ min

If lower sterilisation temperatures are chosen the sterilisation process itself must be carried out for a longer time period.

12.4.5 Tailor-made Processes

When sterilisation is performed on heat-sensitive products the microbiological flora of the product to be sterilised are often screened. A number of D-values and Z-values are defined followed by calculation of a good enough sterilisation process with low enough F_0-values.

Example: The following system is to be used: The D_{121} value is 0.4 min and the Z-value is 15 °C during sterilisation with saturated steam at 121 °C. In each vessel that is to be sterilised at 121 °C is a population of 10 microorganisms (log 1). How long must the sterilisation process continue in order to obtain an SAL-value of 10^{-6}?
The following equation is used:
$F_0 = D_{121} \times (\log N_0 - \log N)$,
where
$D_{121} = 0.4$ min

N_0 = the number of microorganisms at the start of the sterilisation (10)
N = the population after sterilisation (10^{-6})
F_0 = the sterilisation time at 121 °C
$F_0 = 0.4 \times (\log 10 - \log 10^{-6})$
$F_0 = 0.4 \times (1 - (-6))$
$F_0 = 0.4 \times 7 = 2.8$ min
The sterilisation can in this case be performed in 2.8 min at 121 °C.
Example: The product in the example above turns out to be temperature sensitive and can only tolerate 112 °C. Calculate the time, in minutes, F that is needed at this decreased temperature in order to obtain an F_0-value of 2.8 at SAL 10^{-6}.
The following equation is used:
$F_0 = F \times 10^{(T - 121)/Z}$,
where
F_0 = the sterilisation time at 121 °C (2.8 min)
F = the sterilisation time at 112 °C (to be calculated)
T = The sterilisation temperature (112 °C)
Z = 15 °C.
$2.8 = F \times 10^{(112-121)/15}$
$2.8 = F \times 10^{-0.6}$
F = 11.2 min

With these calculations it is possible to prove that other time intervals and lower temperatures than those described in the pharmacopoeias and the GMP will actually give a sterile result (SAL = 10^{-6}) taking the practical details into account.

12.5 Conclusion

Sterilisation by heat means that the microorganisms are affected to such a high degree that they can no longer continue to live. The result from a heat sterilisation process depends mostly on the heat resistance of the microorganisms to be treated, the number of microorganisms in the population at the start of the sterilisation process, and the total heat exposure, i.e., a combination of time and temperature.

13 Steam and Air

13.1 Introduction

Water may exist as a solid, liquid or gas. In a vessel filled with water in the liquid form, high activity exists among the water molecules. All molecules have a certain energy and they are moving around in the liquid all the time. Some of the water molecules have a somewhat larger energy than others and if the energy of these molecules becomes large enough, these molecules in liquid form will leave the water surface and be transported to the gaseous form. This is why a vessel containing water will always contain both the liquid and the gaseous forms of water. If energy is added to liquid water, for instance by increasing the temperature of the water, the energy of the molecules will increase, which in practice means that the higher the temperature of the water, the more water molecules are transformed from the liquid to the gaseous state. Because an equilibrium state always exists between the liquid and the gaseous states in such a vessel, some of the molecules in the gaseous state present close to the water surface will return back to the liquid state when energy is lost to the surroundings.

If an enclosed container filled with water is left for a while, an equilibrium state between water in the liquid and gaseous states will exist. This equilibrium will result in equal numbers of molecules passing from the liquid to the gaseous state and the gaseous state to the liquid state. On increasing the temperature more water molecules will pass from the liquid state to the gaseous state, thereby forcing the equilibrium until a new balance is obtained. When the temperature of the container is increased the pressure in the container will also increase. The relationship between pressure and temperature can be observed in the steam pressure curve for water shown in Fig. 49. This figure shows the borderline separating dry and humid steam. When the temperature is further increased

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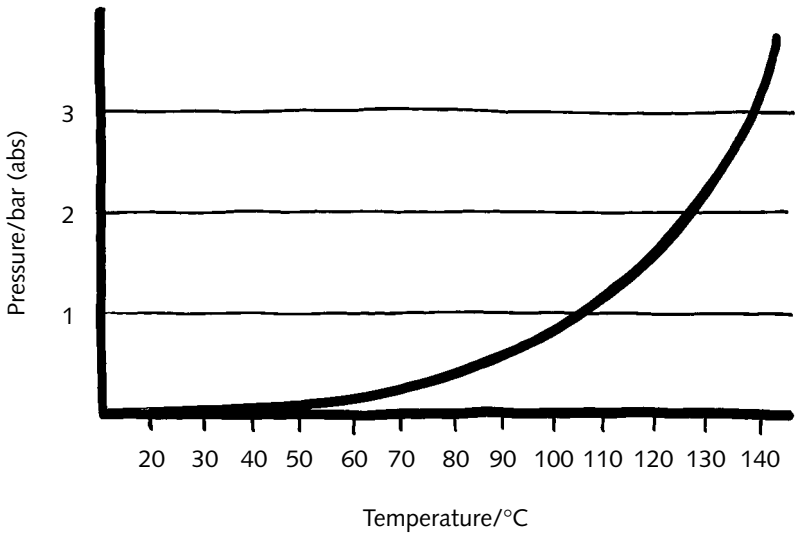

Figure 49. The vapor pressure of water.

and no liquid water is present in the container the steam will be superheated. If the temperature of the dry and saturated steam however is decreased, steam molecules will condense, i.e., the water molecules will pass from the gaseous to the liquid state. Small water droplets looking like smoke will then be formed and the steam is considered as wet or humid. Dry saturated water steam contains no humidity and is thus not visible to the human eye.

13.2 Examples of Heat Transfer

When a cold item is placed in an environment containing dry and saturated steam, the steam in close proximity to the cold item will transfer some of its energy to the item that in turn will be heated. At the same time as this energy transporting process takes place the steam will lose some of its energy to the item and will condense on the colder item. Steam, i.e., water in its gaseous state, has a much larger volume than the condensed water, so that when steam condenses on a colder item, the local steam pressure close to the colder item will decrease. This is also seen as a pressure gradient formed close to the colder item.

This local lower steam pressure close to a colder item will force more steam from the surrounding environment to come closer to the item. In short, more and more steam is forced towards a colder item during the exposure process.

13.2.1 Steam in an Autoclave

An autoclave consists of a totally sealed environment to which steam is added in order to transfer energy to colder items. A traditional steam autoclave can be viewed as a large pressure cooker where steam under pressure is allowed to come into contact with items that are to be sterilised. During the sterilisation process pressure and temperature can be regulated by the addition of more steam. In this way a constant pressure can be obtained in spite of the fact that energy is transferred to colder items and that condensation will take place during this energy transfer. At equilibrium a constant steam pressure will result in a constant sterilisation temperature.

13.3 Saturated Water Vapor

When saturated water vapor is given more and more energy, the pressure will increase rapidly with increasing temperature. This will affect the goods to be sterilised in the autoclave. Such an increased temperature might for instance affect injection vials filled with product.

The difference between the pressure in the autoclave chamber and the internal pressure in the injection vials can result in problems. When an injection vial is to be sterilised in an autoclave, a lower pressure will be obtained in the vials during the initial heating phase of the sterilisation cycle. In practice this means that the vials will be colder than the surrounding steam. Problems might not only occur during the initial phase of an autoclave cycle. The same problems may occur during the cooling phase. When the vials in the final phase of the steam sterilisation are cooled a relative over-pressure in the vial will be created. In certain autoclaves it is possible to regulate the total pressure in an autoclave chamber independent of the temperature in the autoclave. This possibility to independently regulate pressure during autoclaving is used to minimize the risk of vials imploding, exploding or becoming deformed. More details on this type of situation can be found in section 14.

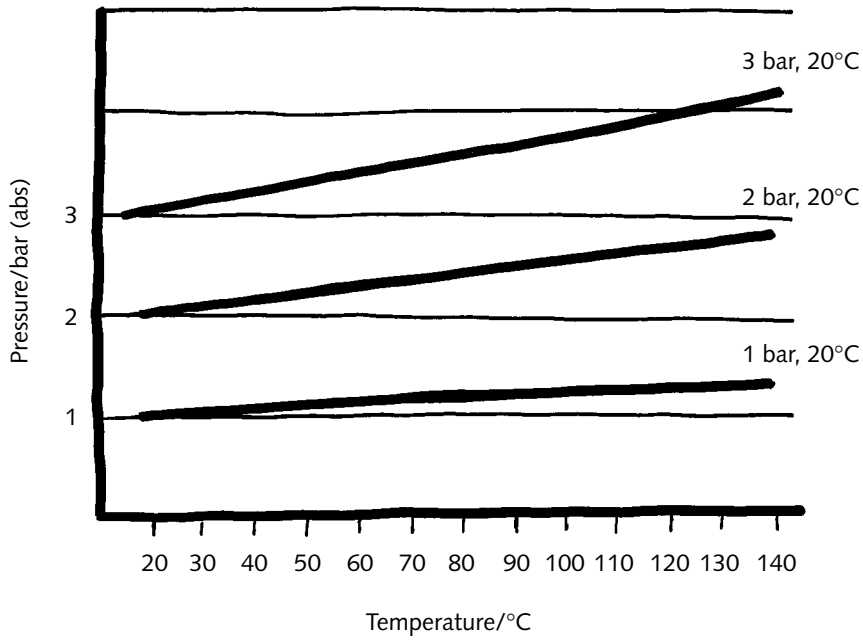

Figure 50. Pressure of air.

