DEPURATION CENTRE MANAGEMENT

COURSE NOTES

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COURSE INTRODUCTION

This course was organised by Bord Iascaigh Mhara (the Irish Sea Fisheries Board) in association with Seafish Industry Authority (U.K.) and ENSA Rennes (France) under a COMETT programme administered by AquaTT UETP Ltd.

The purpose of this course is to train and familiarise people, who are going to build, have just built or are about to modernise a purification plant, in the latest technology available to the industry and also to train people so that they can effectively run a depuration centre.

In general the course aims to:

- Give participants practical experience of different systems prior to committing themselves to a particular system.

- Enable participants to compare systems.

- Provide a minimum training qualification to run an establishment which is essentially a facility to guarantee health and safety of products to the consumer.

- Provide an overview from bacteria testing to plant lay out etc.

The structure of the course is in modules, morning and afternoon sessions, containing lectures on relevant topics and then practical sessions. Throughout the course the teaching principal of hear, see, do is followed. Where the participants will first be given a lecture on a subject then observe a practical demonstration and then finally carry out the procedure themselves.

The people presenting the course have had various experience with purification and shellfish handling and the practical sessions present the time for more informal exchange of ideas.

The practicals were organised on a rotating basis with the course participants being divided up into groups with one group carrying out a practical or a portion of a practical at one stage and with another group doing the same practical at another time. This is to ensure that everyone has the opportunity to have hands on experience with all the equipment.

The course is structured so that the sampling and monitoring techniques are covered first. This is because during the rest of the course the participants will be expected to take various samples and readings which will then be complied and analysed on the last day. This is also designed to get the people into the very important practice of record keeping.

(At this stage the course personnel are introduced and a short tour of the facilities were the course is taking place is carried out.)

GENERAL INTRODUCTION TO SHELLFISH PURIFICATION

Shellfish from certain areas can be unfit for direct human consumption (i.e. cannot be eaten raw) due to them containing harmful bacteria or viruses.

Before these shellfish can be eaten (raw) they have to be cleansed or purified.

Purification is not required just to meet National or EC regulations and directives. People can die or become seriously ill from eating contaminated shellfish. It is up to the person running the depuration centre to see that this does not happen.

The process of purification (or depuration) is essentially a natural process for the shellfish if it is given certain conditions. (There are very new experimental processes where the shellfish is irradiated to kill harmful bacteria and viruses, similar to those being used in the fruit and vegetable business in some countries).

The term shellfish can apply to numerous types of animals from squid to crabs and mussels etc. For the purpose of this course the term shellfish will generally refer to bivalve molluscs i.e. mussels, oysters, clams, cockles and scallops (the class of animals in the phylum mollusca that have two shells).

As the name bivalve suggests the animal has two shells made of hard calcarious material which encloses and protects the soft inner tissues. A very generalised diagram of a bivalve shellfish is below. The main components are the gonads (the tissue that produces the eggs and sperm), adductor muscle(s), foot, mantle, gills, syphons and digestive system including digestive gland (fig.1).

For the most part the bivalves (after a brief planktonic stage) are mainly sessile (i.e. they remain in the one place for the rest of their lives) though some like scallops can display some active movements over distance. As a result of them being sessile if their environment changes they have limited options. If it is a hostile change the shellfish can withdraw into the shell and remain closed. If the change is more long term the shellfish has to live through it or try and move, if it cannot it will die.

All the bivalves are filter feeders, that is they filter out food (small particles and plankton) form the water in which they live by means of syphons and (or) gills. This process is relatively non-selective in that the shellfish will take in everything in the water both good and bad (note what is harmful for man when he eats the shellfish may not be bad for the shellfish). The particles are then passed through the digestive system (gut) where the suitable food particles are digested and the unsuitable or waste particles, are voided in the form of faeces.

Bacteria and viruses can be part of the water borne particles that the shellfish would consider as food and are therefore ingested. Once they enter the digestive system several things can happen. They can; remain in the gut and eventually be voided with other faecal material; stick or become associated with the gut wall; be ingested by cells; pass through the gut wall and enter the haemolymph (or blood) of the shellfish or enter the other tissues of the shellfish such as muscle etc. (fig. 2).

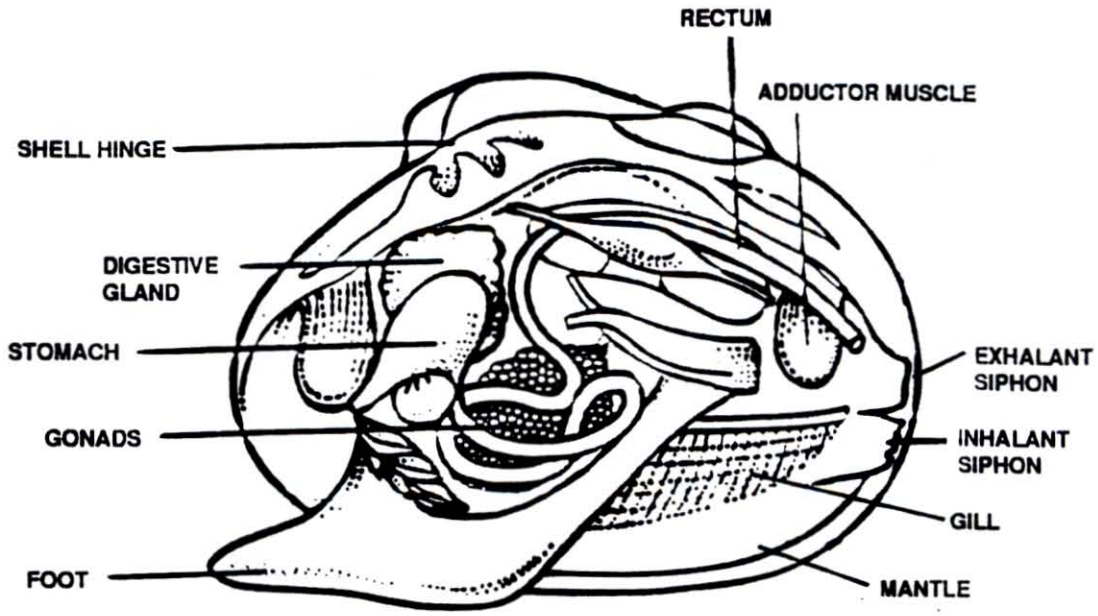


Figure 1. Generalised diagram of a bivalve shellfish.

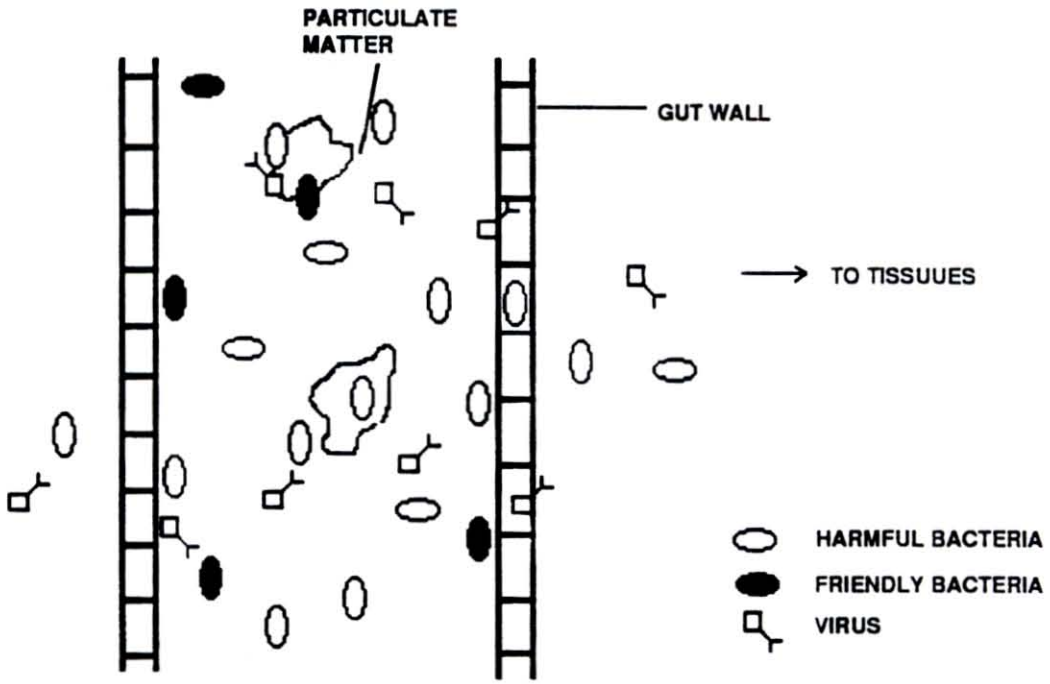


Figure 2. Bacteria and viruses in the gut.

In the gut of the shellfish there are resident bacteria that are usually harmless to man or termed "friendly" or non-pathogenic bacteria. In very general terms if there are bacteria and viruses in the gut they can be cleansed very quickly from the animal (within 24 to 48 hours). If, however, they are in the haemolymph it will take longer to clear them and if they are in other tissues it can take over eight weeks.

Different types of bacteria and viruses can also take different lengths of time to clear. E.Coli and faecal coliforms clean out relatively fast and it is these organisms that normal depuration is designed to eliminate. There are other types of harmful bacteria and viruses that standard commercial depuration times will not eliminate.

In the EU all shellfish growing waters have to be classified. This is done by taking a sample of the shellfish and analysing its flesh for bacteria (not the water as was done under the Shellsan programme).

There are four classifications:

A. less than 3 f. coliforms per gram of flesh
B. from 3 to 60 f. coliforms per gram of flesh
C. from 60 to 300 f. coliforms per gram of flesh
D. above 300 f. coliforms per gram of flesh

In general terms shellfish for direct human consumption from A class waters can be eaten directly. Those from B need to be purified. Shellfish from C areas have to be relayed for a minimum period of 8 weeks in an A area (a suitable distance away from existing shellfish to avoid cross contamination) and those in D areas are not suitable for human consumption (though there are moves to permit this if they are relayed in areas of A or B classification for over six months).

Shellfish from A, B and C can go directly for processing (needs to be an approved processing method sufficient to kill all bacteria and viruses) without purification.

Unfortunately the above classification system has two basic flaws. Firstly one in twenty samples can be over the agreed limit and the area still keeps its classification. So it is possible for one in twenty loads of shellfish from an A area to have bacteria levels that would require it to be purified. This implies that every batch of shellfish should be tested post harvest to ensure that it is fit for human consumption.

Secondly, the above classifications are based on bacteria levels. These levels were picked to correspond to the possible risk of viral contamination. For unfortunately viral testing is very expensive and at present not really practicable in commercial terms. So an A area should not have any risk of viral contamination; B, a slight risk; C will most likely have viral contamination but it is hoped that by relaying for over eight weeks the viruses in the flesh etc. will be cleansed from the shellfish.

A well run purification system will cleanse shellfish with bacterial levels of E. coli

well in excess of 60 E. coli per gram. However, it may not get rid of a virus if it is in the haemolymph or other tissue of the shellfish.

The main problem lies in the fact that it generally takes about 10,000 viable bacteria to be ingested to cause an illness whereas it only takes one viable virus.

This is why it is imperative that a depuration plant manager should not only rely on the bacteria readings of the shellfish leaving his system but should also know the history of the area and the contamination levels of the shellfish at the point of harvest.

All methods of shellfish purification (except irradiation) rely on the shellfish naturally ridding themselves of the bacteria and viruses. Put quite simply if shellfish do not actively filter they will not purify no matter how expensive or fancy your sterilisation apparatus may be.

Shellfish function best in certain optimum conditions, these conditions may vary from species to species and even from within the same species depending on how it was grown and where it was from.

Factors shellfish require to filter actively are:

- clean non toxic water
- the correct salinity
- suitable oxygen levels
- correct temperature range
- no stress or disturbances
- correct light intensity
- ability to be able to physically open
- though not essential, food particles in the water will stimulate active filtration.

Depuration, by definition, is to promote the elimination of waste products from a body, so most depuration systems are just methods of intervention by man to enable shellfish to cleanse themselves. These methods fall into two categories:

- Relaying in clean water.
- Intensive purification.

Intensive purification can be further subdivided by systems.

- Single layer system
- Stacking system
- Multilayer system
- Up-welling and down-welling
- Aeration

Whichever system is used a well designed system, with correct operation, should try and attain as many of the ideal conditions as possible to enable the shellfish to

be actively filtering. In short the purification system should aim to keep the shellfish "happy":

- able to open
- able to filter
- not to be disturbed
- not to be stressed

Most of the newer systems are termed closed recirculating systems (i.e. the same water is treated and recirculated for the period of the purification cycle) though flow through systems, batch water change systems such as "aeration systems" are also in use. (All of these systems will be dealt with in greater detail later).

Components of purification systems.

In general terms purification systems have the following components (fig. 3):

- Intake point where clean sea water is taken into the tanks
- Tank for holding water
- Containers for holding shellfish
- Recirculation pump
- Sterilisation medium to remove bacteria etc.
- Oxygenation system, via weirs or spray bars
- Out-fall where waste water and material is removed from the tank.

In addition most depuration plants are centres where shellfish are collected from various growers or fishermen, washed, graded, packed and distributed. So the buildings are designed to not only purify shellfish but to carry out other functions. Again this will be dealt with in more detail in the relevant sections.

Though in all aspects whether harvesting, handling, purifying or despatching shellfish one crucial fact should be remembered that makes the shellfish industry virtually unique in the food business. This fact is that you are dealing with live animals and usually (unless the shellfish have been processed) the consumer expects them to be alive and fit to be eaten in a raw state for several days after they have been purchased and taken home. So it is up to the depuration centre manager to make sure any product that passes through his plant meets those requirements for unfortunately one bad mistake could cause hundreds of people to become ill or worse.