Steam is used in autoclaves because water is a very good medium in which to store energy, and it is also very easy to transport from the place of production to the point of use.

Steam is also an extremely good medium to use in order to transfer energy to colder items. When steam condenses, large amounts of energy are transferred to the items to be sterilised. At the same time the existing microorganisms will be saturated with water, which in turn has a very large impact on the destruction of organisms at the actual sterilisation temperature.

13.4 Air Pressure

Air is a gas mixture following the general relationship that the total pressure in a constant volume of air will increase linearly with increasing temperature, Fig. 50. When the temperature in an enclosed container containing air is increased from 20 to 120 °C, the pressure in the enclosed air will increase from 1 bar to 1.3–1.4 bars.

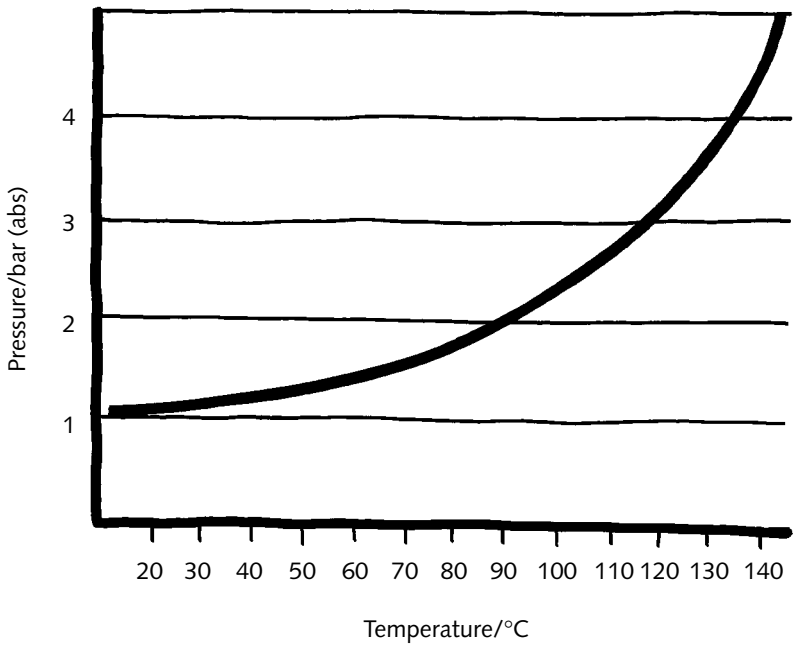

Figure 51. Pressure of mixtures of steam and air.

13.5 Steam and Air Mixtures

When different gases are mixed together, Dalton's law is valid. This law states that the total pressure of a gas mixture corresponds to the sum of the partial pressure of all the different gases in the mixture. This means that a gas mixture comprised of air and steam will have a total pressure corresponding to the sum of the steam pressure and the air pressure, Fig. 51.

The relative humidity is often used to show how much humidity exists in the surrounding air. The term relative humidity describes the degree of humidity, i.e., the amount of water vapor present in the air. From a purely mathematical point of view the relative humidity is equal to the relationship between the existing steam pressure and the pressure of the saturated water vapor at the studied temperature, i.e., $P_{tot} = P_1 + P_2 + P_3 + \ldots$

When the relative humidity is 100% the dew point is reached. The dew point corresponds to the temperature when the water vapor condenses.

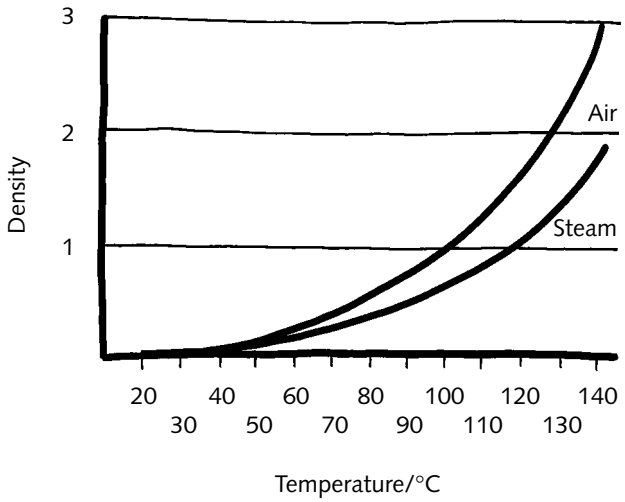

Figure 52. Density of air and steam at different temperatures.

13.6 Density of Steam and Air

Figure 52 shows that the density of air is approximately 50% greater than the density of steam at 120 °C, if the air and steam densities are measured at the same pressure and temperature. If, however, steam and air are mixed homogeneously, the relationship will become different, Fig. 53. The density curves for air and steam at a total pressure of 2 bar, indicates that at 105 °C steam and air have the same density. At a temperature lower than 105 °C the air has a higher density than steam, whereas steam has a higher density than air when measured at a temperature above 105 °C. This means that the combination of pressure and temperature will have a great impact on which component in a steam and air mixture has the highest density.

13.7 The Process Taking Place in an Autoclave

Air present in an autoclave during a sterilisation process will have a negative impact on the effect of the saturated water vapor, effectively humidifying and thereby heating the goods to be sterilised. When steam is initially introduced into an autoclave, steam and air will be separated from one another. This indicates that a temperature difference in the region of 5–10 °C will be obtained between the top and the bottom of

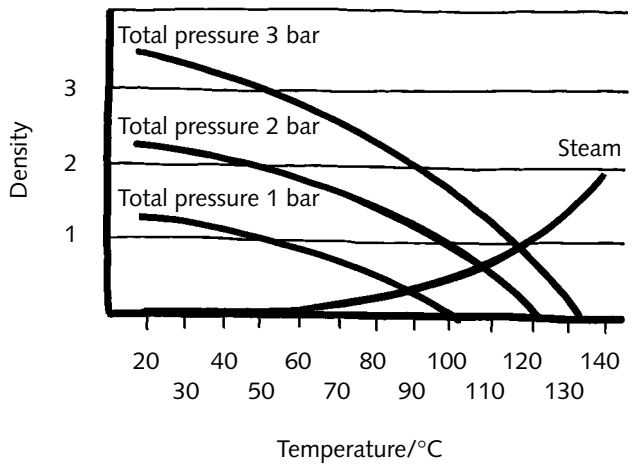

Figure 53. Density of different air and steam mixtures.

the autoclave. Since air has a higher density than steam at lower temperatures, the bottom part of the autoclave will be at a lower temperature. In order to obtain a more homogeneous temperature in the autoclave the air must be removed, for instance by exposing the autoclave chamber to vacuum before steam is introduced. The temperature difference in the autoclave chamber when using vacuum during the initial sterilisation phase becomes less than 0.5 °C. To avoid problems with, for instance, injection vials becoming deformed, imploding or exploding, the total pressure in the autoclave chamber can be controlled by the addition of pressurised air. However, the added pressurised air must in these cases be thoroughly mixed with the steam, for instance by the aid of a fan placed in the autoclave.

13.8 Steam Quality

No official definition of steam quality exists for different pharmaceutical applications. The European standard EN 285, ›Sterilisation – steam sterilisation – large sterilizers‹, defines some parameters for clean steam and for water steam applications used within the medical sector, Tab. 6. A suggestion from the NHS Estates Health Technical Memorandum 2031,

Table 6. Characteristics of condensate and feed water according to EN 285 (valid within the hospital sector).

Contaminant	**Condensate/mg kg^{-1}**	**Feed water/ mg L^{-1}**
Evaporation residue	≤ 1.0	≤ 10
Silicon oxide (SiO_2)	≤ 0.1	≤ 1
Iron	≤ 0.1	≤ 0.2
Cadmium	≤ 0.005	≤ 0.005
Lead	≤ 0.05	≤ 0.05
Heavy metal residues (excluding Fe, Cd, Pb)	≤ 0.1	≤ 0.1
Chloride (Cl^-)	≤ 0.1	≤ 2
Phosphate (P_2O_5)	≤ 0.1	≤ 0.5
Conductivity (20 °C)/ µS cm^{-1}	≤ 3	≤ 15
pH- value	5–7	5–7
Appearance	Free from color, clean without sediment	Free from color, clean without sediment
Hardness (∑ alkaline ions)/mmol L^{-1}	≤ 0.02	≤ 0.02

HTM 2031, give some suggestions on certain parameters for clean steam for pharmaceutical use, Tab. 7.

Contaminants in steam can have a major impact on the goods to be sterilised. Small quantities of contaminants can, for instance, be transferred to surgical instruments, cloths used for wound care and even cause the contamination of pharmaceutical products. Also indirect transfer from different vials and materials can have an impact on the patient to be exposed to the sterilised items.

Water vapor is obtained by steam formation from water in a steam generator. When water is boiled in such a system, small non-volatile water drops in the form of aerosols will be transferred from the liquid water surface to the steam above the liquid surface. These water droplets can leave the steam generator together with the steam. The critical part of such a

Table 7. Characteristics for condensate according to the suggestion in HTM 2031, intended for pharmaceutical use.