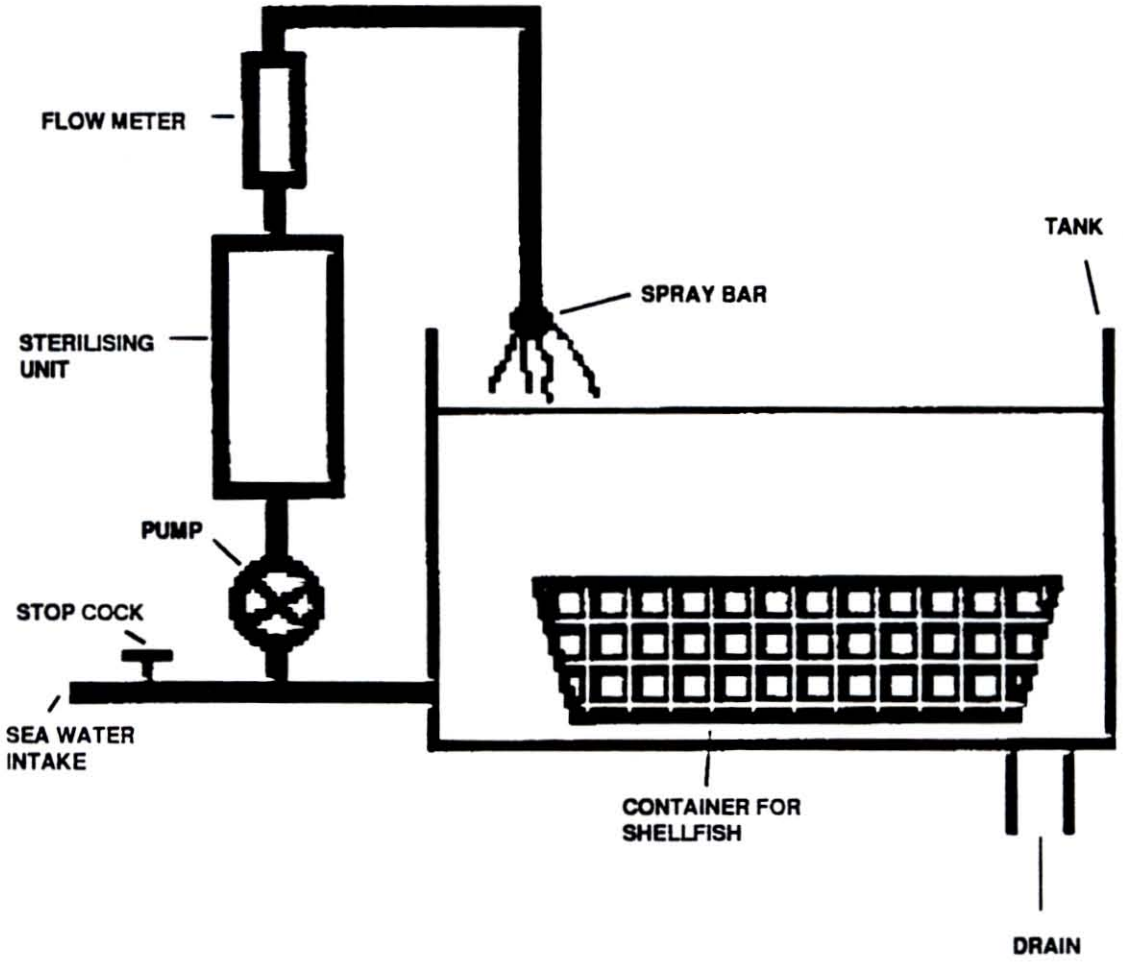


Figure 3. Generalised purification system.

INTRODUCTION TO BACTERIA AND VIRUSES

Definitions:

Germ: A germ is the vernacular for a microorganism especially one that causes disease.

Microbe: A microbe is any microorganism especially a bacterium.

Microorganism: A microorganism is any organism which cannot be seen with the naked eye ie. it can only be visualized using a microscope.

Microorganism group comprises:

- Bacteria
- Viruses
- Fungi (yeasts and moulds)
- Protozoa
- Microscopic algae

Although there are several different types of microorganisms (as outlined above), the ones which are of interest in assessing efficiencies of mollusc depuration and hygiene related to mollusc production, are bacteria and viruses.

Bacteria and viruses are tiny organisms which cannot be seen with the naked eye. Both groups are capable of causing disease in humans ie. are pathogenic, given the right opportunity.

The main differences between bacteria and viruses is their mode of reproduction and their size. In the first instance, viruses are obligate parasites and must invade a living host cell (whether animal, plant or bacterial) in order to reproduce. Bacteria however, reproduce independently, usually by simply dividing in two.

The second differentiating factor is size. Bacteria are generally much larger than viruses ie. between 100-1000 times larger. The fact that bacteria can easily be seen using a light microscope (with fairly low magnification), while viruses can only be visualized using a high magnification electron microscope clearly demonstrates this.

Characteristics of bacteria and viruses.

Characteristics	Bacteria	Viruses
Reproduce independently	+	-
Obligate parasite	-	+
Unicellular	+	+
Mobile	+/-	-

Bacterial and viral contamination of molluscs

Molluscs are filter feeders and in the process of filtering large volumes of water, they collect and concentrate the bacteria and viruses present in the water. When this water is sewage contaminated, there is a high risk that disease inducing (pathogenic) bacteria and viruses will be present and will be ingested by the molluscs during feeding.

There are roughly two possible fates of the bacteria and viruses, often trapped in soil and other particulate matter, ingested during filter feeding;

1) They become trapped in the gills and other tissues
2) They enter the digestive tract of the mollusc and are either destroyed by the animal or are passed through its gut and eliminated with faecal material.

Bacterial contamination

Because bacteria are comparatively large, they generally do not become too deeply embedded in tissues and can be eliminated relatively easily by relaying or depuration. Obviously if large numbers of bacteria are involved, the process of elimination can be lengthy.

Pathogenic bacteria normally found in the gut of man and other warm-blooded animals do not normally grow in molluscs because of their alien environment, indeed many die and others become injured as a result. Even when injured, the bacteria may still be able to grow and reproduce if conditions change. Thence whether injured or completely intact, these bacteria have the potential to cause disease if the contaminated animal is subsequently eaten by humans.

Viral contamination

Unlike bacteria, viruses present in sewage are totally dormant and cannot grow or reproduce in molluscs. However, since viruses are much smaller than bacteria, they can enter muscles and other organs of the mollusc as well as tissues, where they become deeply embedded. When this happens, it is very difficult and almost impossible to eradicate the virus even by relaying and depuration.

When there are high numbers of faecal coliforms (indicators of sewage contamination) present, it can be assumed that there is a high risk that there are also viruses present. The higher the number of bacteria present, the higher the risk will be of viral contamination of the mollusc.

EC legislation has laid down an upper limit of 600 fc/g above which molluscs cannot be harvested. Above this limit there is evidence to suggest that viruses will be so embedded in molluscan tissues that they can never be completely eliminated.

Factors determining bacterial growth and reproduction

1. Temperature

In general, bacteria can only grow between temperatures of 5°C and 65°C. Below 5°C, bacteria remain alive but cannot grow hence the use of refrigeration in food preservation. Above 65°C, bacteria generally cannot grow and indeed most bacteria are killed at temperatures above 70°C.

2. Water Availability

All living organisms including bacteria, require water for growth, repair, reproduction etc. Curtailing water availability limits bacterial growth and is often used as a method of preserving food and extending shelf life eg. dried milk products etc.

3. pH

pH refers to the acidity or alkalinity of a solution and is measured on a scale of 0-14. The lower end of the scale indicates very acidic solutions while the upper end indicates very alkaline solutions. A pH of 7 indicates a neutral solution which is neither acid or alkaline. Most bacteria grow best in a narrow range of pH, between 6.5 and 7.5. This is the reason why some foods eg. marinades are preserved by acidic solutions such as vinegar.

4. Gaseous Environment

Many bacteria require the presence of oxygen for growth and these are referred to as aerobes. Other bacteria cannot grow when oxygen is present and these are known as anaerobic bacteria or anaerobes. Some bacteria fall between these two categories and require varying gaseous mixtures eg. carbon dioxide and nitrogen gas, for growth.

5. Nutrient Availability

Like all living organisms, bacteria require a nutrient/food source from which they derive their energy. Some bacteria have fairly simple nutrient requirements while others have more complex ones. The major nutrients required are carbon, nitrogen and sulphur and these are derived from a variety of sources eg. carbon from carbon dioxide in the air, nitrogen from soil, sulphur from inorganic compounds such as hydrogen sulphide etc.

Factors determining viral multiplication

1. Availability of a suitable living host.

Viruses are inert and dormant outside living host cells. However, once a virus enters a host cell, it becomes active and multiplies.

Viruses can be divided into 3 main categories;

(a) animal viruses
(b) plant viruses
(c) bacterial viruses (bacteriophages)

Animal viruses can thus only invade and cause disease in animals while plant and bacterial viruses can only invade plant and bacteria, respectively.

This division of viral groups is due to the presence of "recognition" or receptor parts on the surfaces of viruses. For example, an animal virus recognizes an animal cell via its receptor parts. These receptors allow the virus to bind to and invade the animal cell.

To date, research has found that viruses of warm-blooded animals, eg. man, do not invade and multiply in shellfish. Therefore, if a mollusc becomes contaminated via sewage with a virus from human intestines, it cannot invade and multiply in this mollusc, simply because it doesn't have the receptors to recognize the mollusc cells.

Microbes, seafood and food-borne disease

Outbreaks of diarrhoeal food poisoning and other illnesses associated with the consumption or handling of shellfish adversely affects public confidence in the shellfish industry. It is for this reason that people involved in the industry must understand the mechanisms by which these illnesses occur and control the harvesting and placing on the market of their produce. If the market for live mussels, oysters and other bivalve molluscs expands in the future, health risks associated with their consumption will also increase unless the proper controls are put in place and adhered to by the industry.

Although there are many health risks associated with the consumption of raw or lightly processed bivalves, as listed below, those addressed here relate only to disease caused by bacteria and viruses.

<u>Diseases transmitted via seafood</u>

- Bacterial: Infections
  Intoxications
- Viral
- Parasitic
- Intoxications due to chemical poisons
- Intoxications due to biotoxins
- Undetermined infectious agents

<u>Bacterial food-borne disease</u>

Bacteria produce 2 types of gastro-enteritis. The first type is an infection where the bacterial pathogen ingested in the food penetrates the intestinal wall of the person,

multiplies and this leads to diarrhoea, vomiting and other symptoms. Bacterial pathogens which operate in this way include the salmonellae and shigellae (which can cause dysentery).

The other type of disease is where the bacterium produces a toxin which causes the symptoms. In many cases this toxin unlike the bacterium which produces it, is heat resistant. Thus even if the mollusc is cooked, the toxin may still be present and can cause the disease. It is important to note however that high numbers of the bacteria must be present before the toxin is produced so it is possible to prevent the disease by controlling the growth of the bacteria eg. by refrigeration.

Bacteria which produce toxins include Vibrio cholerae (causing cholera), Clostridium botulinum (causing botulism) and Staphylococcus aureus (a bacterium present on the skin of humans which causes food poisoning).

The onset of symptoms in bacterial infections and intoxications range from a couple hours to 2-3 days although there are some exceptions to this (eg. a salmonellae infection may take 2 weeks before taking effect).

Viral food-borne disease

The most well known food borne viral infection is infectious hepatitis, caused by the Hepatitis A virus. This virus causes a liver and kidney infection as well as the usual food poisoning symptoms. The onset of these symptoms is 15 to 45 days.

Other viruses which can cause food poisoning include the Norwalk and Rota viruses. The onset of symptoms for these viruses is considerably shorter and ranges from 1-3 days.

The diseases mentioned above and their associated potential health problems clearly underline the necessity of purification of molluscs (whether by relaying or depuration) where there is an unacceptable risk to the consumer.

Sources of microbial contamination, infection and intoxication in seafood

Molluscs, like other foods can be contaminated with bacteria and viruses from a variety of sources as outlined below.

1. **Workers**,

   Because the human body is the home for so many bacteria and may also be infected by a various viruses, there is an inherent risk of human contamination of the molluscan product. Many people carry a bacterium, S.aureus on their skin which if present in high numbers in a food can cause food poisoning. Obviously, the shell of bivalves offers protection against this type of contamination however, it is important to note that this potential risk does exist.

2. Equipment and processing environment

As outlined earlier, bacteria require a food source, water and higher temperatures to grow. If the equipment is not properly cleaned and temperatures are high, then the risk of contamination will be much higher. It is important therefore, to ensure that the cleaning operation is effective and the water used, potable, in order to minimize any dangers of contamination.

Equipment should be designed for rapid, efficient handling and be suitable for easy and thorough cleaning.

The processing environment should be clean, pest-proofed and temperature controlled as far as possible. Structurally, it should be in good condition with smooth, easily cleanable, corrosion-resistant surfaces and proper drainage facilities.

3. Sewage pollution of the aquatic habitat

Sewage contains very high numbers of bacteria and often viruses and is thus the single most important source of contamination of live bivalve molluscs. Where its untreated discharge cannot be controlled in molluscan growing waters, there will be an extremely high risk of contamination of the product.

When governmental authorities monitor these waters bacteriologically, then the actual risks can be assessed. It is important therefore, for molluscan producers to be able to understand the nature of these bacteriological assessments, interpret the results and make an informed assessment of any risks relating to their own product.

However, where sewage contamination occurs, purification (whether by relaying or depuration) of the product is a way of guaranteeing its safety.

BACTERIA SAMPLING PROCEDURES AND TESTING

Microbial analysis and molluscs

Why test?

Regular bacterial testing is an essential factor in the production and placing on the market of live bivalve molluscs for direct human consumption. Testing can be used as an effective monitoring device to ensure that animals are free from disease-inducing (pathogenic) bacteria and viruses.

1 Nature of the beast

Molluscs have been singled out among shellfish groups for rigorous scrutiny by health authorities because of their inherent nature to filter-feed, thus enabling them to accumulate high numbers of pathogens from sewage-contaminated waters. They are potentially a significant source of gastro-enteritis and thus a real public health hazard if their production is not monitored.

2 Product safety

Direct and indirect sewage contamination of molluscs and molluscan growing waters can be measured using the Faecal coliform test. The test is crucial in determining the efficiency of purification and relaying procedures to ensure product safety and the elimination of potential pathogens from the mollusc flesh.

3 Legislation compliance

New legislation effective from January 1st 1993, lays down a strict code of practice for the production and placing on the market of live bivalve molluscs. The Directive concerned. 492/91/EC, in summary dictates that final product must contain less than 3 Faecal coliforms (FC) per gram, product containing 3-60 FC's must be purified, and product containing 60-600 FC's must be relayed.

Council Directive (91/492/EEC) - Health conditions for the production and placing on the market of live bivalve molluscs

Under this directive, two of the annex chapters, 1 and 5, outline the microbiological requirements for mollusc product.

Chapter 5

Requirements concerning live bivalve molluscs.

(a) Must contain less than 3 FC or 2.3 E. coli per gram.

(b) No salmonella in 25g.

(c) In absence of virus testing procedures, health checks must be based on faecal bacterial counts.

Chapter 1

Conditions for production areas.

(a) Must comply with requirements in chapter 5.

(b) Areas >3 fc/g and <60 fc/g must purify.

(c) Areas >60 fc/g and <600 fc/g in 90% samples must relay for >2 months.

(d) Areas >600 fc/g cannot harvest.

NOTE

1 Relayed and purified product must comply with (a).
2 Sample size 100g.

Microbial indicator systems

The use of "Indicator Bacteria" in shellfish sanitation dates back to the beginning of the century. Since then these indicator organisms, have been used routinely in demonstrating sewage contamination and potential health risks from shellfish product. Given the technical difficulties in isolating certain pathogens, as well as the wide range of pathogens possibly present in a molluscan product, use of indicator bacteria represents a practical compromise in determining a possible health hazard from such product.

The indicator organisms most widely accepted and outlined in legislation are the faecal coliform group of bacteria including E.coli. These bacteria are found in the intestines of warm blooded animal (like enteric pathogens) and are easily isolated in the laboratory.

Characteristics of coliforms and faecal coliforms

1. Coliform is a general term given to bacteria which inhabit the intestinal tract of man and other animals without causing disease.

2. The term "Coliform" is generally limited to those bacteria which when cultivated in the lab can ferment lactose (generally at 37°C - body temp.) within 48 hours. By and large coliforms are represented by 4 genera belonging to the bacterial family of Enterobacteriacea, ie. Escherischia (E.coli), Klebsiella Enterobacter and Citrobacter. Some strains of Edwardsiella, Arizona, Hafnia, Pantoea, Serratia, Aeromonas however

ferment lactose though usually not within 48 hours.

3. Faecal coliforms are defined as coliform bacteria which can ferment lactose at a cultivating temperature of between 44° and 46°C (usually 44.5°C). A test for faecal coliforms is essentially a test for E.coli, although some Citrobacter, Enterobacter and Klebsiella strains can in fact ferment lactose between 44 and 46°C. 75-95 % of faecal coliforms in shellfish growing waters are E.coli.

4 E.coli unlike other faecal coliforms is always found in the human intestine and is excreted in vast numbers in the faeces. It's presence, therefore, is a good indication of faecal pollution.

Faecal coliforms - effective indicators ?

The requirements of a perfect indicator are outlined below. Faecal coliforms, including E.coli, fulfil many of these requirements. Thus, in theory, they should give a relatively good indication of the presence of bacterial pathogens.

- History of constant association with pathogen.
  Faecal coliforms originate in the gut where many pathogens originate.

- Distinguishable from other organisms/agents.
  Faecal coliforms are a distinct group of bacteria easily distinguishable from other bacteria.

- Always present when pathogen present.
  Faecal coliforms are normally present with other enteric bacteria including pathogens, enteric viruses however survive longer.

- Possess equal/similar growth rates to pathogen
  Faecal coliform possess similar growth rates to bacterial pathogens but not viral pathogens.

- Be easily and rapidly detectable.
  The isolation of faecal coliforms is easy and economical and detection time varies from 24-72 hours.

Viral indicators

Faecal coliforms are fairly effective in indicating many bacterial pathogens, but, their use as indicators for viral pathogens is dubious. Much research is currently being done on finding a suitable virus to indicate the presence of viral pathogens. However, because of technical difficulties in growing viruses, research is slow and it may be some years before a suitable indicator virus is identified and accepted. Several viruses have been proposed as indicators, mainly those which infect the gut ie. enteric viruses* and also the group of viruses which infect bacteria ie. bacteriophages**.

* One enteric virus which has been suggested as an indicator is Poliovirus. Most people are vaccinated against polio by means of an oral vaccine containing a live but non disease-producing (non-pathogenic) form of the virus. At any given time then, there will be people excreting the virus and its presence in water could be used to indicate faecal contamination which possibly contains viral pathogens.

** Bacteriophages (also known as Phages) are viruses which infect certain bacteria.

Sampling requirements

A <u>Water</u>

(a) Sample container

Water samples for analysis must always be collected in a sterile container. Containers which can be sterilized easily are most suitable. Plastic containers are recommended because they are unbreakable and re-usable. Many laboratories will supply sterile containers for water samples however, it is possible to sterilize your own if required. To do this, simply pour boiling water into the container, around the neck and also into the lid. Quickly, but thoroughly rinse and pour out the water, replacing the lid tightly without touching either the neck or the inside of the lid. The container should now be sterile and ready for sampling.

(b) Container capacity

Capacity of the sample container is important. Large samples are cumbersome and unnecessary while small samples may not be representative of the water under investigation.

A 500 ml capacity container will give a sample volume which allows for all usual bacterial isolation procedures. If samples are required for viral testing however, much larger volumes are required ie. up to 100 litres may be needed. 500 ml plastic (polypropylene) bottles which are leak proof and can be sterilized are available commercially for about £2 each.

(c) Methylated spirits, cotton wool, flame

Water outlet taps are often contaminated with bacteria. It is important therefore, to ensure that the tap/outlet pipe from which the water sample is being taken is clean. If the tap itself isn't clean then the water sample analysed may not represent the bacteria present in the water but instead those present on the tap.

The area around the water outlet should be sterilized by swabbing first using cotton wool or clean cloth dipped in meths. and then flaming. If the outlet is

flammable, then it should simply be cleaned using meths.

B Molluscs

(a) Sterile, heavy duty plastic bag

The container used for sampling here ideally should be sterile to ensure that it doesn't contaminate the mollusc flesh. Sterile plastic bags are widely available commercially and are very cheap. If however, sterile bags are not available then the molluscs should be packed in such a way as to prevent opening, loss of fluid and contamination. It is important not to use open or partially opened molluscs in this instance as the flesh could already be or could become contaminated.

(b) 1 kg to ensure 100g flesh

Under Directive 492/91/EC, a sample size of 100g of mollusc flesh and intra-valvular fluid is required. To ensure this is obtained in the laboratory, a 1 Kg sample of live molluscs should be taken.

(c) Remove mud etc. from shell

Only animals with intact shells should be taken. In addition, their shell should be free from mud etc. and if they are not then it should be removed carefully.

Packing requirements

A. Water

The lid should be secure on the water container to prevent leakage and possible contamination. The container should be placed in an insulated bo eg. polystyrene, if possible with ice packs to ensure the water is kept cool.

B. Molluscs

Molluscs should be packed tightly in the bag in such a way as to prevent opening and loss of fluid. They should also be placed in a clean insulated box and kept cool using ice packs.

Critical factors in sample transportation

1. Keeping samples cool ie. below 10°C.

It is important to keep samples as cool as possible (without freezing) to ensure that bacteria therein do not multiply and affect the true test results. This is particularly relevant during the summer months.

2. Transportation time is crucial.

(a) For water samples this is particularly important as faecal coliforms and other bacteria only survive a few hours to a couple of days in water. If the test water arrives in the laboratory later than 8 hours after sampling, it probably won't be representative. In fact, the results obtained will be relatively useless in determining the extent of contamination of the water body sampled. Ideally samples should be delivered to the laboratory within 6 hours of sampling.

(b) Mollusc samples should also be transported rapidly to the laboratory for several reasons;

(i) Although faecal coliforms survive better in mollusc flesh than in water, they are nevertheless in a stressed environment and may die as a result.

(ii) If the sample is transported at room temperature, faecal coliforms may be able to multiply, increasing in numbers to give an over representation of the level of contamination. As a general rule then, mollusc samples should be transported to the laboratory as soon as possible after sampling and definitely within 8-10 hours of sampling.

3. Ensuring no damage occurs.

Obviously if a sample becomes damaged during transport, it may become contaminated. Analysis of such samples will be futile.

4. Correct delivery.

Samples should be delivered directly to the laboratory where they will be refrigerated briefly if they are not tested immediately. They should never be simply abandoned at the laboratory door.

Microbiological test methods

There are many different methods employed to estimate bacterial numbers in samples. Most of the traditional methods are based on a direct count to estimate bacterial numbers ie. the bacteria are cultured on a selective medium and their growth can be seen as colonies on a culture plate which can be easily identified and counted. The newer rapid methods tend to employ techniques of enumeration which reflect bacterial activity or presence indirectly. Many of these techniques measure some aspect of bacterial metabolism which can then be used to estimate numbers present eg. in the Malthus system, the change in impedance produced by by-products of bacterial metabolism is measured and used to estimate bacterial numbers.

Systems like the Malthus give rapid results and are being used increasingly in

Ireland for estimating faecal coliform and other bacterial counts.

In general however, the cost of such systems together with the level of expertise required to install them is prohibitive. Most laboratories here still use traditional methods and these are the methods which are used in the workshop practicals.

The two most common methods used for faecal coliform detection and enumeration are:

1. Membrane Filtration Test

2. Most Probable Number (MPN) Test

Membrane filtration test

This method is used only for water and liquid samples. It is rarely employed to enumerate faecal coliforms in molluscs as the flesh (unless very diluted) remains on the filter making visualization of the bacteria impossible.

Methodology involved

Step A. - Total coliform test

a) The water sample is passed through a filter of small pore size (-0.45 um) which retains the bacteria.

b) This filter containing the bacteria is then placed on a growth medium selective for total and faecal coliforms (eg. MacConkey Lactose medium) and incubated at 37°C.

c) The filter absorbs the growth medium allowing any coliforms present to grow and reproduce.

d) After 24 hrs, the filters are examined for colonies. Since the medium used is selective for coliforms the colonies present are taken as the Total Coliform count.

Step B. - Faecal coliform test

e) Colonies which grow on the filter are subcultured into two broth mediums

   (i) Brilliant Green Bile Broth (BGBB) - to indicate acid and gas production.
   (ii) Tryptone Water (TW) - to indicate indole production.

f) After transferring the colonies into these media they are incubated at 44°C for 24 hours.

When acid and gas from BGBB and indole from Tryptone Water are produced, the test is positive for faecal coliforms.

For further confirmation of E.coli and differentiation between different faecal coliforms species, further tests are required. These tests, collectively called IMViC tests, are not normally carried out because of the time and labour involved.

Most Probable Number (MPN) test

This method can be used for both mollusc and water samples and although it is laborious, it is relatively simple to do and results are easily readable.

It is a statistical estimating method based on three assumptions;

1) Bacteria are distributed evenly in all parts of a sample (this can be helped by good mixing or homogenizing of the sample);

2) If a sample is diluted sufficiently, eventually, a point will be reached where there will be no bacteria left;

3) The bacteria are grown in a medium where their growth is easily detectable (In MacConkey Broth, coliforms produce gas in tubes and acid which changes the colour of the medium).

Basis of test

A. In the test, different volumes of the sample (of the same dilution) are put into a series of 3 sets of tubes of different volumes and strengths.

B. These 3 different volumes have been precalculated to give a constant dilution factor, and tables have been prepared to calculate the results.

C. If the sample gives a positive result in all of the tubes, then it has not been diluted sufficiently to give a readable result.

D. If the sample is diluted enough, then there will be both positive and negative tubes, ie some tubes will have shown growth and therefore the presence of bacteria and others which have not. If there are negative tubes present then the pattern of tubes can be translated into a readable result.

Methodology

1. 3 Tube Method (for molluscs)

A. Total coliforms:

(i) The mussel flesh is diluted and mixed (homogenized).

(ii) The diluted flesh (diluent), is then added to the 9 MPN tubes as

shown below ie.

a) 10 ml is added to each of the 3 tubes containing 10 ml of Double Strength MacConkey Broth
b) 1 ml is added to 3 of the tubes containing 10 ml of Single Strength MacConkey Broth
c) 0.1 ml diluent is added to the remaining 3 tubes containing 10 mls of Single Strength MacConkey Broth

Tubes numbers

--- add 10 mls diluent 1 2 3 (containing 10 mls Double Strength)
--- add 1 ml diluent 4 5 6 (containing 10 mls Single Strength)
--- add 0.1 ml diluent 7 8 9 (containing 10 mls Single Strength)

(iii) After incubation at 37°C for 48 hours, any growth in the tubes will be indicated by a colour change in the medium (for MacConkey Broth the medium will change from purple to yellow and become turbid) and by the presence of a gas bubble in the inside (Durham) tube.

Interpretation of results

This can only be done using the 3 Tube MPN Tables. Please consult these tables in the appendix.

Examples

1. If only one tube is positive, say tubes 1, 2 or 3 then the result will be 1,0,0 which equals 3 coliforms per gram.

2. If one tube is positive in each row of the dilutions eg. tubes 1, 4 and 7 then the result will be 1,1,1 which equals 11 coliforms per gram.

3. If all tubes ie. 1-9 inclusive, are positive then the result will be 3, 3, 3 which equals > 1100 coliforms per gram.

B. Faecal coliforms

1. This method is identical for that outlined for Step B in the Membrane Filtration Test except that instead of taking a colony from membrane filter, a 1ml volume is taken from the positive tube.

2. 5 Tube Most Probable (MPN) Test (for water)

The basis of this test is identical to the 3 Tube Method but the volumes and numbers of tubes are different. The water sample is added to 11 tubes as outlined below. The water sample can be diluted if high numbers of bacteria are expected.

(a) 50 mls of water is added to 1 tube containing 50 mls of Double

Strength MacConkey Broth

(b) 10 mls of water is added to each of the 5 tubes containing 10 mls of Double Strength MacConkey Broth

(c) 1 ml of water is added to each of the 5 tubes containing 5 mls of Single Strength MacConkey Broth

Tube numbers

--- add 50 ml water	1					(containing 50 mls D.S)
--- add 10 ml water	2	3	4	5	6	(containing 10 mls D.S)
--- add 1 ml water	7	8	9	10	11	(containing 5 mls S.S)

Interpretation of results

This can only be done using the 5 Tube MPN Tables. Please consults these tables in the appendix.

Examples

1. If only Tube No. 1 is positive then the result will be 1, 0, 0 which equals 2.3 coliforms per 100 mls

2. If one Tube is positive in all rows eg. Nos. 1, 3 and 10 then the result will be 1, 1, 1 which equals 6 coliforms per 100 mls.

3. If all tubes are positive in all rows then the result will be 1, 5, 5 which equals >180 coliforms per 100 mls.

Variations in methodologies

The methods outlined above are those recommended by the International Commission on Microbiological Specifications for Food (ICMSF) and the American Public Health Association (APHA). There are however a diverse array of methodologies outlined in the literature for both the MPN Test and the Membrane Filtration. These methodologies merely differ from those outlined here, in the bacterial growth medias used as well as the temperature and time factors. As well as the differing methodologies for these tests there also many other tests used for enumeration of coliforms and these are also outlined in the literature.

SETTING UP A LABORATORY

Depending on the size of an operation involved in shellfish production, particularly one involved in shellfish depuration, a company may consider setting up a laboratory to carry out basic microbiological tests of product on site.

Several factors must be considered before making a decision, however the main question is whether the need justifies the expenditure.

Areas to consider before setting up

1. Level of testing; does it make financial sense?

2. Product specification relating to quality

Does your product specification at present involve limitations on bacterial numbers, specifically relating to faecal coliform and/or salmonella presence in order to

   a) satisfy legal requirements ?

   b) satisfy customer specifications ?

3. Harvest water quality; is it likely to cause a problem ?

   If you are harvesting from an approved zone do you envisage

   a) Any changes in classification of your harvest area in the immediate future ?

   b) Any problems in proving that your product has in the past and will continue to meet the requirements of product taken from an approved zone ? ie. do you possess bacteriological records of product which comply with current legislation ?

If you are harvesting from a conditional zone are you

   a) Complying with legislation most or all of the time or do you know ?

   b) Evading monitoring of the product to an extent that it may lead to food-borne disease and thus jeopardize your business and that of other neighbouring businesses ?

   c) Selling product direct to supplier for further value-added processing or depuration and if so, do you envisage a change in this arrangement where you may be forced to or decide voluntarily to carry out these further operations on site ?

4. Laboratory services available

If you are getting your product tested regularly are you satisfied with

a) The laboratory service regarding speed of results and level of expertise ?

b) The cost of this service and if so do you envisage future prohibitive costs as perhaps production increases or charges increase dramatically ?

If you are one hundred percent committed to compliance with legal product specifications and probably stricter specifications required by buyers, and have a medium to large scale operation, then laboratory set-up will probably be justified economically, particularly if harvesting from a conditional zone.