Contaminant	**Condensate**
Evaporation residue	≤ 10 mg L^{-1}
Silicon oxide (SiO_2)	≤ 0.1 mg L^{-1}
Iron	Not specified
Cadmium	Not specified
Lead	Not specified
Chloride (Cl^-)	≤ 0.5 mg L^{-1}
Phosphate (P_2O_5)	≤ 0.1 mg L^{-1}
Sulphate	
Ammonia	≤ 0.2 mg L^{-1}
Endotoxins	≤ 0.25 IU ml^{-1}
Conductivity (20 °C)	
pH value	
Appearance	Clear, free from color, smell and taste
Hardness (Σ alkaline ions)	

situation is that these water droplets are not as clean as the steam and can incorporate, for instance, ions and other soluble and non-soluble material, volatile material and pyrogens (endotoxins), that can contaminate the goods to be sterilised.

Within the pharmaceutical industry the steam can be divided, from a cleanliness point of view, in four different qualities, factory steam, filtered steam, clean steam and pure steam.

13.8.1 Factory Steam

Factory steam is sometimes also referred to as house steam. This type of steam is often produced in an industrial steam generator. It contains different types of contaminants, for instance additives added to the feed water to the generator in order to reduce the possibility of corrosion and the formation of insoluble substances such as limestone. Different types of iron oxides and other types of contaminants from the feed water and residues from the steam generator or arising from the steam distribution system may also be present. These types of contaminants are often quite easily detected, since they discolor the goods to be sterilised or even give a thin discolored surface coating on the inside of the autoclave chamber.

13.8.2 Filtered Steam

Filtered steam is factory steam that has been allowed to pass a filter, removing some of the particles present in it. Filters for steam applications are traditionally manufactured from stainless steel and have a removal rating of approximately 10–20 μm. Colloidal iron particles from the steam generator or from the distribution system are very difficult to remove since these minute particles often quite easily penetrate the steam filter, where they later aggregate to form larger particles. When too high flow rates are allowed through a steam filter, condensate and small droplets of liquid may also penetrate the filter.

13.8.3 Clean Steam

Clean steam is the second highest quality of steam. Clean steam is produced in a steam generator constructed from acid-proof stainless steel. The steam producing system does not include a separator system. The feed water entering the steam generator is often pre-treated, for instance by allowing it to pass a softening filter where calcium and magnesium ions are eliminated. The water can also be further de-ionised or treated in a so-called reversed osmosis unit.

There are no absolute guarantees that the feed water in these treatments will be freed from all ions, particles or pyrogens. These types of contaminants can be transported far away in the form of small water droplets in the steam. In order to increase the quality of the steam, it is filtered through a stainless steel filter with a removal rating of between 3 and 5 μm. At higher steam velocities even condensate and small water droplets can pass such a filter. Clean steam often corresponds to the quality demands stated in EN 285 together with the fact that the quality of the condensate should correspond to the demands of Water for Injection (WFI) according to most pharmacopoeias. The most important thing to remember is that the feed water used for steam production has a major impact on the total quality of the steam.

13.8.4 Pure Steam

Pure steam is the highest quality of steam. The quality of the condensate from pure steam should correspond to the demands of Water for Injection (WFI). In practice this means that the condensate should have a low content of ions, the concentration of endotoxin should be lower than

0.25 IU ml^{-1}. In most cases there also exists a demand for freedom from particulate contaminants. Pure steam is produced in a steam generator manufactured from a high degree acid-proof stainless steel. The steam production system is also equipped with a separation system in order to prevent small water droplets and other particles leaving the system. The steam distribution system after the steam generator should be manufactured from steel of the same quality as that of the generator.

The separation system used in the production of pure steam is intended for collecting very small water droplets and other particulate contaminants to be transported along with the steam to the point of use. Separation systems are generally based on removal due to gravitation, cyclone removal and/or even mechanical removal. In many cases a combination of these techniques is used, Fig. 54. The feed water to the steam generator is often softened, de-ionised and (or) RO-treated.

13.9 Feed Water

The features and quality of the feed water play an important role in the quality of the produced steam as well as in the impact the feed water has on the steam generator itself. EN 285 defines to a certain degree quality aspects of the feed water, Tab. 6.

In some cases softened water, i.e., water of drinking quality that has been freed from calcium and magnesium ions in a bed-type filter, is used. In many cases softening of the water is not a good enough pre-treatment method in order to give the correct quality of steam. This is why different water treatment methods are often combined, for instance deionisation or reversed osmosis, in order to obtain as low ion contents in the water as possible. In order to avoid insufficiently high quality steam that damages the steam generator by corrosion and gives residues that cannot be volatilised and are collected within the steam generator, these destructive components must be removed from the generator continuously or at certain pre-determined time intervals.

Microorganisms also have a great impact on steam quality. Steam is used for sterilisation purposes, which in practice means that the steam itself is sterile, but up to 98% of all the

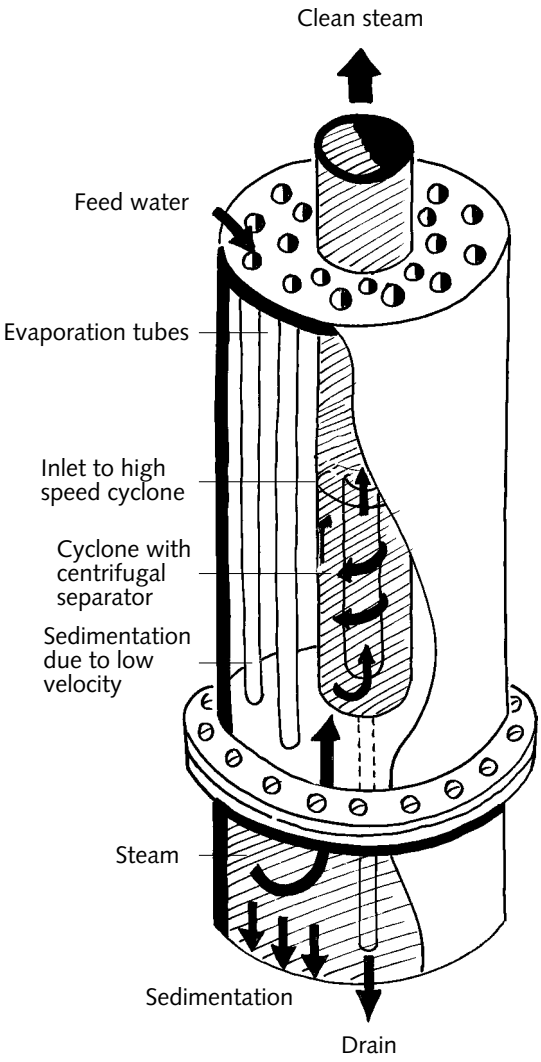

Figure 54. A combined separation system for steam.

microorganisms that are present in systems for feed water have the ability to form endotoxins. This is why microorganisms in the feed water must also be controlled.

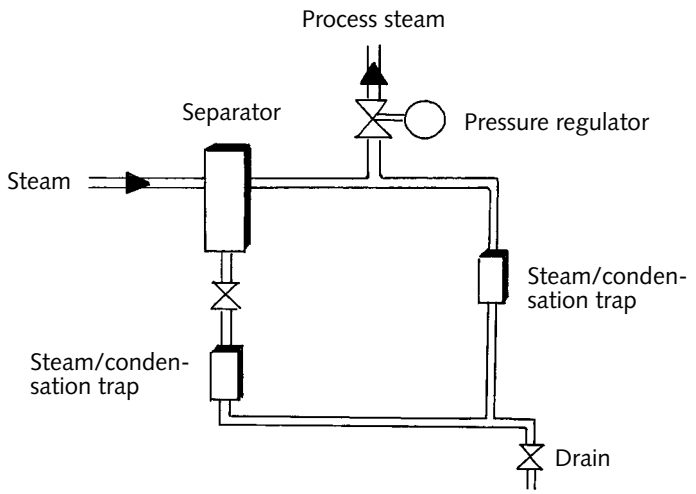

Figure 55. Schematic representation of a steam distribution system.

13.10 Steam Generator and Distribution System

The low ion content of the feed water and of the steam itself makes both the liquid feed water and the steam aggressive towards the materials used in the generator and the distribution system and can result in corrosion. The material in the steam generator as well as in the distribution system should therefore be stainless steel, for instance AISI 316, 361 L and 316 Ti. The steam distribution system should be constructed without any dead-legs and be equipped for instance with automatically working steam traps. The steam should be removed for use from the top of the steam pipe, Figure 55.

13.11 The Physical Features of Steam

The physical features of steam including steam dryness, superheating of steam and the impact of so-called non-condensable gases, will affect the possibility of the steam sterilising the goods. Examples of such features are given in EN 285, Tab. 8.

Steam for sterilisation purposes should always be dry and saturated. A too high level of humidity will automatically create large amounts of condensates. The condensate is formed on the colder goods to be sterilised, resulting in them being more or less covered with condensate, which in turn

Table 8. Characteristics of steam according to EN 285

Dryness value	≥ 0.95 (≥ 0.90)
Non condensable gases	≤ 3.5%
Super heating	≤ 25 K
Fluctuations	≤ 10%
Reduction	≤ 2 : 1

will mean that the wetted goods will not reach the sterilisation temperature as fast as if they were dry. Large amounts of condensate might also result in difficulties in obtaining the sterilised goods in a dry state after sterilisation is complete.

Superheated steam on the other hand will dry out the goods and also eventually any microorganisms present, which can cause great problems. Dehydrated microorganisms or microbiological spores will thus become much more heat resistant and will demand a longer sterilisation time as compared to the use of dry and saturated steam. Non-condensable gases in the steam will also more or less hinder the steam from coming close enough to the goods.

13.12 The Impact of Steam Quality

Many sterilised items are used in situations where the natural defence mechanisms of the body are out of control. Contaminants that are added to the sterilised items can have a direct and often quite harmful impact on a patient. Within the pharmaceutical industry pure steam should be used when sterilising materials are to come into direct contact with a pharmaceutical product. This is because one should avoid the possibility of contaminants within a product being injected or coming into direct contact with unprotected skin.

Within the countries of the European Union, clean steam is often used for pharmaceutical products that are sterilised in their own container, provided that a validated closure process and leak-test system has been used. A suggestion from HTM 2031 (see page 139) states that Water for Injection has been documented as safe for medicinal use, during the past years. The quality demands in EN 285 are in many cases higher, but limit values are not stated for endotoxins and microorganisms. Steam with a quality according to EN 285 is also more

Table 9. Comparison between EN 285, HTM 2031 (suggested) and USP XXII.

Contaminant	**Condensate**		
	EN 285/mg kg^{-1}	HTM 2031/mg L^{-1}	USP XXII/mg L^{-1}
Evaporation residue	≤ 1.0	≤ 10	≤ 10
Silicon oxide (SiO_2)	≤ 0.1	≤ 0.1	
Iron	≤ 0.1	Not specified	≤ 0.1
Cadmium	≤ 0.005	Not specified	
Lead	≤ 0.05	Not specified	
Heavy metal residues (excluding Fe, Cd, Pb)	≤ 0.1		≤ 0.5
Chloride (Cl-)	< 0.1	≤ 0.5	≤ 0.5
Phosphate	≤ 0.1	≤ 0.1	
Sulfate			≤ 0.5
Ammonia		≤ 0.2	≤ 0.3
Calcium			≤ 0.5
Carbon dioxide			≤ 4.0
Copper			≤ 0.01
Chromium			≤ 0.01
Cobalt			≤ 0.1
Manganese			≤ 0.1
Nickel			≤ 0.1
Oxidisable substances			Test decides
Microbiology			≤ 10 CFU/100 ml
Endotoxins		≤ 0.25 IU mL^{-1}	≤ 0.25 IU ml^{-1}
Conductivity (20 °C)	≤ 3 µS cm^{-1}		≤ 10 µS cm^{-1}
pH value	5–7		5–7
Appearance	Free from color, clean without sediment	Clear, free from color, smell and taste	Free from color, clean without sediment
Harness Σ alkaline ions)	≤ 0.02 mmol L^{-1}		

(NB! A direct comparison is not totally feasible)

aggressive towards the steam generator and the distribution system, see comparison in Tab. 9. Steam giving a condensate of Water for Injection-quality is therefore more suitable for pharmaceutical use.

13.13 Conclusion

When planning a system for steam production used for sterilisation purposes, steam quality should be carefully discussed and defined. A clean steam generator producing steam with a

condensate quality corresponding to Water for Injection (WFI) should be used. It is also of vital importance to design and install the distribution system in such a way that physical and microbiological demands are fulfilled. The steam and feed water system must be checked on a regular basis in order to secure the correct quality.

14 Autoclaves and Processes for the Pharmaceutical Industry

14.1 Introduction

According to the ISO (International Standards Organisation) there are five different types of sterilisation processes within the pharmaceutical industry. The different processes make different demands in regard to the construction of the autoclave. The most commonly used autoclaves are the conventional steam autoclave, the circulating water autoclave and the ventilator autoclave. The type of autoclaving process, as well as the type of autoclave, depend on the features of the product to be sterilised together with the features of the product after sterilisation.

14.2 Conventional Steam Autoclave

In a conventional steam autoclave, Figure 56, heat transfer to the goods to be sterilised is obtained by using dry and saturated steam of a good enough quality. Steam and air do not mix spontaneously, resulting in a situation in which air might be incorporated inside the goods and also in the packaging material covering the goods. This is the reason why it is very important to replace all the air in the autoclave with steam. This replacement can be obtained for instance by allowing steam to flow through the autoclave. This method is, however, not effective if air is included in the packed goods or if, for example, air is incorporated between small ampoules placed very close together. In these cases a better and more effective way to remove air from the autoclave is by applying a vacuum prior to the introduction of steam. The application of the vacuum can be repeated several times if necessary.

The sterilisation phase is maintained by the addition of dry and saturated steam for a predetermined time period. If the goods to be sterilised are sensitive to pressure differences, an external supporting pressure consisting of pressurised air can be supplied to the autoclave chamber during the final cooling

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phase. Cooling of the goods is achieved by compensating the chamber pressure, adding compressed air, the aid of a vacuum, cooling with the aid of a jacket surrounding the chamber of the autoclave or spraying the goods in the chamber with water of good chemical and microbiological quality. In order to avoid too high temperature changes of the goods in the autoclave, so-called temperature stress on the goods which can be observed on larger glass vials, the water used for initial spraying should be warm. Another way to cool the goods with water is to introduce the cold water as a high pressure aerosol. When the goods need to be obtained in a dry state after sterilisation, a final vacuum step can be applied and/or heating of the surrounding jacket of the autoclave.

The water used for cooling the goods should be sterile or of a good microbiological quality. This is not easy to accomplish and therefore it is necessary to have thorough control of the cooling water in order to avoid contamination of the goods. Furthermore the ionic content of the cooling water should be low in order to avoid crystallisation of salt on the goods and also on the interior surfaces of the autoclave chamber.

A conventional steam autoclave is suitable for the sterilisation of:

- Porous goods, for instance textile material
- Equipment (also packed equipment)
- Packaging material where a certain amount of residual humidity is allowed
- Ampoules
- Injection bottles
- Infusion bottles (glass)
- Liquids in open containers (for instance microbiological growth media)

A conventional steam autoclave should however not be used for sterilisation of products that must be completely dry after sterilisation. This process is not suitable for sterilisation of products in blister packages because too much condensate will be left in the package after drying.

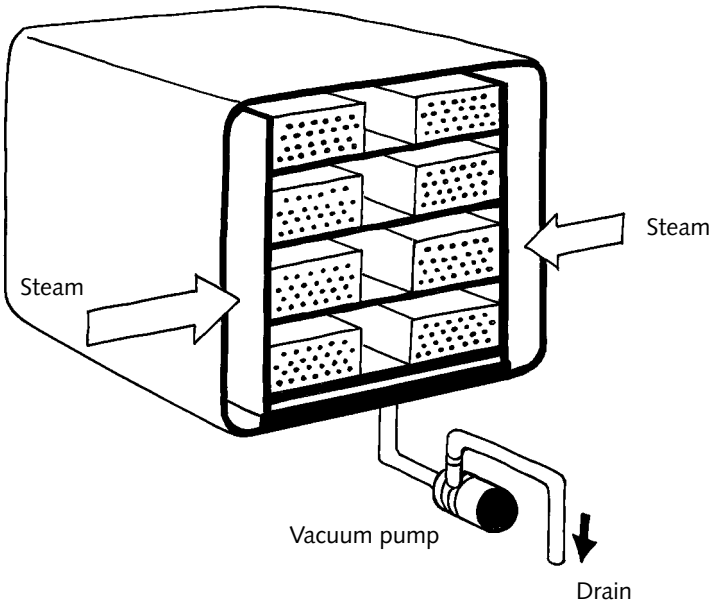

Figure 56. Conventional steam autoclave.

14.3 Circulating Water Autoclaves

Heat transfer in a circulating water autoclave (Figure 57) is obtained by using water that is heated to the sterilisation temperature. The water, preferably of a quality corresponding to Water for Injection or purified water, is supplied to the autoclave chamber at a level just above the goods. The water is then continuously recirculated in the autoclave with a circulation pump and is sprayed over the goods. During the circulation an external heat exchanger is used to regulate the temperature of the water. During heating and the entire sterilisation phase, steam is used on the primary side of the heat exchanger. Since this steam never comes into direct contact with the water heating the goods, a poorer quality of steam, for instance factory steam, can be used.