Having considered all of the factors involved, you may decide that setting up a laboratory either independently or in conjunction with another farmer/processor to fulfil your testing needs. If this is the case then a lot of planning is involved to avoid any pit-falls.

The first step is to identify the basic requirements involved and to assess their cost.

These could be summarized as follows;

A. Capital Investment

1. Basic laboratory structure
2. Fixtures and fittings
3. Equipment

B. Current Expenditure

1. Laboratory technicians salary
2. Laboratory consumables
3. Gas & Electricity
4. Office Expenses (Phone. Fax etc.)
5. Maintenance Costs
6. Rent & Rates
7. Insurance
8. Depreciation

Structural requirements:

The basic demands of the laboratory structure is to allow safe testing of the product without escape of any pathogen into the production area or community. It must be protected from the outside by correct barriers ie. proper ventilation so that windows don't need to be opened, doors which do not open onto the processing area or directly to the outside. Obviously, to prevent cross-contamination, staff access to the laboratory must be curtailed.

The structural requirements could be summarized as follows;

* AMPLE SPACE
* SMOOTH SURFACES
* ADEQUATE LIGHT, VENTILATION AND HEATING
* HAND BASIN AND LAB SINK
* ADEQUATE STORAGE
* MINIMUM 6 POWER POINTS
* GAS SUPPLY

Equipment requirements

The list of equipment could be endless but there are some basic essentials such as autoclave, incubator, water bath, hot plate, distillation unit, glassware etc.

Before purchasing any equipment though, it is best to seek the advice of a trained food microbiologist or laboratory technician. Total reliance on scientific supplier companies for advice is bad policy as you risk buying equipment which doesn't suit your requirements or which you don't need at all.

1. Water Distillation Unit: This is a water purification system which is necessary for production of purified water used in the preparation of bacterial culture media.

   A small model which produces about 4 litres per hour costs about £350. It is much more cost effective to buy a unit rather than buying purified water.

2. Incubator: This is a controlled temperature air chamber for bacterial cultivation. As the temperature settings are adjustable, different microbes can be cultured.

   Larger models are recommended as they do not suffer from wide fluctuations in temperature and they also allow higher volume testing. For ease of monitoring, an in built display is recommended. Two incubators are recommended for initial laboratory set-up, each costs about £500.

3. Waterbath: This is a controlled water chamber for bacterial cultivation. Like the incubator, it has adjustable temperature settings which allows cultivation of different types of bacteria. The unit should have a heater, thermostat and stirrer while the bath should have an insulated base and detachable lid. A water bath is required when carrying out the MPN test. Two water baths are necessary if the MPN is going to be carried out, at a cost of about £500 each.

4. Lab blender: This is a machine used for mixing and blending test samples. It is essential to produce homogeneous test samples to ensure uniform results. An essential requirement of a blender is that it doesn't create aerosols thereby contaminating the laboratory. The blender most frequently used in food microbiological laboratories is a "Stomacher" model. Stomacher models cost about £1,400.

5. Hotplate: This is necessary for dissolving culture media which is usually in powder form. A simple, sturdy model with an in built magnetic stirrer is recommended. Hotplates usually cost about £200.

6. Autoclave: This machine is for steam sterilisation and has two roles in the microbiological laboratory. In the first instance, it is used for sterilising the culture media for bacterial cultivation. Secondly, it is used for inactivation of possible infectious bacterial cultures and other material produced as a result of bacterial cultivation.

   There are two types of autoclave which can be purchased when setting up a laboratory. The first type is a small portable model, which is basically a larger version of the traditional pressure cooker. It is manually operated and costs in the region of £400-600. The second type is much larger, with a capacity in the region of 48 litres. This type can generally costs between £4,500 to £9,000.

7. Balance: The balance, a more sophisticated version of the mechanical weighing scales, must be a top-pan model. It is essential for accurately weighing out small volumes of culture medias etc. and ideally should have a weighing range of between 0.1 grams and 2 Kg. The model purchased should be self calibrating with a digital display. Although prices vary enormously, the average price of a balance would be about £700.

8. Fridge: A fridge is essential for chilling samples which are awaiting testing and it can also be used for storing pre-prepared culture media. A good domestic fridge will suffice and costs about £250.

Optional equipment

The following pieces of equipment while not essential will improve the overall capability and possibly the capacity of the laboratory. Since these optional extras are a considerable additional expense, it is not advisable initially at least, to consider these investments unless they can be justified by high volume testing etc.

Colony counter: This is a magnification system which allows easier visualization of bacterial colonies. A colony counter usually costs about £400.

Microwave: This is used for the speedy liquefaction of bacterial growth media however as yet there is no official recommendation for microwave use in laboratories. Microwaves are now very competitively priced and 600 Watt models can be purchased for about £120.

Oven: This allows heat sterilization of utensils and costs about £450.

Vortex Mixer: This piece of equipment is recommended for use in the MPN test as it ensures better mixing of the sample and therefore in theory should give better results. As with any mixing device for bacterial cultures, it can lead to the

generation of aerosols and thence contamination of the laboratory so its use should be controlled. Vortex mixers are relatively cheap and can be purchased for about £130.

Air flow safety cabinet: This is used to protect workers from infection and limit possible contamination of the laboratory. It is essential in laboratories where staff are exposed to very dangerous or infectious pathogens. The major advantage with this cabinet is that testing can be carried out in a very controlled atmosphere which should be free of contaminating bacteria. If the laboratory atmosphere itself cannot be controlled to minimize the chances of contamination from the outside then an air flow cabinet, however simple, would be necessary.

There are three types of cabinet available,

Class 1 type has a negative pressure system which sucks aerosols into the filter. It offers no real protection for the worker but is efficient in limiting contamination.

Class 2 type sterilizes the air in the cabinet, 70 % of which is recirculated. The air barrier created protects the worker.

Class 3 is identical to Class 1 type except that the cabinet is totally enclosed. The cabinet gives full protection to the worker.

Air flow cabinets vary enormously in price with simple models costing about 2.5 K and more sophisticated ones costing over 10 K.

Dispenser: This is used for dispensing medias solutions semi-automatically and is extremely useful for high volume testing.

Microscope: This is necessary for confirmation of bacterial types etc. It is not absolutely essential in a small routine testing laboratory however it may be desirable to be able to visualize the bacteria under isolation. Microscopes can be purchased for as little as £100 however the higher resolution, binocular models cost about £800.

Other Laboratory Requirements

Glassware: Various pieces of glassware are essential to carry out bacterial testing. These include;

Universal bottles
Durham Tubes
Glass Rods
Beakers (500-1000 ml)
Media Storage Bottles

Plastics: Some plastic apparatus is also essential in laboratory set-up and this includes;

Specimen containers (250/500 ml)
Graduated sterile pipettes
Distilled water container

Miscellaneous items: Besides equipment, glassware and plastics there are also some miscellaneous items which are essential in setting up a laboratory and these include the following;

Bunsen burner
Tweezers
Office knife
Magnetic stirrer
Racks for holding Universal bottles
Stop clock
Pipette fillers

MONITORING PARAMETERS AND EQUIPMENT

Architects, engineers and builders will provide a facility that may look fantastic and be the latest technology. However, if the correct monitoring is not carried out and the plant operated properly then it is questionable whether the plant would purify shellfish.

The factors or parameters that need regular checking are as follows:

- Shellfish
- Bacteria levels
- Water quality
  - Salinity
  - Turbidity
  - pH, Nitrates, Ammonia, etc.
  - Oxygen
  - Toxic algae
  - Toxic compounds
- Water flow
- Temperature
- Sterilisation apparatus

There are quite sophisticated and expensive pieces of apparatus and tests that can be used to monitor most of the above. However, experienced personnel using the senses they were born with can go a long way with certain pieces of the necessary equipment.

Shellfish

The shellfish is the first thing that needs to be checked.

It cannot be stressed enough that it is very important for the plant manage to know the area from which the shellfish are coming from and also who is supplying it and how they treat their product.

The bacteria counts on harvest are important. If for example there has been heavy rainfall (in an agricultural area) after a fairly dry spell, the agricultural run off being washed down the rivers to the sea could cause bacteria levels to increase dramatically. It is possible for shellfish in an A area with counts normally below 3 F. coliforms/g to have a contamination level 100 times greater after heavy rainfall. If this is the case then the normal purification time (36-48 hours depending on national regulations) may not be sufficient.

This is why it is a good practice to have each batch of shellfish tested before purification, and after. Though the results are usually not available for over 24 hours, at least at the end of a purification run the contamination level of the shellfish going in will be known and the plant manager should know from experience whether his system will have cleared the shellfish in the time period.

From studies where shellfish are artificially contaminated with bacteria and viruses it has been generally found that a quick contamination, even at very high levels, will also have a quick elimination time. However, if the same levels of contamination are reached over a long period then the purification time is also increased (this relates to whether the bacteria and viruses etc. have time to penetrate to the tissues of the shellfish). Therefore if the area the shellfish is obtained from has consistent naturally high levels it will most likely take longer to purify this animal than it would in the example given above due to rainfall run off.

Other factors that need to be considered are what is causing the microbiological contamination in an area. If bacteria counts are due to agricultural run off then that area would be less likely to have harmful viruses than an area with the same counts from domestic sewage. Also the risk of clinical outbreaks from contaminated shellfish is generally related to the species and how it is consumed.

Mussels are very active filter feeders and if mussels and oysters were side by side mussels would most likely have higher contamination levels. However because mussels are usually cooked whereas oysters are eaten raw it is the oysters (or other shellfish eaten raw) that would most likely result in food poisoning if the two species were put through a faulty purification run.

As stated shellfish have to be "happy" to purify properly. It is important to check the condition of each batch when it arrives. If shellfish are weak because they have been badly treated or are in a spawning condition then they may not purify.

It is important to check shellfish for gaping and the condition of the gonads for spawning before they are put in a tank. It is also important not to subject the animals to temperature shock. Bringing animals from a chill room/truck into a tank 10-15^{0}C warmer or from standing on a pier/boat in sunlight to a tank 10-15^{0}C colder can result in animals spawning even if they are in a healthy firm condition. (This heat shocking technique is used by hatcheries etc. to induce spawning). Rough handling from bad grading etc. can also cause animals to spawn. If the animals spawn in a tank the spawn and eggs have to be flushed out otherwise the tank will become fouled and all the shellfish die. In addition effective purification will not take place.

Once shellfish are in a tank they should be visually checked after about an hour to see that they are open and filtering. Mussels will start to byss up after several hours. If they are not then it is an indication that something is wrong. Other species such as oysters are harder to tell if they are actively filtering. In species such as clams the syphons should become apparent. If the water is in any way cloudy at the start of a run it should be cleaned by the shellfish themselves within an hour or two if they are actively filtering.

If any smell is noticed from the tanks it is usually a sign that things have gone wrong (either shellfish spawning or dying) the tanks should be drained and shellfish checked and discarded if necessary.

A build up of foam is normal, however excessive foaming again is usually a bad sign. Visual checks should also be made to see if there is not excessive aeration or spray that is disturbing the shellfish and causing them to close and not purify. The clarity and colour of the water should also be visually checked.

Water quality: salinity

The sea water quality is one of the most important factors. Special care should be taken when selecting an intake point. Ideally it should be possible to pump in water at all times and the intake point should be sited so as not to be subjected to variations in salinity. If salinity variations are unavoidable (i.e. when sited in an estuarine area) pumping in of water should only take place at the correct tidal phase to ensure the correct salinity water is taken in. Generally full strength seawater at about 33 ppt (parts per thousand) is recommended. However it is again important to know the salinity of the area from which the shellfish come as best results are obtained when the salinity of the tanks are within 10 to 20% (not ppt) of the salinity of the water the shellfish are grown in, i.e. if shellfish grow in estuarine conditions with a salinity of 23 ppt it would most likely stress them to be purified in water at 33 ppt (even though they would survive they may not be actively filtering).

Again, different species are more or less tolerant to salinity changes. With mussels being the most tolerant and scallops being the least. (See specific gravity chart in appendix).

Though some people will claim to be able to tell you how salty the water is by tasting it, it is recommended that a more reliable method is used.

Hydrometers are one of the cheapest methods. There are many different types of hydrometers on the market. The range that is most useful for testing seawater is from 1.000 to 1.030. Hydrometers measure specific gravity for a liquid (i.e. how dense the liquid is relative to fresh water). The saltier a solution the heavier it is, this means that the hydrometer will float higher out of the water and give a higher reading on the scale.

Note the lowest number 1.000 is at the top of the hydrometer and the higher numbers, 1.030, are lower down nearer the body of the hydrometer. The hydrometers should never be placed in the tanks as false readings can be obtained by looking at the scale at the wrong angle or if the hydrometer is not floating freely but resting on something. A clear graduated cylinder of sufficient size to float the hydrometer in should be used. A water sample is taken in the cylinder, it's temperature is measured (as the density varies with temperature), and the hydrometer is placed floating free in the cylinder. Keep the hydrometer away from the sides of the cylinder. Put the cylinder on a level bench and bend down so that your eye is level with the water level in the cylinder and read the level at the bottom of the meniscus (fig. 4). The specific gravity given to you off the hydrometer is then read off the specific gravity chart, corresponding to the temperature of the water. It is recommended that water samples are brought back to a central bench for testing as this cuts down on breakages and also the salinity/specific gravity

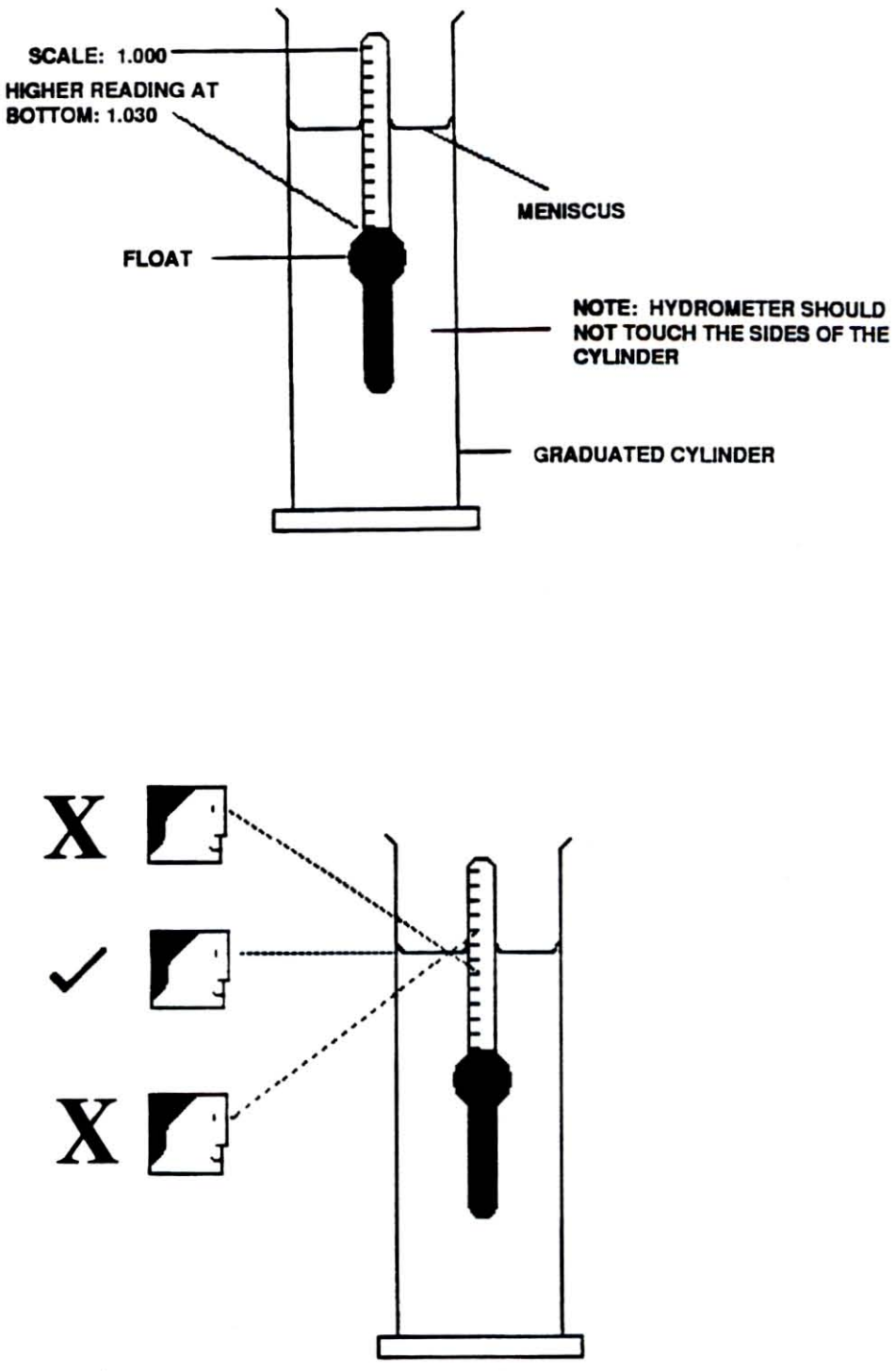


Figure 4. Diagram of hydrometer, note the meniscus should be at eye level for a correct reading

chart can be kept on the wall.