After the sterilisation phase is complete, the goods can be cooled by decreasing the temperature of the circulating water. The temperature decrease is obtained by exchanging the steam on the primary side of the heat exchanger with cold water. The circulating water within the autoclave chamber is then cooled and in turn cools the goods. Since water and air

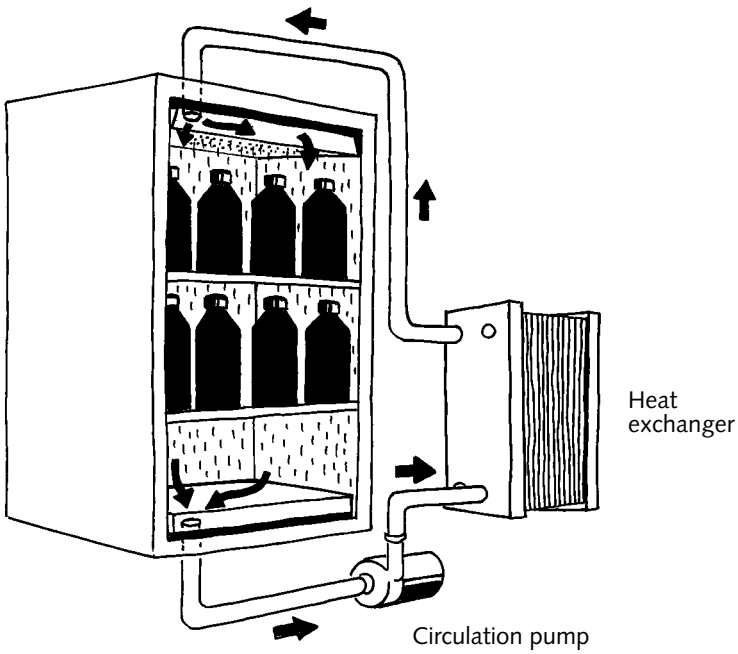

Figure 57. Circulating water autoclave.

are mixed effectively during the process, a support pressure supplied by pressurised air can be used during the entire sterilisation process. This pressure can be altered independently of the temperature in order to avoid the goods becoming deformed.

A circulating water autoclave is primarily used for sterilisation of injection bottles and infusion solutions. An advantage of this sterilisation process is the very effective heat exchange, an even temperature distribution and a guaranteed sterile cooling medium since the water is sterilised together with the goods during the sterilisation cycle. The circulating water autoclave does not generate risks of cracks in glass bottles due to temperature stress, since the cooling phase is performed gradually.

The goods are however wet after the sterilisation process, but if the goods are removed from the autoclave when still relatively hot (approximately 50–70 °C) the residual water will have a good chance to evaporate.

Certain circulating water autoclaves require that the entire autoclave chamber is filled with water that is then circulated during the process. The energy needed for such a process will however become unnecessarily high, and the total weight load on the floor on which the autoclave is placed will increase dramatically.

14.4 Ventilator Autoclaves

In a ventilator autoclave (Figure 58), also sometimes referred to as an air/steam autoclave, heat exchange is obtained by using a mixture of air and steam. It is extremely important that this gas mixture is homogeneous and this is achieved by the aid of very powerful fans that can be operated at two different speeds. The temperature and pressure can be varied independently of one another during the circulation of the steam and air mixture by using a heat exchanger placed above the goods in the autoclave chamber. The heat exchanger is used for both heating and drying and cooling of the goods.

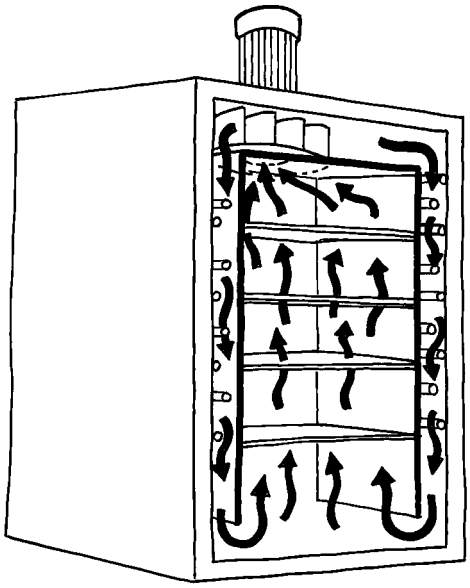

Figure 58. Ventilator autoclave.

Nearly all types of goods can be sterilised in a ventilator autoclave. This type of autoclave is used when there is a demand for dry goods after sterilisation, for instance products packed in blister packaging, rubber sealing, pre-filled syringes, PVC bags (that will be transparent after sterilisation) and porous goods such as textile material.

The goods to be sterilised may be preheated, for instance by introducing hot and dry air through the heat exchanger. Preheating will minimise the formation of condensate on the goods. Residual moisture after terminating the sterilisation cycle can be eliminated quite easily by circulation of hot air in the chamber.

14.5 Air to the Autoclave Chamber

All air that is to be introduced into an autoclave chamber must be sterile filtered. The filters used in this type of application must be able to be sterilised and integrity tested. Sterilisation and integrity testing must be documented, preferably on the internal printer of the autoclave.

14.6 Leakage Test of Ampoules

Autoclaves used for sterilisation of ampoules can also incorporate a program for leakage testing. Such a test can start with quite a large under-pressure, followed by the addition of a strong colored test solution into the chamber. During the next step an over-pressure is then applied to the chamber, forcing the colored test solution into small cracks in the ampoules. Using this type of test it becomes easier to remove colored and thereby unapproved ampoules.

14.7 Autoclave Control System

It is not only vital to choose the correct autoclave type for the sterilisation of a certain type of goods but also to use a good enough control system and software in order to optimise the sterilisation process. The control system must be able to incorporate all the different process steps that are used. In some cases it is possible to run several autoclaves from the same control system. In order to use a new sterilisation process the control unit should be able to be re-programmed or to use a new software. An automatic calibration program both

saves time and minimises the sources of problems. It is necessary to be able to change parameters like pressure, time and temperature. The task of changing parameters should however only be able to be performed by specially assigned personnel, by using different types of authorisation codes to get into the program. The control system should be able to document the entire process in regard to time, pressure, temperature and F_0-value, alphanumerically as well as in the form of a diagram. Furthermore, different types of alarms should be documented. It is a great advantage also to have an independent supervision system, i.e., a system that is able to collect signals independent of the actual control system and also able to compare these two signals, Figure 59. If the two signals are different, this will indicate that something unexpected has occurred. An alarm should then be registered. There should also be a backup that can store all data from the latest autoclavation.

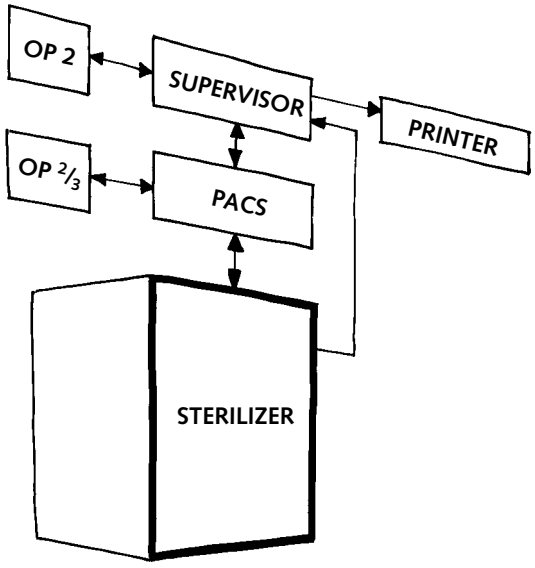

Figure 59. Control system for autoclaves.

14.8 Conclusion

It is the features of the product prior to sterilisation and also the desired features after sterilisation, that are the basis for choice of autoclave, i.e., conventional steam autoclave, circulating water autoclave or ventilator autoclave.

15 Validation

15.1 Introduction

In simple terms validation can be explained as an activity performed in order to ensure that a process really works or functions in a desired way. Validation is thereby an important part of the quality assurance and is used in most production industries. A development during recent years is that validation has been used more systematically. A very big difference is observed when viewing validation documentation.

The term validation was first introduced within the American space industry in order to save money and lives. The pharmaceutical industry soon followed an initiative from government authorities. The major aim of validation in this context is to secure the safety of the end user of the pharmaceutical product, namely the patient, i.e., to give the patient a safe pharmaceutical product with the desired effect. The reason why validation was introduced within the pharmaceutical industry was a number of serious accidents, mostly concerning products that had been declared as sterile but were manufactured without the necessary quality assurance.

This chapter is mainly based on the European norms EN 554; Sterilisation of Medical Devices – Validation and Routine Control of Sterilisation by Moist Heat and European Community GMP, Good Manufacturing Practice for Medicinal Products in the European Community.

15.2 Definitions and Explanation

15.2.1 EN 554

Commissioning: obtaining and documenting evidence that equipment has been provided and installed in accordance with its specifications and that it functions within predetermined limits when operated in accordance with its operational instructions.

Installation qualification: see commissioning.

Performance qualification: obtaining and documenting evi-

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dence that the equipment as commissioned will produce acceptable products (goods) when operated in accordance with the operating instructions (process specification).
Validation: a documented procedure for obtaining, recording and interpreting data required in order that the process will consistently comply with predetermined specifications.
Note: for moist heat sterilisation, *validation* is considered as a total program that consists of *commissioning and performance qualifications*.
Sterile: Condition of a medical device that is free of viable microorganisms.

15.2.2 EC GMP

Validation: action of proving in accordance with the principles of Good Manufacturing Practice that any procedure, process, equipment, material, activity or system actually leads to the expected results (see also Qualifications).
Qualification: action of proving that any equipment works correctly and actually leads to the expected results. The word validation is sometimes widened to incorporate the concept of qualification.
Sterility: sterility is the absence of living organisms. The condition of the sterility test is given in the European Pharmacopoeia.

15.2.3 Sterilisation

Sterilisation processes follow statistical relations and sterility can therefore not be guaranteed in absolute terms. According to current Good Manufacturing Practice, routines and processes should be such that, after a sterilisation process, at the maximum one surviving microorganism per one million sterilised units is allowed. This can also be expressed as a sterility assurance level (SAL) of 10^{-6} or better.

The result from a sterilisation process is dependent on the heat resistance of the microorganisms (the *D*-value of the microorganism), the number of microorganisms in the unsterilised package (often referred to as the bio-burdened, presterilisation level) together with the heat exposure (time and temperature).

A microbiological declination curve is obtained when the logarithm of the number of living microorganisms is plotted against time during a sterilisation process, see Fig. 48. At a

predetermined temperature a straight line is obtained with a slope dependent on the actual *D*-value. The process time needed in order to reach a certain SAL-value is dependent not only on the slope of this line but also on the number of microorganisms present in the unsterilised product.

A sterilisation process can be designed as overkill, often 120 °C for 15 min, or tailor-made, based on known microbiological parameters.The overkill process often needs less complex validations than tailor-made processes.

When a sterility test is performed according to the European Pharmacopoeia there is a minor possibility of detecting whether a batch is sterile or not.

Within EN 554 it is stated ›… process efficacy cannot be verified by inspection and testing of the product‹. It also states ›… labelling a medical device ›Sterile‹ is only permissible when a validated sterilisation process has been used‹.

The only way to guarantee sterility is to work with validated processes and to control that the process has performed as intended. Release of a batch without final control based on a validated and documented process is called parametric release and can be used after admittance of concerned authorities.

15.3 What is Validation

Within the EC GMP the following is stated in regards to validation:

›1.3 the basic requirements of GMP are that:

II. Critical steps of manufacturing processes and significant changes to the process are validated‹.

In annex one the following is stated:

›55. All sterilisation processes should be validated … Where possible, heat sterilisation is the method of choice.‹ This statement is in itself totally clear, all sterilisation processes, for instance of products, appliances and preparation systems, must be validated. It also states, together with other documents, that heat treatment with saturated water vapor is the method of choice.

›56. Before any sterilisation process is adopted, its suitability for the product and its efficacy in achieving the desired sterilising conditions in all parts of each type of load to be

processed should be demonstrated by physical measurements and by biological indicators where appropriate. The validity of the process should be verified at scheduled intervals, at least annually, and whenever significant modifications have been made to the equipment. Records should be kept of the results.‹

Before a sterilisation process is used for routine production it must have been shown to be suitable for each type of load and product.

›57. For effective sterilisation the whole of the material must be subjected to the required treatment and the process should be designed to ensure that this is achieved.‹

›58. Validated loading patterns should be established for all sterilisation processes.‹

All load patterns must be validated. This is important when using the same process for different sizes of loads. Maximum and minimum loading should under all circumstances be validated.

›91. The sterility test applied to the finished product should only be regarded as the last in a series of control measures by which sterility is assured. The test should be validated for the product(s) concerned.‹

Sterility testing, as explained in the pharmacopoeia, gives little or no chance of finding whether a badge is sterile.

›92. In those cases where parametric release has been authorised, special attention should be paid to the validation and the monitoring of the entire manufacturing process.‹

15.3.1 Practical Validation

It is always the user who through a specially assigned qualified person is responsible for the validation. This does however not mean that an external consultant cannot perform the entire validation. The user should approve the validation program, the performance and the result.

Validation should be performed when, for instance, a new process, a new autoclave or a new load pattern is to be used. It should also be performed after maintenance or changes in the process or in the media supplies. If, after a change in, for instance, the steam distribution system, less steam is supplied to the autoclave than before the change, it is not possible to perform the process for the same time as before the change. It

is not even certain that the sterilisation temperature will be obtained. Revalidation should also be performed at certain time intervals, at least once a year.

Validation should follow a validation program. This program should incorporate

- Test plan
- Specification of the autoclave and the processes
- Criteria of acceptance
- Test methods

It is often quite difficult to perform a validation without deviating from the protocol. Such deviations must be approved by the persons who originally approved the protocols. Measuring instruments must be calibrated before and after each test. The calibration should in some way be traced back to some type of national standard.

Before a validation, the system and routines that the validation object is dependent on must be validated or shown to function correctly. Examples of such systems and routines are test methods, premises, the surrounding environment, media, methods of work, standard operating procedures and personnel.

15.3.2 Qualification Documentation

A thorough documentation will facilitate the validation. The documentation can comprise

- Description of function
- Design specification, i.e., the documented design that the manufacturer and the user have approved
- Overall drawings
- The deliverer's quality policy
- The deliverer's control and acceptance tests
- Description of control system and application program
- The deliverer's calibration procedure
- Pressure vessel certificate

15.3.3 Pre-Qualification

A pre-qualification is performed at the deliverer's or manufacturer's site. It should as far as possible be identical with the validation later performed by the user and it is therefore necessary to design the pre-qualification with the user. An example of documentation and tests is:

- Test protocol from the receiving of material of use
- Process control from manufacturing
- Delivery testing, including test manuals, compliance of specification, safety testing and test certificate

15.3.4 Installation Qualification

Commissioning or Installation Qualification (IQ) means that control and documentation of the equipment are in compliance with specifications and that it has been installed correctly. The manufacturer can perform most parts of the installation qualification.

15.3.5 Operation Qualification

Operation Qualification (OQ) means a functional test with the IQ approved empty autoclave. An example of an OQ activity is a test of incoming media, a test of analogue and digital signals, calibration and mechanical function tests. Other examples are tests of closures and alarms, leakage tests, test runs of programs and temperature distribution in the empty chamber.

15.3.6 Performance Qualification

Performance qualification (PQ) means testing with product in relevant load patterns. It is during this phase that the process is defined. Example of PQ activity is that for each and every process a load pattern test must be performed regarding heat transfer in the product, ability to reproduce (normally performed on three repeated processes) as well as spore testing.

Spore samples are frequently used in order to biologically verify a process. Commercially available spores have the feature that no survivals are expected after the sterilisation process. Certain companies have their own spores with very high *D*-values. Dilution series give the number of surviving spores at a certain F_0-value and this technique is used at the same time to verify the actual F_0 level.

15.3.7 Acceptance Criteria

All companies can define their own acceptance criteria. Examples of such found in EN 554 are:

- Time and temperature to be within defined limits during the sterilisation phase
- The temperature should be kept within sterilisation temperature limits with the highest temperature +3 K above the sterile temperature

- The temperature from a sensor should not vary by more than 1 K during the sterilisation phase
- The temperature from different sensors should not differ by more than 2 K during the sterilisation phase

According to EN 554 the validation should be performed with 12 temperature sensors per cubic meter of chamber volume. Within the pharmaceutical industry fewer temperature sensors are used when utilising large chamber volumes. EN 554 is also valid for sterilisation of medical devices and is not specifically designed for the pharmaceutical industry.

15.3.8 Certification

Certification means a formal collection and judgement of the validation results. It should be performed by persons not directly involved in the practical validation work. It is also recommended that a conclusion of the validation is produced.

15.3.9 Revalidation

Revalidation should be performed at defined time intervals or when changes have been made in the process or in the media system, but at least once a year. In order to interpret the result the same type of protocol and methods used during the first validation should also be used during revalidation.

15.3.10 Product Compatibility

The product and the packaging should comply with the autoclavation and this should be defined during product development. The product should however be controlled during its shelf life after taking a new autoclave in to use.

15.4 Conclusion

Validation is an activity undertaken in order to ensure that the process really performs according to the desired criteria. Validation may follow the European norms EN 554 and EC GMP and is an extremely important part of quality assurance.

16 Summary

This book deals with two very important and often used techniques, filtration and sterilisation, in order to deal with contaminants in products and in different types of process flows. The techniques differ in both their mode of operation and their respective final result.

During filtration a partly permeable barrier is used in order to capture contaminants, in this book the focus is on particles of different types. These contaminants are collected and held by the filter material but will not be eliminated from the total process. What actually happens is that particles present in the process flow are stopped and retained in the filter. This means that if such a filter is not maintained correctly, for instance by exchange of filter cartridges, cleaning or sterilisation of the filter, there is a major risk that the filter will not function satisfactorily.

During sterilisation, for instance with steam in an autoclave, dead particles will not be affected whereas microorganisms will be killed. Basic demands for this type of action are that all parameters in the process are adjusted in relation to the type of microorganism, the medium in which the microorganisms are located and the number of microorganisms that are to be killed.

The two methods discussed in this book differ from one another and the natural question is therefore : Which should I use?

As a natural conclusion we can consult the GMP ›Annex on the manufacture of sterile medicinal products‹ in regard to filtration, sterilisation and sterilisation with humid heat. In this document the following is stated:

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Filtration of medicinal products which cannot be sterilised in their final container:
Filtration alone is not considered sufficient when sterilisation in the final container is practicable … steam sterilisation is to be preferred …
… If the product cannot be sterilised in the final container, solutions or liquids can be filtered through a sterile filter of nominal pore size of 0.22 µm (or less) …
… Such a filter can remove most bacteria and molds, but not all viruses and mycoplasma …
… The integrity of the filter should be checked by an appropriate method such as a bubble point, diffusive flow or pressure hold test immediately after each use (it may also be useful to test the filter in this way before use) …
… The integrity of gas and air vent filters should also be tested after installation, sterilisation and other appropriate intervals…

Sterilisation:
All sterilisation processes should be validated …
… Where possible and practical, heat sterilisation is the method of choice. In any case, the sterilisation process must be in accordance with the marketing and manufacturing authorizations.