Titration or chemical methods for determining salt concentration are not recommended as they are time consuming and disposing of the resulting chemicals can be problematic.

Refractometers are instruments that make use of the fact that light is bent when it passes through water. It will also bend, more or less, depending on the salinity. A drop of water to be tested is placed as thin film on the prism of the refractometer and the reading is read off a scale viewed down the eye piece. Most refractometers are calibrated for use at 20°C. which is near normal room temperature; so if the refractometer is near this ambient temperature the small water drop will reach this temperature when it is put on the refractometer. Thus the refractometer is extremely easy to use but they are relatively expensive (several hundred pounds). Care should be taken to wipe off the sea water with lens tissue. The instrument should be stored in a dry environment.

Electronic salinity meters or temperature salinity meters are extremely useful for measuring both temperature and salinity. They vary from model to model and manufacturer to manufacturer.

The conductivity of the seawater is measured by an electrode (again the more saline the more current) which is then converted by the machine (depending on model) directly to salinity. On some models the temperature has to be measured first and set for an accurate reading. Depending on how much you wish to spend (from £200 to £2,000) the salinity meters can be more automated, come with more robust electrodes, longer flexes or be more weather/water proof. It should be noted though they are easy to use and can measure salinity at depth (useful when selecting your water intake point) for most purification plants one of the other methods will suffice and be cheaper. Electrodes should be washed in fresh water (distilled or deionised if possible) after immersion in salt water.

Salinity should be checked when starting to fill a tank and when it is full. If species such as scallops (and many crustacea) are immersed even for a short period in water of the wrong salinity then death can result. So checking the water once the tank is full and the shellfish covered many to too late.

Intake water should be checked, prior to siting the intake point for hydrocarbons, heavy metals, bacteria, viruses, toxic plankton. Periodic checks should also be carried out once the plant is established. For it may happen that the intake water, if it had toxic substances in it, could make the shellfish being purified more harmful than they may have been originally. If it is not possible to intake water directly from the sea then water may be tankered to a plant or artificial seawater may be made up and used. (This will be dealt with further in the section on the multilayer system).

Temperature

Temperature is also an important factor. Not only is it important to monitor and

control the temperature of the water in the tanks but as mentioned above the shellfish should not be exposed to fluctuations in temperature, especially rapid rises and falls. It is important to also know the actual temperature the shellfish have been stored in or transported in prior to it reaching the purification centre along with the temperature of the workplace in which it will be later handled and graded.

The water temperature is crucial. If it is too high the animals may be over stressed or spawning induced; if it is too low the shellfish may look open but they may not be actively filtering. The effective temperature range varies for different species. Generally temperatures should be maintained over 8^{0}C but should not exceed much over 18^{0}C, as above 18^{0}C oxygenation of systems may become problematic. For the native clam temperatures need to be about 13^{0}C before it starts filtering effectively.

Various types of glass thermometers are available. Care should be taken in selecting a thermometer with an appropriate temperature range, 0-60^{0}C is usually adequate. It should be emphasised that alcohol thermometers should be used in preference to mercury thermometers for shellfish, for if the mercury thermometer breaks the mercury could contaminate the shellfish consignment which would then have to be dumped.

Metal sleeves are available to protect thermometers. It should be noted that time is required to equilibrate the sleeve and thermometer to the ambient temperature.

All thermometers should be left until the temperature reading is constant. This time can vary according to the type of instrument used. The temperature should be read with the thermometer or probe in place (i.e. bulb of glass thermometer in the liquid). If the reading cannot be taken while the thermometer is immersed then care must be taken to read it immediately on removal as the thermometer will quickly adapt to the ambient temperature.

Glass thermometers, even in protective sleeves, are fragile, though they are cheap and are maintenance free. A problem though is that they cannot measure temperature remotely i.e. you have to be near the thermometer and tank etc. to read it, also they cannot be used to measure temperature at depth etc. if you are at the surface.

Digital thermometers with steel probes are robust and can be pushed into the centre of bags on trays of shellfish etc. They are relatively inexpensive (under £100) and apart from batteries are relatively maintenance free.

Temperature/salinity probes as mentioned earlier are expensive but can be used to measure temperature remotely, i.e. at the bottom of tanks etc. This will be discussed later in the section on data loggers.

Purification plants should have a mounted thermometer in the main work/packing shed and chill room with separate thermometers for holding and purification tanks. Maximum/minimum thermometers are ideal for static use in the above mentioned places as they will give the maximum, minimum and actual temperature (at time of

reading) for the time interval since previously being read and reset.

Oxygen

There is one piece of equipment that can tell you more about your tank system than any other and should be compulsory in any purification centre and that is an **oxygen meter**.

Oxygen levels can either be measured in mg/l or in % saturation levels. When measured in mg/l levels this can be converted readily to % saturation by using a temperature conversion chart.

During a purification run there should be no part of the tank that should have an oxygen level below 50% saturation. Generally with oxygen levels below 50%, shellfish do not purify efficiently so oxygen meters are invaluable in finding dead spots in tanks and also if tanks are overloaded, i.e. the aeration system is not sufficient for the amount of shellfish the tank contains. They will also show, for example, with long tanks that by the time the water flows to the end whether or not the oxygen levels in the water has fallen dangerously low.

The amount of oxygen dissolved in the water varies with temperature. The colder the water the more oxygen it contains. Once water reaches 24^{0}C it contains very little dissolved oxygen. So though shellfish will survive in the sea at temperatures above 24^{0}C in artificial systems, that are heavily stocked per cubic metre of water, when temperatures near 24^{0}C are reached additional aeration is usually required to prevent animals from dying. That is why if the temperature of the tanks can be kept below 18^{0}C it tends to avoid critical lowering of oxygen levels.

It should be noted that though tanks can take a certain tonnage during the winter once temperatures rise in the spring or summer that the stocking density of the tanks maya have to be reduced depending on oxygen levels in the system. It should also be noted that just because a system can take for example one tonne o bottom mussels it does not mean that it should be able to take one tonne of rope mussels. If you check the respective meat yields you could find that there may be twice as much meat and therefore respiring, oxygen using tissue in the rope mussels. This will cause the oxygen levels in the tank to drop and the loading capacity of the tank may need to be reduced. Therefore the oxygen meter can also be used to determine loading capacities of systems for different types and species of shellfish.

There are numerous oxygen meters on the market but care should be taken to stress that they will be used in a marine environment. Certain oxygen meters will automatically compensate for temperature where with others the reading given by the machine will have to be converted on a temperature graph or chart. Most machines have a switch for them to be set for either fresh or sea water. Hand held probes have to be moved slightly for they require a flow of water over the membrane of the electrode to give an accurate reading. If the probe is left stationary the electrode itself will use up all the oxygen in the water immediately surrounding it and the readings will show a sharp drop in oxygen levels. There are

special oxygen probes designed for use in slow moving waters or when they are left in position for a long period.

The oxygen probe takes considerable maintenance compared to most other pieces of equipment mentioned. Membranes on the probes have to be changed periodically along with the electrolyte solution and the electrodes cleaned. Depending on the model you may also have to calibrate the electrodes periodically by placing them in either water with no oxygen in it or water at 100% saturation.

Data loggers are a piece of equipment that can store information from various probes. This information can then be down-loaded onto computers. If oxygen and temperature probes are linked to a data logger then fluctuations in temperature and oxygen can be measured at different points in a tank throughout a complete purification cycle. This level of monitoring is usually only needed when commissioning or designing a new system. However, probes can also be put into different tanks and linked back through a data logger to alarm systems. Though initially expensive (5-probe oxygen data logger is around £5,000, enough to do five tanks) these systems can pay for themselves in one night if something goes wrong. For example if a pump breaks down in a tank and the oxygen levels goes below a predetermine point an alarm will sound. The alarm can either be linked to a standby pump or solenoid valve to drain the tank as well as notifying the appropriate person (via phone alarm system).

Water flow

The amount of water or flow rate of water through a system is also critical to oxygen levels. Depending on the system used various flow rates or exchanges of water are required per hour. The most efficient method of determining this is to have flow meters fitted to all tank input pipes.

Certain systems will only function properly if the correct flow is passing into the tank. A flow meter will also give you an indication on how well your pump is performing. A drop in flow rate may mean the impeller has to be replaced.

Turbidity

The turbidity or clearness of the water needs to be checked. A secci disc is traditionally used to give an indication of turbidity. This is basically a disc divided into quarters which are alternatively painted black and white. This disc is lowered into the water, the distance down where you can no longer see the divisions between the white and black is the first reading. The second reading is when you cannot see the disc at all. The secci disc method is not really practicable in a purification plant.

A more realistic approach is to put marks down the side of the tank and depending on how many marks you can see you can determine how cloudy or turbid the water is. A spectrophotometer can also be used to give a reading on how much light can pass through the water.

Turbidity is important especially when ultra violet lights are being used as the sterilisation medium. For if the water is too cloudy the U.V. light cannot function efficiently. As stated normally if the shellfish are active they will clear a tank within one to two hours and then the U.V. will start being fully effective.

Other parameters

Apart from U.V. units being fitted with clocks to log the hours used, detectors can be fitted that will give you the actual dosage the U.V. lamps are giving. If this falls below a certain level (depends on the U.V. system) then bulbs will have to be replaced. Unfortunately not all U.V. systems are conducive to having monitors fitted.

Other things that have to be checked (more when using artificial seawater or reusing the same water for several purification runs), are pH, ammonia and nitrate levels. These factors are even more important if you are also using your facility to hold crustacea as they are more sensitive than most bivalves.

Ammonia, nitrates and pH can all be measured adequately from cheap test kits available from most aquarium shops. pH can be measured using a pH meter and also some meters that measure conductivity (salinity meters) can also be used to determine pH by using conversion tables. If more accuracy is required for ammonia and nitrates colorimeters can be purchased along with slightly more specialised test kits. These however would only be supplied by scientific specialist dealers. Normally for purification runs where the water is changed after each run pH ammonia and nitrates will not be a problem.

TOXIC PLANKTON MONITORING

A factor that seems to be becoming increasingly problematic for anyone dealing in shellfish is the presence of toxic algal blooms or "red tides" in the water. These blooms can either directly affect and kill the shellfish themselves or make anyone eating the shellfish ill.

In both fresh and sea water microscopic plants and animals live which are generally termed plankton. The small animals (which often include larvae of shellfish) are termed zooplankton whereas the small plants or algae are termed phytoplankton. Plankton is the major food source for bivalve shellfish which filter it out of the water. It is certain species of these microscopic phytoplankton that are responsible for toxic blooms or "red tides".

If certain conditions occur in the sea very large concentrations of these toxic algae may concentrate together forming what is generally termed a bloom. Certain of these blooms actually colour the water red if they occur in high enough concentrations. This had lead many people (especially in Ireland) to refer to any toxic algal populations as "red tide", even if they are visible or not to the human eye.

In Irish waters there are two common problem groups; Gyrodinium aureolum and Dinophysis species. Both are a type of algae known as dinoflaquellates but they differ from each other in their ecology and effects on aquaculture.

Dinophysis Sp.: There are three common species, *D. acuminata*, *D. acuta* and *D. rotundata*. All these occur in coastal waters but generally do not reach very dense concentrations (<40,000 cells per litre). All species can cause illness (but not death) in humans who eat shellfish contaminated by these species. This type of poisoning is termed diarrhoetic shellfish poisoning (D.S.P.) as it causes people to have varying degrees of diarrhoea.

The species generally grow during mid and late summer especially near areas of stratified water. They do not colour the water and can only be detected either microscopically or by their effects on seafood.

Gyrodinium aureolum does not contaminate shellfish directly (in relation to being a health hazard for human consumption) but it can kill a variety of marine organisms including fish, snails and bivalves. Toxic affects are usually associated with large areas of discoloured or reddish brown water. Again the species is commonest in mid and late summer near stratified water.

Both species cause ongoing problems in the aquaculture industry especially in Ireland along the south and southwest coasts.

A variety of other species are potentially dangerous and have caused serious problems in other parts of Europe and the world. One such species is Alexandrium (formerly called Gonyaulax). This species has been responsible for deaths in

Canada and the United States when people have eaten contaminated shellfish. This species causes paralysis and is known as Paralytic Shellfish Poisoning (P.S.P.). While an Alexandrium species is common in Irish waters it has only been found to be toxic in Belfast Lough. However, Scotland has had extensive P.S.P. blooms around its coast and it is relatively common in other European countries.

Recently a species of a different group, the diatoms, has caused serious problems in Eastern Canada, including deaths and loss of memory (termed Amnesic Shellfish Poisoning, A.S.P.) in those who ate contaminated mussels. Again species of this genus, Nitzchia, are very abundant especially off the south west coast of Ireland to date no toxic affects have been reported in Ireland.

A number of species of phytoplankton cause dramatic changes in water colour but appear to have no toxic effects. Phaeocystis caused brown water along the west coast of Ireland in May 1990 while Prorocentrum often colours water in Galway Bay.