Sterilisation by heat:
Each heat sterilisation cycle should be recorded on a time/temperature chart …
… The chart, or a photocopy thereof, should form part of the batch record.
… After the high temperature phase of a heat sterilisation cycle, precautions should be taken against contamination of a sterilised load during cooling. Any cooling fluid or gas in contact with the product should be sterilised, unless it can be shown that any leaking container would not be approved for use.

Sterilisation by moist heat:
… The items to be sterilised, other than products in sealed containers, should be wrapped in a material which allows

removal of air and penetration of steam, but which prevents recontamination after sterilisation…
… Care should be taken to ensure that steam used for sterilisation is of suitable quality …

Glossary

Absolute rating	A way to describe the function of a filter used for the elimination of dead particles. Absolute rating is defined as the diameter of the largest, hard, spherical particle that under specified conditions can pass through a filter medium. This definition is stated by the National Fluid Power Association in the United States.
Absorption	The retaining of a component of a mixture due to the fact that the component is transferred to another state as compared to the rest of the mixture.
AC Fine Test Dust	Short for Air Cleaner Fine Test Dust. A special test dust obtained from the Arizona desert. This test dust is used in several different applications in order to study the function of a filter. This type of test dust, as compared to the more traditional ones (mostly spherical particles), more accurately resembles industrial solid contaminants.
ACFTD	See AC Fine Test Dust.
Adsorption	The capture of material on a surface due to physical as well as chemical forces. Mostly only a single layer of material is captured on the surface.
Agar	Growth medium for microorganisms
Analytical membrane	A special filter material with a very exact rating.
Autoclave	A pressure vessel used for sterilisation with humid heat.

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Bacteria	Large group of microorganisms. The largest group numerically in the surrounding environment and also on man.
Beta-value	A way to describe the function of a filter. The beta value of a filter is defined as the relation between the number of particles of a given size and larger on the upstream side and the number of particles with the same size and larger on the downstream side of the filter.
Brevundimonas diminuta	The microorganisms used when testing microbiologically rated filters in order to characterize a sterile filter.
Brownian motion	The physical phenomenon arising due to the fact that extremely small particles suspended in a gas (air) will have a randomised motion pattern. This motion pattern arises because the particles are continuously bombarded by the moving gas molecules.
Bubble point	See bubble point pressure.
Bubble point pressure	The pressure at which the largest pore of a wet filter material releases its content of liquid due to the increasing gas pressure applied to the filter. In practice the bubble point pressure is used in integrity testing of filters by the bubble point method.
Centrifugation	A forced form of separation obtained by increasing the gravity force. The gravity force is increased by allowing the sample to be purified to rotate at high speed.
Challenge tests	Test performed on filter materials intended for the elimination of microorganisms. The tests are performed in such a way that the filters are exposed to (challenged with) a fluid or a liquid in which a known microorganism is sus-

	pended in order to examine the separation effect of the filter. Challenge tests are often called destructive testing because the filter cannot be used in active service after the test.
Channeling	A major drawback of unbound filter materials in which the pores of the filter material are enlarged and thereby form channels through which the fluid can pass not fully filtered. Channeling is very common when using filters that are subject to fiber release.
CIP	See cleaning in place.
Cleaning in place	A way to perform cleaning of equipment without having to disconnect all the various parts of the process. The cleaning process is achieved by allowing cleaning solutions to recirculate at high flow rates through the entire system.
Clean steam	A quality term used for steam.
Clarifying filtration	A filter that has the ability to eliminate all visible particles. The visibility of a particle for a naked, healthy human eye in normal daylight is approximately 40 µm.
Coarse filter	A type of filter that is intended for the elimination of coarser material, for instance algae, leaves and other organic material in nature. This type of filtration is usually performed by using some type of sieve.
Colloidal particles	Very minute particles, approximately 0.1–0.001 µm, that when observed in a microscope tend to be floating totally free in a liquid or a gas.
Compatibility	A term usually used to express the degree by which the process, i.e., the product, cleaning solutions, steam and other more physical factors have an impact on the process equipment. Inert

	materials are generally stated to have a good compatibility.
Condensate	The water in liquid form that is formed on cooling steam.
Condensation	The process by which a gas is transferred into the liquid form.
Cross flow	A technical way to perform a filtration process. The fluid to be filtered is allowed to pass the filter in a tangential mode, whereby the filter surface will be flushed with the fluid. Particles in the fluid will have a much lower capability to travel into the filter material and will instead be kept in the fluid. This technique is often called one in-two out because there is one inlet to the filter and two outlets.
Culinary steam	A designation of steam quality mainly intended for the food industry. Culinary steam is defined in 3A Accepted Practices for the food industry USA. From a practical point of view this steam quality is produced from water of drinking quality and filtered prior to use through a filter with a minimum rating of 5 µm.
D-value	A term used in connection with the killing of microorganisms in an autoclave. The *D*-value is the time needed at a certain temperature in order to reduce the microbiological population by one logarithmic unit, for example to reduce the population from 1000 bacteria to 100 bacteria.
Dalton's law	This physical law states that the pressure of a gas mixture is the sum of the partial pressures of the gases in the mixture.
Dead-end	A way to perform a filtration process. The fluid to be filtered will face the fil-

	ter in a dead-end way and the only way the fluid can leave the system is by passing the filter and thus being cleaned. Dead-end filtration is in some cases called one in-one out filtration.
Dead-legs	This is the expression used for pipes in a process system that are not passed by any fluid. A dead-end will be a potential hazard with respect to hygiene and cleanliness.
Declination phase	The last part of the growth pattern of microorganisms kept in an isolated vessel to which nothing is added or from which nothing is removed. The number of microorganisms will decrease following a geometrical pattern.
Denaturation	A three-dimensional change in the structure of a protein, causing the molecule to lose its function.
Depth-type filter	A type of filter having a certain thickness or depth through which the fluid has to pass. The action of this type of filter is based merely on adsorption of particles in the fluid to the filter material.
Dialysis	A way to perform an ultrafiltration, used in hospital for the treatment of patients and in the purification and treatment of biochemical solutions.
Differential pressure	The difference in pressure on two sides of a component placed in a flow. The differential pressure across a filter is defined as the pressure on the upstream side minus the pressure on the downstream side of the filter. Differential pressure is sometimes called pressure drop.
Diffusion test	A type of integrity test method intended for a filter used for the removal of microorganisms.

Diffusive flow	The flow of gas initially passing through a wet filter material exposed to a pressurised gas. When applying an increasing gas pressure to a wet filter material, the gas will pass the liquid film in the filter due to diffusion. When the gas pressure becomes higher the diffusive flow will increase until the bubble point is reached. At the bubble point pressure and higher, the gas flow through the filter will be a result of both massive gas flow and diffusive flow of gas. Diffusive flow is used in connection with integrity testing of membrane filters used as microbiologically rated filters.
Disinfection	The destruction or inactivation of microorganisms. Disinfection means in practice that the majority of microorganisms, but not all, are destroyed, i.e., sterility is not necessarily obtained.
Duplex filter	A filter system comprising two filter housings, normally used in filtration systems for continuous filtration. Duplex filters are most commonly used where there is a need and desire to have a secured and reliable supply of filtered gas or liquid to a process and in systems where there will be no service needed at other times than those planned.
Endotoxins	Lipopolysaccarides that are building blocks of the outer layer of some microorganisms that upon injection into a patient will give rise to a certain reaction, such as elevated body temperature. Endotoxins can in some cases have a lethal outcome.
Enzyme	A protein with catalytic properties.
Erosion	Damage due to physical forces, for in-

	stance in an unbound filter material that will be subjected to a very high flow rate.
Extractable substances	Substances in a process system or a process component (such as a filter) that can be dissolved and thereby contaminate the product.
Factory steam	See house steam.
FDA	Short for Food and Drug Administration in the United States.
Feed water	The water that is fed into the steam generator in order to be heated during the production of steam.
Fermentor	A vessel designated for growth of microorganisms.
Fiber release	A negative phenomenon based on the fact that the fiber-based construction material of a filter is unbound and thereby released from the filter. The released fibers can then contaminate the filtered and cleaner product. Fiber release will finally result in channeling, another drawback of unbound filter materials.
Filtered steam	Steam that in one way or another has been filtered. Culinary steam is one type of filtered steam.
Filter mechanisms	The overall designation of the various ways by which a filter performs its task of cleaning a process fluid.
Filter aids	Solid and (or) liquid additives that have the ability to aggregate smaller and often colloidal material and form larger and more easily separable particles.
Filtration	The overall designation of the various techniques that can be used in order to clean a fluid by allowing it to pass through a semipermeable material, usually called a filter.

Final filter	A common name for the last filter in a total filter system.
Flocculation	A process by which smaller particles that normally are much more difficult to separate, form aggregates. These aggregates will be much more easy to separate either by sedimentation or by filtration.
Flotation	A separation process in which particles in the fluid are made to accumulate gas bubbles in the fluid at their surfaces. The attracted gas bubbles will then act as balloons and force the particles towards the surface of the liquid.
Flow density	A term often used when dimensioning a filter system. Flow density is calculated by dividing the process flow by the surface area of the filter that the fluid is to pass.
F_0-value	This value is used to describe the killing of microorganisms and is defined as the equivalent time in minutes at a temperature of 121 °C to kill microorganisms with a Z-value of 10 °C. An example is that a sterilisation at 121 °C for 15 min gives an F_0-value of 15 min.
Hot water sanitation	A technique for disinfecting interior surfaces of production equipment by using water at 85–90 °C. The hot water will to some extent dissolve residual product and also have some influence on the killing (inactivation) of microorganisms.
Inactivation	A term used to describe a technique rendering microorganisms harmless.
Inert	Something that is unwilling to react.
Inertial impaction	A mechanism by which a filter performs its action. This mechanism is based on the fact that particles in a fluid flow have inertia, i.e., mass times ve-

	locity. This inertia will allow particles suspended in the flow not to follow the center line of the flow when this is deviating, but instead to follow a tangential path and collide with the pore wall of the filter where the particle is adsorbed.
Integrity test	See non-destructive test methods.
Interaction	Mutual influence.
Ion exchanger	A filter that has the ability to separate ions from a water solution.
Irreversible reaction	A reaction that cannot be forced in the opposite direction. See reversible reaction.
Lag phase	the first phase of the growth curve for microorganisms, in which the microorganisms are extremely active but no cell division occurs.
Logarithmic phase	The second phase of a growth curve for microorganisms. During this phase the cell will reproduce and multiply according to a geometric series, i.e., 1 cell becomes 2, 2 become 4, 4 become 8 and so forth.
Mechanical retention	The filter mechanisms that in a purely mechanical way capture particles. The capture of particles can occur on the filter surface or in the interior parts of the filter material. Mechanical retention is sometimes referred to as sieving.
Membrane filter	A thin paper-like filter material that is constructed from a very strongly bound part, either fibers that are connected together or polymeric cast filter materials.
Microbial retention	The ability of a filter to retain and thereby stop microorganisms from passing a filter material.
Microfiltration	The filtration technique that is used in the separation of smaller particles, so-

	called microparticles, ranging from approximately 90 µm down to 0.01 µm. Sterile filtration is one of the cleanliness levels included in microfiltration.
Nominal rating	A rating of filters used for the separation of dead particles from a process flow. According to the definition nominal rating is ›a micrometer value stated by the filter manufacturer, based on the elimination of a percentage of all particles of a given size or larger‹.
Non-destructive	A type of test method that will not have an influence on the material that is tested. During testing of a filter this type of test technique is utilised by both the manufacturer and the users of filters in order to decide whether or not the filter has retained its integrity.
Polishing filtration	Type of microfiltration that is used in order to separate particulate material that is so small that it is not individually visible to the naked eye. Due to higher concentrations of these particles, the solutions tend to contain a haze.
Polymeric filter	Filter materials constructed from polymeric material that has been cast during manufacturing. Examples of polymeric filter materials are nylon, PTFE and PVDF.
Pore	The small holes penetrating a filter medium through which the fluid has to pass.
Pore size	A commonly used term to describe the action of a filter in service. Unfortunately not a good way to describe a filter since the term pore size might give the impression that the process filter has perfectly circular holes through which the process flow is passing. The term should instead be changed to the

	rating of the filter material. See filter rating.
Porosity	The amount of open space existing in a filter material, i.e., the total area of the pores in a filter divided by the total area of the filter. The higher the porosity the lower the pressure drop across the filter during filtration.
Pre-coat filter	This type of filter is the nearest one can get to a surface filter. The filter has a coating of a porous material that is used for separation of particles in the flow. After filtration the pre-coat layer can be released together with the collected material and allowed to drain out of the system. A new and fresh pre-coat is added and then the filtration process can start all over again. This type of filter is generally used in automatic processes in which the material to be collected by the filter is considered hazardous to the personnel, for example in nuclear power plants.
Pre-filter	A very commonly used description of a filter placed before another filter, often called the final filter.
Pressure drop	See differential pressure.
Pressure decay test	An integrity test method for membrane filters. See pressure hold test.
Pressure hold test	An integrity test method in which the pressure decrease in an isolated volume is measured. The filter to be tested is wetted with a suitable liquid and placed in a filter housing. Pressurised air at a specific test pressure is applied to the wet filter. The air valve is then closed and the diffusion of air through the wet filter material is measured as the decrease in pressure in the isolated volume.
Protozoa	Type of microorganism that is highly

	developed. This type of microorganism is normally found in water environments where they mostly live on decaying organic material.
Pseudomonas diminuta	Former name of the microorganism used in order to characterize sterile filters.
Pure steam	Quality term used for steam and in practice the highest quality. The condensate should correspond to WFI, Water for Injection. This means low ion concentrations, endotoxin content below 25 IU ml^{-1} and often also freedom from particles. The steam is produced in a steam generator manufactured from high degree stainless steel and is equipped with a separation system. The distribution system is also manufactured from high degree stainless steel.
PVDF	Poly vinylidine difluoride.
R_a	Term for surface finish or roughness, given in micrometers R_a.
Reversed osmosis	A separation method that has the ability to allow water molecules to pass whereas other ionic material will be retained by the semipermeable membrane.
SAL	Short for sterility assurance level.
Sand filter	A filter for coarser separation of particles in liquids. The filter can for instance be comprised of a tank filled with sand of different sizes through which the flow is allowed to pass.
Sanitation	Sanitation comprises cleaning followed by disinfection.
Sanitation in place	A form of sanitation taking place online.
Sedimentation	A separation technique based on the attractive force of the earth. This means that if a particle suspended in gas or a

	liquid is large enough, i.e., has a large enough mass, it will be drawn downwards due to the attractive force present and will be collected on the ground or on the bottom of, for example, lakes.
Separation	The overall term for what happens during a separation process, i.e., a technique that has the ability to separate different substances or materials from mixtures.
Septa	The hardware of a pre-coat filter to which the pre-coat material is added in order to form a filter cake through which the fluid to be filtered is allowed to pass.
Serratia marcescens	Test organisms that are used to characterize a membrane filter with a rating of 0.45 μm.
Sieving	A simpler form of separation (filtration) through which the process flow is allowed to pass. A sieve is comprised of a woven material, often a net, that will capture larger particles, whereas smaller particles and soluble material in the flow will pass quite unhindered. Sometimes also a way of expressing the filter mechanism's mechanical retention.
SIP	Short for sterilisation in place, steaming in place and (or) sanitation in place.
Sheet filter	A type of loosely bound filter material, a depth type filter, in the form of plates that are placed in between frames in a filter press. They are most commonly used within the food and beverage industry as well as in basic chemical industry.
Stationary phase	A phase in the total growth pattern of microorganisms in which the major reproduction of organisms is reduced so that the number of cells formed equals

	the number of cells that are dying.
Steam sterilisation	A procedure used for sterilisation using dry saturated steam, either in an autoclave or on-line in a process system.
Steam trap	A component, either manual or automatic, that is mounted in a process system and that releases condensate formed in the process system. This type of component is commonly used in connection with steam sterilisation of process systems.
Sterile	Something that is totally free from living organisms.
Sterile filter	A filter used in the separation of microorganisms in gases and in liquids and which corresponds to a definition stated by the FDA, USA. The definition of a sterile filter is a filter that when challenged with 10,000,000 (10^7) cells of *Brevundimonas dimuta* per cm^2 of filter area, will give a sterile effluent. If a filter corresponds to this definition it will be called a sterile filter and it will be given the rating 0.2 or 0.22 μm nominal rating.
Sterilisation	Techniques used in order to obtain sterility.
Sterilisation in place	Sometimes referred to as SIP. Sterilisation in place means that the actual sterilisation process takes place on-line in the process system, meaning that the process system is kept intact during the sterilisation. A commonly used way to perform sterilisation in place is by using steam at high temperature and pressure.
Sterility assurance level	Traditionally shortened to SAL. This term expresses to what degree of probability the sterilisation process will give a sterile result.
Surface filter	A filter that only collects particles on

	the surface and not in the interior parts of the filter.
Surface finish	The surface finish of a construction material is defined as the deviation from an ideal and totally even state of an object. The deviation is commonly stated in the form of roughness. See R_a.
Titre reduction	A filter rating term which shows the possibility of the filter to reduce the number of microorganisms in a fluid.
Total pressure	The total pressure that is applied to a system. Normally the total pressure of a system will affect the hardware of the system. The filter and other critical components are normally only affected by the pressure differential across the component, and not the total pressure of the system.
Ultrafiltration	A filter technique that is used in order to separate soluble materials from one another. The ultrafiltration membrane will only allow the passage of low molecular weight substances, whereas high molecular weight substances will be retained by the membrane. The high molecular weight substances that are affected by ultrafiltration are for example different proteins (antibodies etc.) and complex carbohydrates.
Unbound filter	Filter materials that are constructed of loosely connected material or totally unconnected material, for example sand filter (multimedia filter), wound fiber filters or glued fiber filters.
USP	Short for United States Pharmacopoeia.
Validation	A commonly used term within the pharmaceutical industry. To validate can easily be defined as to prove that a

	process is doing what it is intended to do.
Vegetative	A state in which microorganisms have the ability and possibility to reproduce.
Virus	A group of microorganisms that depend on the presence of other organisms in order to reproduce
Viscosity	An expression used to describe the ability of a liquid to flow. The higher the viscosity, the harder it will be for the liquid to flow through a process system.
Z-value	A term used in the killing of microorganisms. The *Z*-value is the temperature in °C needed to change the *D*-value of a bacterial population by one log unit.

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