Generally, all growing areas for shellfish are monitored for toxic algae. However a purification centre may be in an area that is not tested. It is important to check your water intake periodically. There are no ways of preventing "red tides" but some early warning is possible by collecting water samples and examining them microscopically.

By siting your intake pipe in deep water it may be possible to escape the worst effects of blooms. However, if toxic blooms are detected in the locality it is essential to maintain a strick testing procedure.

Given the correct conditions shellfish naturally clean themselves of the toxins.

Detoxification trials for D.S.P. are taking place. As of yet though toxic shellfish should be left in their harvest areas until they clear as movement of contamination species may lead to transfer of toxic algae to previously uncontaminated areas.

Apart from the species toxic to man, species such as Gyrodinium aureolum need to be monitored because if high concentrations of the algae are pumped into a purification system they may kill the shellfish. Passing water through ultra violet light may kill the algae but it will not prevent it from having a toxic effect. If the purification centre is operation in periods of likely blooms at least weekly water samples should be taken.

Samples are taken by collecting 100 mls of water in a clean container and adding 5 mls of either 40% formalin or Lugal's iodine (100g Potassium iodide in one litre of distilled water plus 50g iodine and 100 mls of acetic acid). These should then be sent to a competent laboratory for checking. Care should also be taken if species mentioned above (eg. Nitzchia and Alexandrium) which have not had toxic affects to date are noticed in large quantities. Samples of shellfish should be sent for toxicity testing just to be sure.

DOCUMENTATION REQUIREMENTS AND RECORD KEEPING

As part of the requirements of the Council Directive (91/492/EEC) proper records are to be kept so that any batch of shellfish can be traced from the retailer right back to the producer.

The harvester whether he is a fisherman or a shellfish farmer has to be registered and be issued with a Shellfish Gatherer's Document Book (issued by competent national authority). This book is a duplicate book and after the gatherer fills out the required information (sample Irish documentation in appendix), he retains a copy and a copy goes with the consignment either to the dispatch centre (if the shellfish is from an A category area) or to a depuration centre.

A gatherer must fill in date of harvest, location of harvest, classification of harvest area, species harvested, quantity, destination and approval number. This must be signed by the gatherer and date stamped on receipt by the recipient on arrival of the shellfish.

The dispatch/purification centre in turn has to be registered and issued with a ledger book (see samples in appendix). It is possible that the shellfish will go to a purification centre and then sent to a different dispatch centre. However it is more usual for the purification centre to also be the dispatch centre.

At the purification centre the following must be recorded for each batch of shellfish.

Date of delivery, quantity and species, registration document number, unpurified shellfish test results, source water test results, time of start of purification, time of end of run, purified shellfish test results, dispatch date, destination information. In addition water temperature, dissolved oxygen and salinity levels are also required to be recorded.

At the dispatch centre, (even if the same plant) dispatch centre details must be filled in. Again these contain date of handling, species, source of shellfish, quantity, registration document number, tests carried out along with their results, dispatch date and destination details.

Once shellfish leave the dispatch centre each bag or container must contain a label or wrapping which gives details of country of dispatch, species name, dispatch centres approval number and date of wrapping. This "health mark or label" must be durable and waterproof and the information presented must be legible, and in easily decipherable characters.

If a retailer splits a bag and sells in smaller quantities he must keep the above mentioned health mark and label.

There is a requirement for each person in the chain to retain copies of relevant documentation for a period of 60 days after a consignment leaving them.

All the above are legal requirements. These are the bare minimum of

documentation that an efficient plant manager should record. In addition notes should be recorded on turbidity, if artificial or recycled sea water is used then number of runs should be recorded, top up levels, ammonia and nitrate levels etc; toxic algae cell counts; flow rate; draining and training times; equipment checks, e.g. U.V. hours; system cleaning records; rodent control records.

If possible complete records should be received or you should have access to farms records where important environmental and harvest details should be recorded.

At dispatch a quality report sheet of the consignment should also be filled out, giving details of meat yield, condition, size, appearance etc. of shellfish. Samples of these records from a draft quality manual for rope mussels are in the appendix.

Many operators and managers question the use of all this form filling and paper work. If people are lucky then most of it will never be needed. However, detailed records are essential if something goes wrong with a consignment or someone disputes the quality of the shellfish on arrival.

Many complaints do not filter back for a period of weeks and only usually surface if someone has become ill or when payment is sought for a consignment. If you cannot accurately trace all aspects of a problem consignment you will most likely not be able to argue a valid case with the person who received the batch and may be complaining that the shellfish died after two days or that he didn't get the size etc. that he specified.

It is a good practice to retain several bags of a consignment and store them in a chill room until you would have expected the batch to be consumed. Also it is worth phoning or faxing the day after a consignment arrives at its destination getting the consignee to state that he is happy with the batch. If however it is stated there is something wrong you can immediately check the samples in your cold room. If these also show the same problem you will have to accept the loss. However, if they are fine and you find nothing went wrong with the transport you should be able to come to some arrangement where the consignee takes some if not all the blame for the loss because of how they were treated when they arrived at his premises.

If the complaint is something as simple a disagreement over size you will be able to refer to your records and see if the consignee again has a valid complaint.

So simply, good record keeping though initially it may seem a lot of extra work, once a system is established it will become easy to maintain and second nature and more importantly in the long run, will save you money.

TRADITIONAL SINGLE LAYER AND STACKING SYSTEM

Up until very recently most purification systems were variations of the single layer system which consisted of shallow tanks usually outdoor in which shellfish were placed in a single layer. However, with the advent of Council Directive (91/492/EEC) it was stated that all tanks had to be covered with the shellfish not being exposed to extremes in temperature and restricting access by rodents.

The Directive is open to interpretation but many National authorities (especially in colder countries) stated that all tanks had to be enclosed in buildings in order to meet the requirements of the Directives. Covering larger outdoor tanks is expensive and as a consequence it may be cheaper to erect smaller buildings with more space efficient purification systems (these will be dealt with later).

If outdoor tanks have to be covered for the above mentioned reasons by inference it means that placing shellfish from B areas into bays in A areas, especially intertidal areas where rats and seagulls, carrying all sorts of diseases have access to the shellfish, will no longer be possible.

Most purification systems in Ireland are what is known as recirculating systems, where the same water is recirculated around a closed system. There are other systems though; flow through for example, where clean water is pumped through to waste. This is normally only done when pumping costs are cheap i.e. a header reservoir is filled and gravity can then be used to flow the water through the tanks or if there is a small head height between seawater intake and the tanks. Depending on the cleanliness of the intake water it may or may not have to pass through sterilisers such as a U.V. system.

The batch system is also common in some countries where tanks are filled (usually at high tide) and the same water is left (but aerated) in the tank for 8-12 hours and then drained and refilled. This is where the aeration systems originate from and will be discussed in more detail later. Again either clean or sterilised water must be used.

The single layer and stacked systems described below are both closed recirculating systems (though it is possible to have them flow through).

In general most closed recirculating systems will have intake points and an outfall to respectively fill and drain a tank at the start and end of a purification run. Once a run starts these must both be closed off.

The tanks themselves must have a smooth, hard and impermeable surface and be easy to clean. The base of the tanks must be sufficiently sloped and be equipped with an adequate drainage system.

Shellfish are usually placed inside the tanks in containers (but not always), the design of which depends on the system.

On the recirculating system there may be a sump or outlet pipe (different to

drainage pipe). a recirculating pump, sterilisation unit and spray bar or filling pipe. Oxygenation is either carried out by the spray bar or cascading weirs.

Water sterilisation methods will be dealt with in detail later. Usually, however, U.V. is used as the sterilisation medium.

Single Layer System.

In the earlier systems shellfish were laid directly onto the floor of the tanks at a depth of 7.5cm (3 inches), now they are usually placed in baskets at the same depth. To allow for improved water flow and to avoid the shellfish on the bottom being covered in faeces and silt etc. either a false floor is fitted to the tanks, raised baskets are used or baskets are placed in such a way that one end is resting on the edge of another and thus allowing water to flow under them (fig. 5).

The stocking density of shellfish is approximately 50-60kg/m^2.

No shellfish should be placed near where the water enters the tanks as the impact of the water will cause the shellfish to close and therefore not filter and purify. Water is aerated either by a spray bar or cascading weir, some systems will incorporate both.

At least 15 cm (6") of water must be above the shellfish. The flow rate for the single layer system is generally one complete water volume change every two hours. Water tends to flow around obstructions so the main flow will be above and below the baskets (fig. 6). The shellfish obtain oxygen etc. by passive diffusion and limited water circulation in the 7.5 cm layer. It may be necessary to fit baffles or flow screens in tanks to direct water flow to ensure there are no dead spaces in a system.

Sump systems can be beneficial as they allow sediments to settle etc. and also increase the ratio of water to shellfish. Wherever possible all recirculation pumps should be fitted with filters. This prevents them clogging with debris from the tanks and has the benefit of lengthening impeller life.

As a general rule long tanks should be avoided. It is better to have wide short tanks, for as the water flows down the tank the oxygen is used up. In long tanks, during warm weather especially, it is possible for most of the oxygen to be used up by the time it is half way down the tank.

Advantages of single layer system:

- Suits all types of shellfish
- Slow flow rate means low pumping costs
- Well proven and simple system
- Tanks relatively inexpensive.

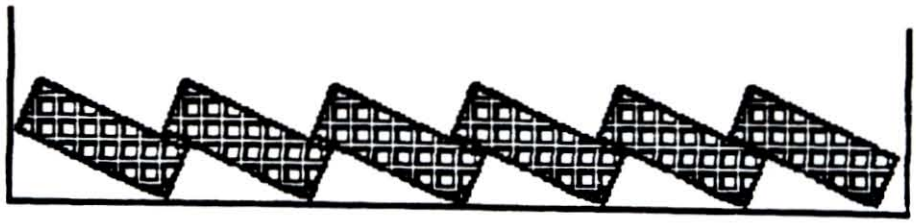


Figure 5. Staggered baskets to improve water flow.

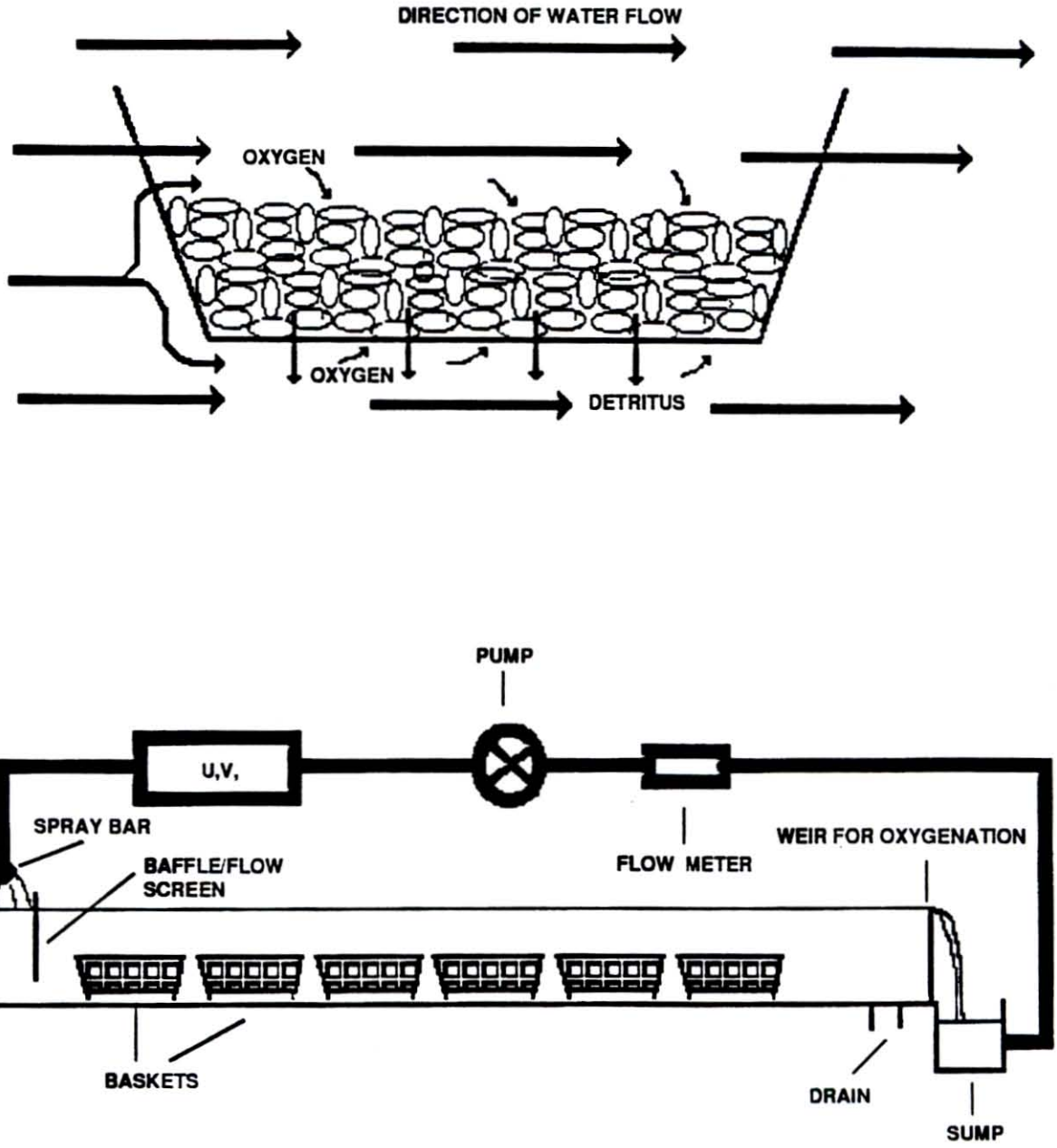


Figure 6. Traditional single layer system (below) with basket showing water flow (above)

Disadvantages

- Tanks have to be covered - large surface area and buildings are expensive.
- Loading and unloading shellfish can be labour intensive
- Because of large surface area hard to temperature control.

Stacked System

The stacked system is quite an old system and is essentially a single layer system stacked vertically (fig.7).

Shellfish are placed in stacked trays (which should have a false bottom to allow faeces to settle). Again to a depth of 7.5cm (3"). Water is pumped from a sump via a U.V. system to the top tank. The water flows across each tank and cascades down to the next.

Trays are modified so that they direct flow down into an area with a baffle to avoid disturbing the shellfish. The flow rate is the same as for a single layer system i.e. one water change every two hours. However, the head height is greater so pumping costs are slightly more expensive. Oxygenation is achieved when the water cascades down from one tray to the next.

Advantages:

- Suits all types of shellfish
- Low pumping costs
- Efficient per square meter of floor space, number of stacked trays governed by available height and ease of working.
- Individual trays can be bypassed and removed without affecting rest of system.
- Good oxygenation throughout system.
- Heat regulation can be carried out relatively efficiently in sump etc.

Disadvantages;

- Need vertical racking system
- Loading and unloading relatively labour intensive
- Large number trays required which are more expensive than baskets.

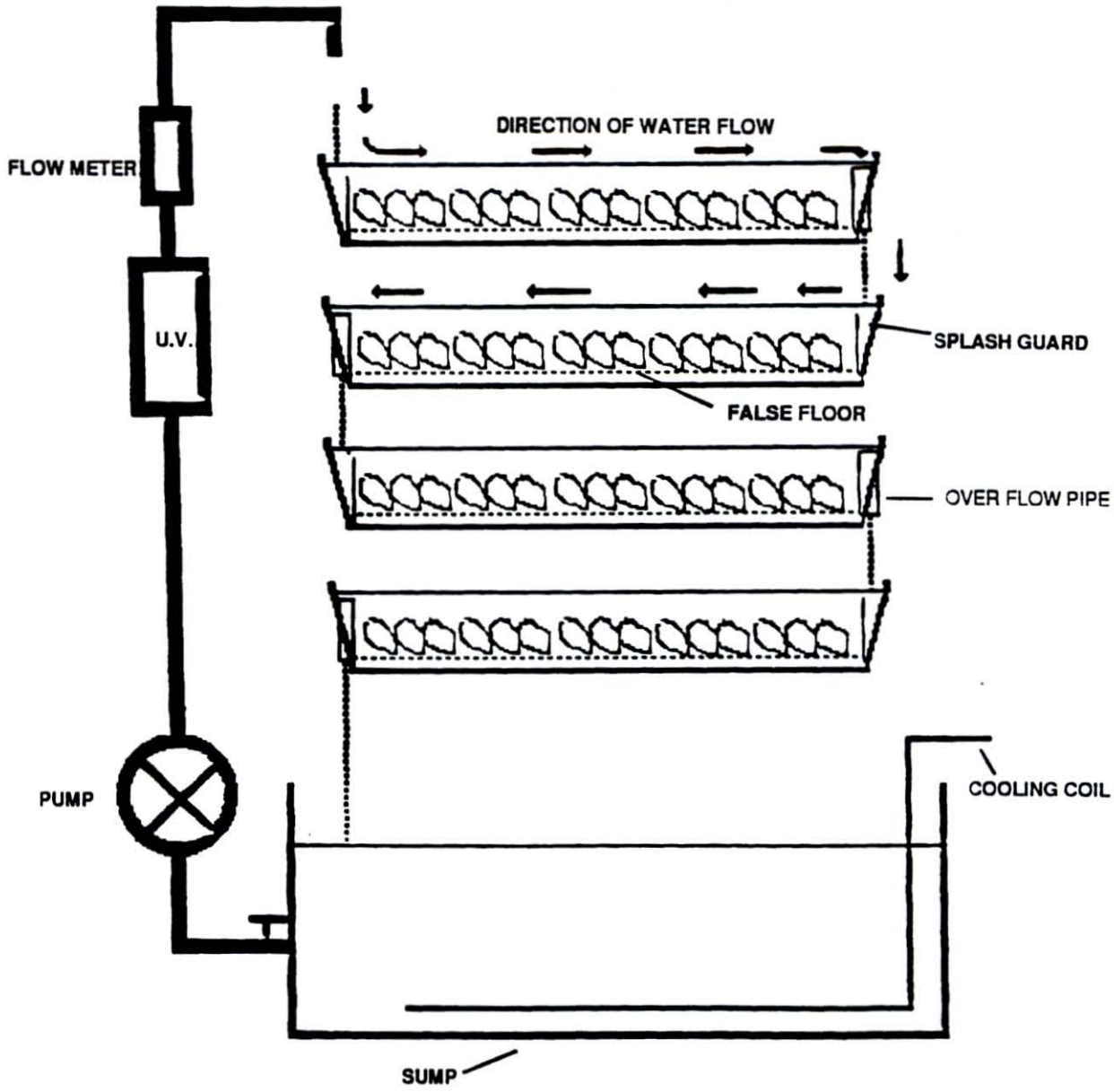


Figure 7. Stacked system.

STERILISATION MEDIUM - U.V. 0_3 Cl_2 AND IRRADIATION

At present there are three systems of water sterilisation in commercial use in depuration facilities, these are chlorination, ultra violet light and ozonolysis. Ionisation is not suitable for use with shellfish tanks but has use in sterilising sea water for washing.

Irradiation as a means of purifying shellfish will also be considered.

Chlorination

Most of the first commercial purification centres used Chlorination. Now a days this has been replaced in most countries by U.V. systems, though there are still plants operating using Chlorine.

Chlorine gas (Cl_2) (from cylinders) is pumped into a mixing chamber with water (to levels of approximately 0.3ppm) where it has its sterilising action. Most of the chlorine is then removed by an aeration system before it enters into the holding tank (fig. 8).

Dechlorination can also be achieved with sulphur bearing compounds or activated charcoal. Problems with buffering the system may occur with the former as hydrochloric acid is a by-product of the chlorine removal process. The cost of reactivating the charcoal would be a problem with the latter.

Chlorine acts in aqueous solution by diffusing through bacteria cell walls and attacking enzymes groups which lead to death of the organism. Chlorination is not very effective on viruses. One of the main problems with chlorine however is that residual chlorine can taint the shellfish and also leaks of chlorine gas are highly dangerous.

Ultra violet light

In Ireland and the U.K. ultra violet light (U.V.) sterilisation is the most common system and is preferred by the national authorities. A fluorescent tube containing mercury is used to generate ultra violet light at a wavelength of 253.7 nanometres. Light at this wavelength is germicidal. It acts by damaging the D.N.A. and R.N.A. of micro-organisms (bacteria and viruses) which inhibits them from reproducing or leads to death.

In the past weir systems have been used where the U.V. tube is situated several centimetres above water cascading over a weir. These systems were not very efficient. Now closed systems are used. In these systems the U.V. tubes are placed in quartz sleeves which are inserted into a chamber through which water flows (fig. 9). The tubes are separated from the water by a quartz sleeve (quartz glass has to be used to allow U.V. light to pass through). The distance of the tube from the water is very small, usually less than 0.5 cm. For U.V. lights to operate effectively they have to be at a temperature of approx. 40°C. The cooling effect of water directly in contact with the tubes is one of the reasons they are separated by

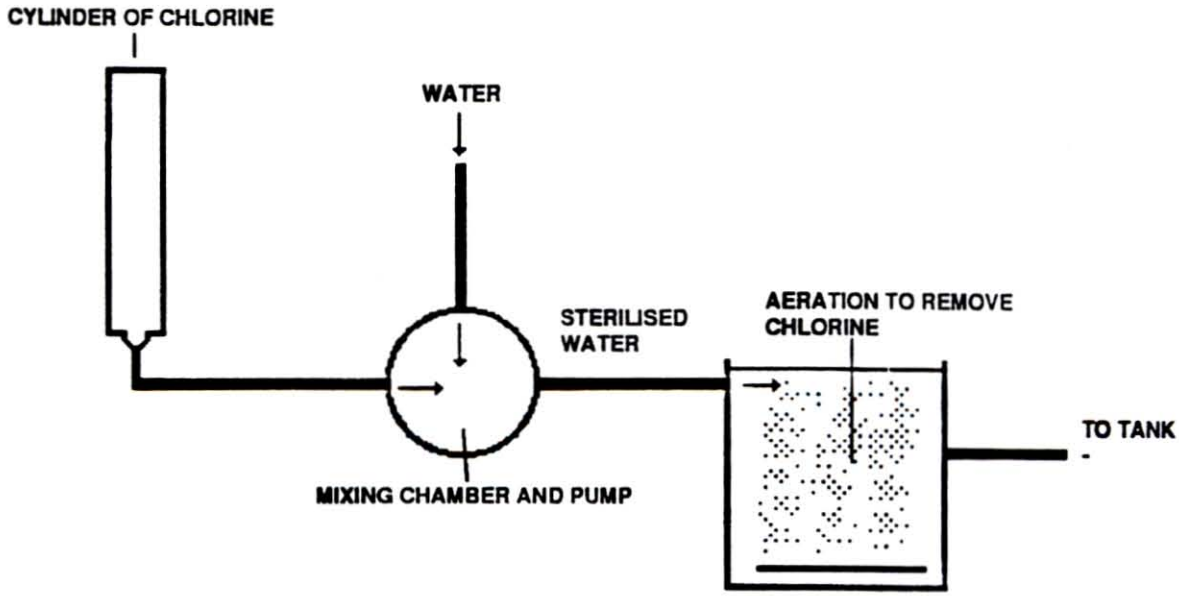


Figure 8. Diagram of chlorine sterilisation system.

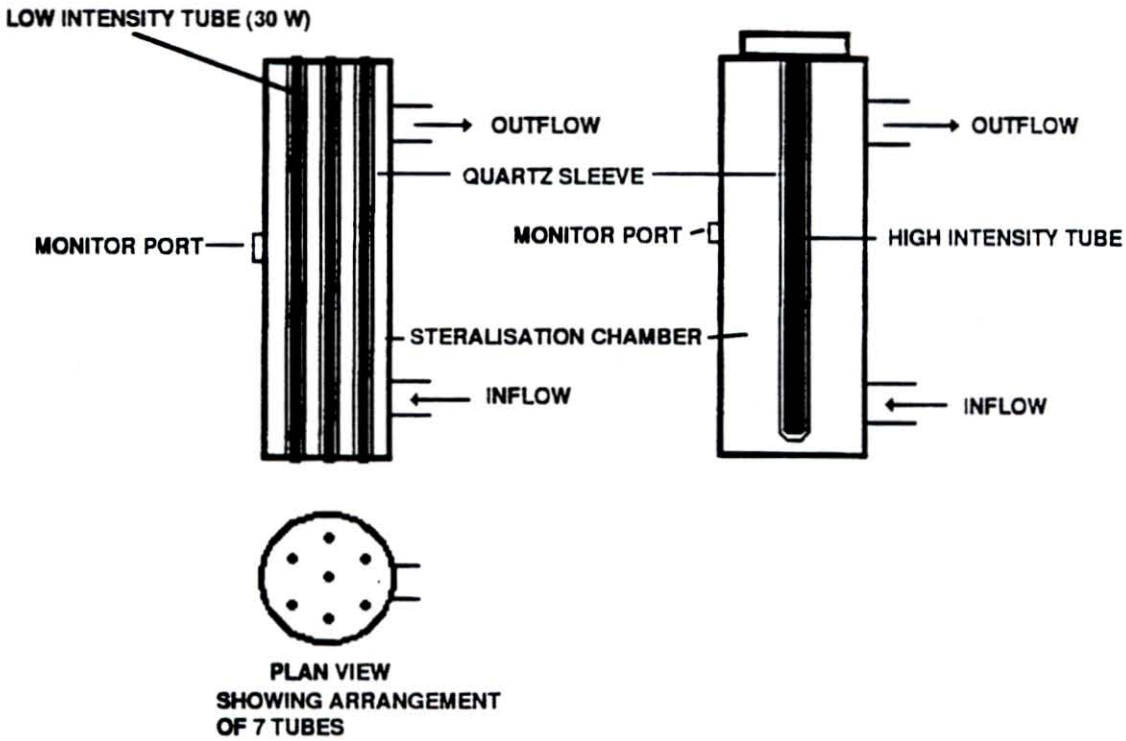


Figure 9. Diagram of low intensity and high intensity ultra violet systems.

the quartz sleeves. U.V. lights do not function well in turbid or cloudy water as (as a light in fog) the light cannot penetrate through the particles thus it will not be able to kill bacteria and viruses. If water quality is a problem even after shellfish have been filtering for an hour or so then filters will have to be installed to clean up the water.

Different countries have slightly different regulations in relation to dosage levels of U.V. Britain is one of the strictest and requires a minimum of a 30 watts U.V. tube for each 2,200L of seawater in a system, this corresponds to a dose of 10 mw/cm.sq./s for a single pass once an hour.

It is essential that tubes can either be visually checked to see if they are operating or that a U.V. monitor is fitted. Clocks (which cannot be accidentally reset) should be used to record hours of usage. Tubes should be replaced after 2,000-2,500 hours usage. If U.V. monitors are fitted it should be at the outer sleeve for a single tube and in an area which tubes are not being shielded in a multiple unit. When dosage figures are cited by manufacturers make sure they are the dosage given at the outer edge of the chamber and not in the middle.

Increasing the turbulence of the sterilising chamber helps to achieve a better kill rate. Depending on the flow rate and the model from 90 to 99% kill rates are achievable on a single pass.

There are basically two systems of U.V. in use in depuration plants. One uses multiples of the 30w tube, usually with separate units for each tank. The second system uses only one high intensity tube (measured in kilowatts) and is usually used when the water is only being sterilised once when entering the system. The advantage of the high intensity system is that very large flow rates can be pumped through the system.

The main problem with the U.V. system is that the emission of the germicidal wavelength decreases with time therefore it is important to change the bulbs at the recommended time (unless U.V. monitor indicates otherwise), even though they may appear to be working properly. Care should be taken never to be exposed to U.V. light, all tubes should be shielded as they can damage vision and cause severe skin burns. The quartz sleeves have to be cleaned at regular intervals to stop build up of scum and scale which could prevent U.V. penetration. Again in turbid or cloudy conditions the effectiveness of U.V. sterilisation will be severely reduced and increased wattage and turbulence in the chamber may be necessary.

Ozone

Ozone is an allotrope of oxygen i.e. an oxygen (0_2) molecule has two atoms of oxygen whereas an ozone (0_3) molecule has three. It is obtained by passing air or oxygen over two electrodes across which there is a large voltage or potential difference.

Ozone is made in an ozone generator and is then sucked (by venturi pump system) to a mixing tower where it is bubbled up through the tower (fig. 10). This tower is

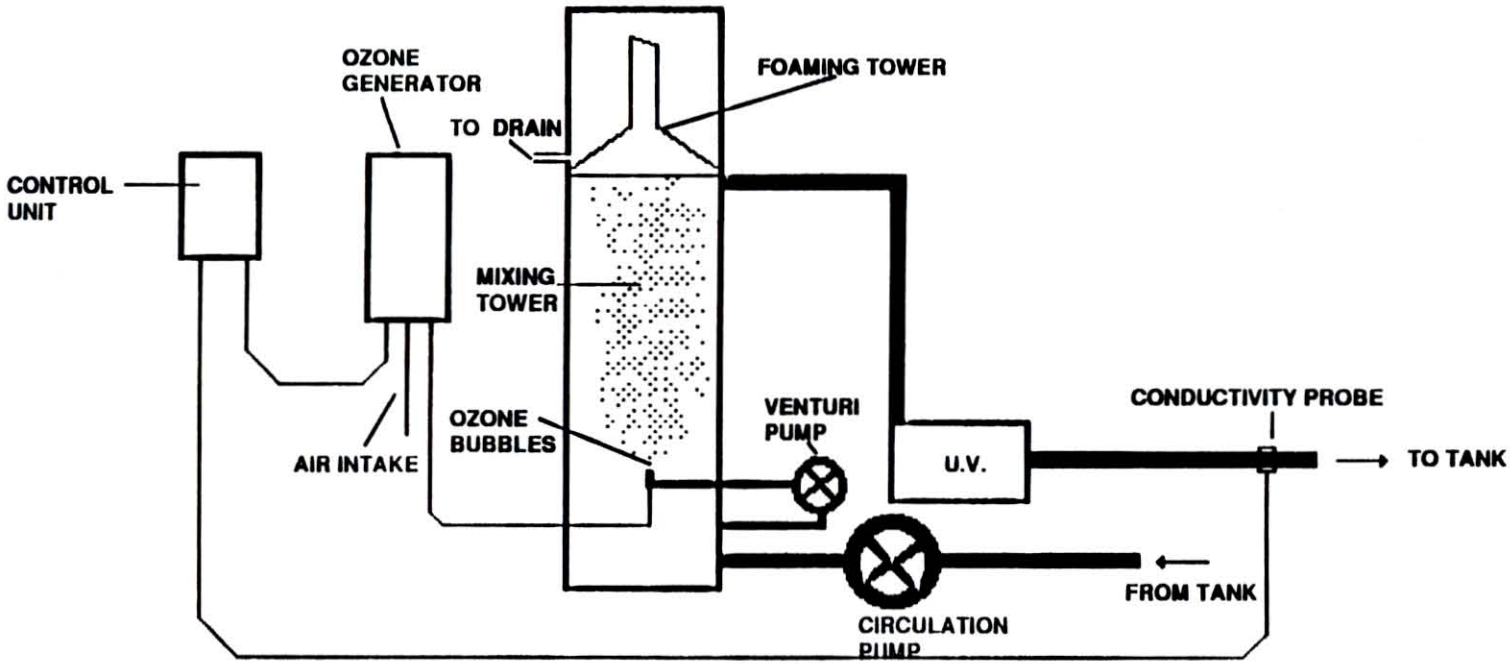


Figure 10. Diagram of ozone system.

essential to provide sufficient contact time for the ozone to work. At the top of the tower foam collects and is removed via a foam tower (or skimmer). The water is then passed through a U.V. system to remove any residual ozone (as ozone is toxic to shellfish). A conductivity probe is fitted after the U.V. to ensure that the ozone has been removed. This is linked back to a control panel which in turn can control the ozone generator. The tower should be large enough to give a contact time of between to 10 minutes, depending on ozone dosage. In shellfish purification ozone levels of between 0.5 - 2g/m^3 are used. Shellfish will tolerate small amounts of residual ozone whereas finfish are highly sensitive.

Ozone has potent germicidal properties which are attributed to its high oxidation potential. It has a direct effect by disintegrating the cell walls of bacteria and viruses.

A major advantage of ozone over other systems is that it can work on turbid water it also improves the taste, odour and colour of water and can also destroy organic traces. This fact is being used in trials using ozone for the depuration of D.S.P. contaminated shellfish.

The design of the mixing tower with a protein skimmer will also be functioning in removing unwanted wastes and bacteria etc. as with the aeration system, foam is being produced and continually being removed. Another advantage of the ozone is it very efficiently oxygenates the water. The main disadvantage of the ozone system is its initial capital costs. Though the ozone generators themselves are relatively cheap the control boxes etc. tend to make small systems expensive. Larger systems become proportionally cheaper as one control box can regulate several tanks. Every year ozone systems are becoming more refined and cost effective. There are now U.V. tubes that make ozone as a by-product. It is important that the ozone system is correctly installed and vented properly as ozone is toxic to humans.

Irradiation

Considerable work is being carried out in America in the use of irradiation in the purification of shellfish. As stated previously depuration systems only remove bacteria and viruses that can be quickly purged (i.e. in the gut or to a certain degree the haemolymph [or blood]). If viruses and bacteria are in the tissues then depuration for 48 hours will not be sufficient. In certain cases shellfish tested clear for F. coliforms after being depurated have never the less caused food poisoning or other illnesses. This is because there are still viable viruses or bacteria present. In America in the Gulf of Mexico the oyster industry has had to be shut down for over six months because of a bacteria, Vibrio vulnificus, which cannot be purified by traditional systems and which can cause death in immunocompromised people.

In light of this there are only three courses of action available to people dealing with shellfish from areas that have viral or vibrio problems. Firstly relaying for long periods of time in clean water; secondly cooking/processing all products by a suitable procedure that will kill the viruses etc.; and thirdly trying to kill the viruses and bacteria in situ, in the shellfish, while leaving the shellfish alive.

To date irradiation trials using low dose Cobalt 60 gamma radiation at levels of 0.85 K Gy have been relatively successful.

Radiation is used extensively in some countries in the fruit and vegetable industries to sterilise their products. This is the technology that is currently being adopted for shellfish. The gamma radiation penetrates the shellfish and kills the viruses and bacteria. The radiation does affect the shellfish but not enough to kill it. To date the oysters have had a two week shelf life after irradiation treatment with complete elimination of viruses etc. It should be noted this work is still at an experimental stage.

MULTILAYER AND DOWNWELLING SYSTEMS

The development work and testing for both the multilayer and downwelling systems described below was carried out by Seafish Industry Authority in association with members of the British Shellfish Industry. (Reports in relation to various trials are available from Seafish).

The multilayer system is currently available in three models sizes, a 750 kg. unit, a 1.5 tonne unit and a 3 tonne unit. The system is designed as a modular system so if you want a 24 tonne capacity per run you buy eight 3 tonne units. Once the tank is built to the right specifications and installed correctly it should perform efficiently and should be easily passed by the local authorities responsible. Because of the construction and self contained nature even the 3 tonne units can be moved if required with only the water supply pipes usually needing to be disconnected.

The design of the 750 kg. unit is illustrated in fig. 11. The small units are usually constructed in G.R.P. (fibreglass) whereas the larger ones are constructed in 316 stainless steel.

The main noticeable feature of the system is the perforated stackable baskets. These baskets hold approximately 15 kg. of shellfish in 7.5 cm layer. The baskets are deep enough that even when the shellfish open there is at least 7.5 cm free space between layers to allow an adequate water flow. The 750 kg. models hold 50 baskets, they are stacked five high, two abreast and five long (fig. 12).

Unfortunately they have to be loaded and unload manually. The larger systems are designed to take baskets on pallets. The 1.5 tonne unit takes four pallets in a single line, whereas the 3 tonne unit takes 8 pallets (2 wide, four long). Each pallet contains 24 baskets (four baskets on the base and six high). The pallets can be mechanically loaded and unloaded by either an overhead crane or an adapted fork lift.

By necessity (to take the stacked baskets) the tanks are far deeper than tradition purification tanks, yet the most important design feature of the system is that all layers get a uniform flow of water across them. This is achieved by a perforated flow screen situated at each end of the tank. The size and number of holes in the flow screen are designed so that a positive pressure is established on the water in flow side, thus ensuring an even flow of water over the whole screen.

In the small tanks there is a 3.5 : 1 water mussel ratio, in the larger it is 6.4 : 1 (i.e. for every 3.5 kg. of water there is 1 kg. of mussels). The small tanks require a flow rate of approximately 9 M^3/hr. (approximately 3 volume changes) whereas the large tanks can operate on one change per hour (approx. 12 M^3/hr and 24 M^3) if extra aeration of 20 L and 40 L of air per minute is added. If air isn't added the flow rate has to be increased in order to keep oxygen levels over 50% saturation throughout the tanks. If tanks are built longer than four pallets it is found that the oxygen levels cannot be maintained to the end of the tanks. Aeration is achieved by the use of a spray bar alone on the small tank and by a spray bar and

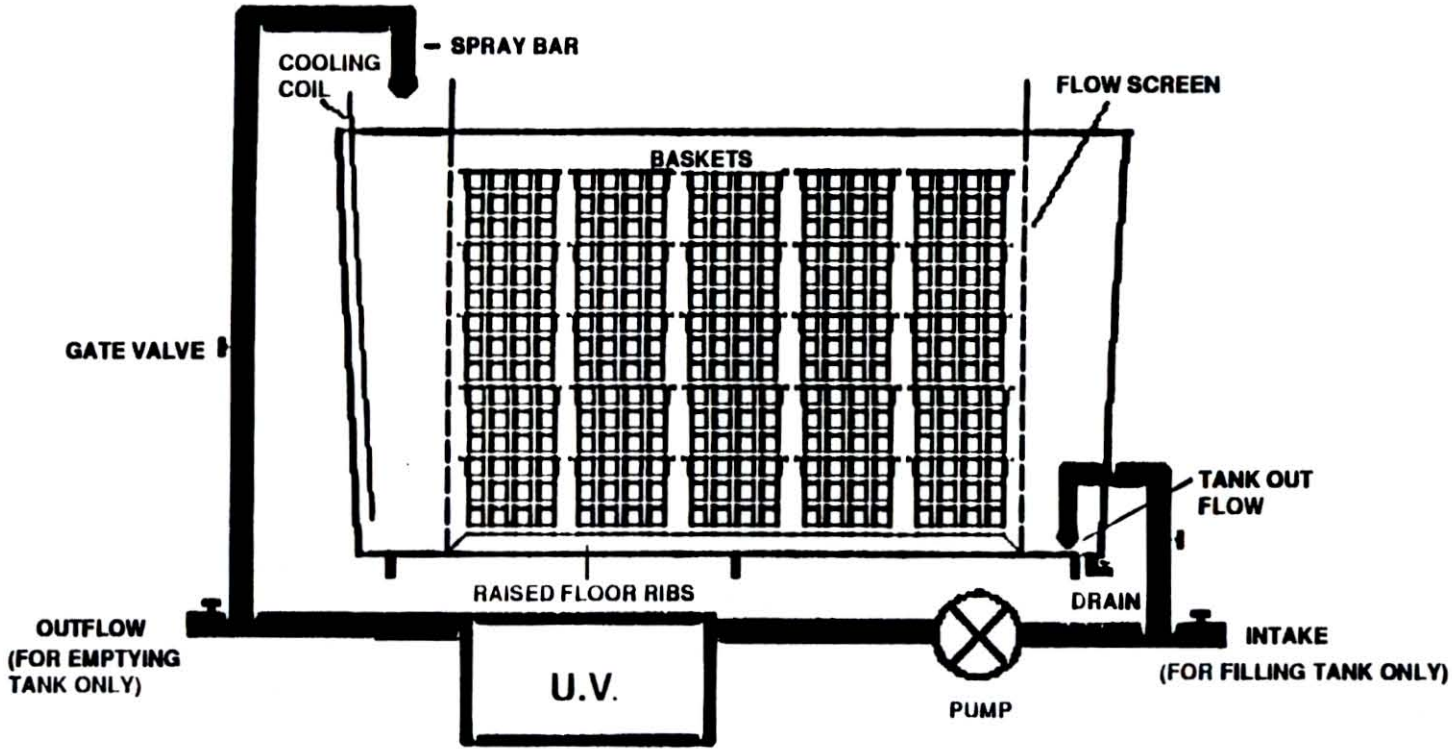


Figure 11. Diagram of multilayer system.

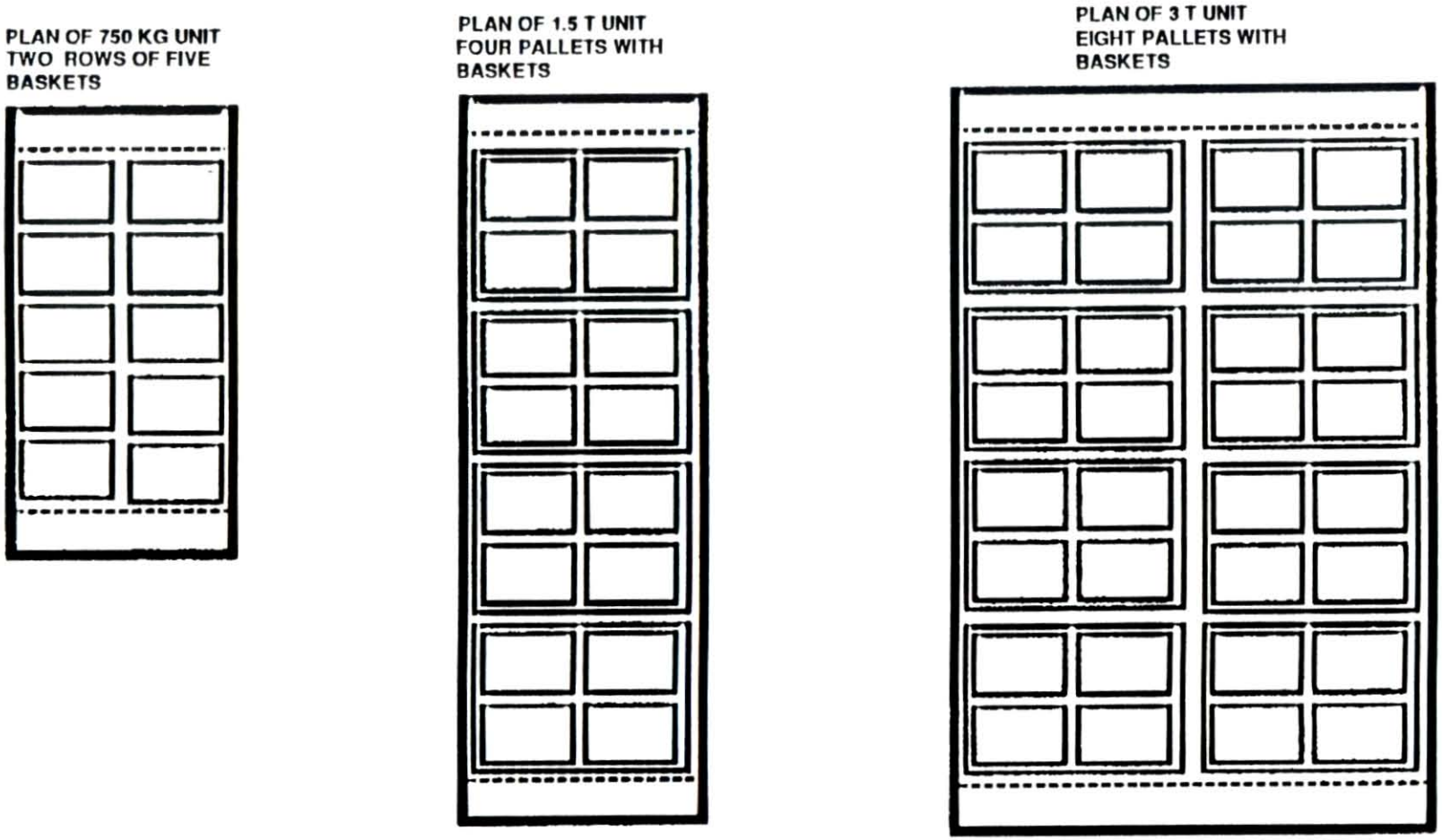


Figure 12. Diagram showing plan basket layout of the various multilayer systems.

compressed air on the larger tanks.

The tanks are designed so that detritus collects on the bottom of the tank in a dead spot that is not disturbed by the recirculating water. The baskets are held above this dead space. Reingestion of detritus from upper layers is not a problem due to the high flow rates and the eventual accumulation of much of the material at the bottom of the tank.

The temperature of the tanks can be controlled by either indirect chilling of the water or by controlling the ambient air temperature. The first option works best for GRP tanks because of their inherent insulating properties whereas the later works well for stainless steel tanks.

Advantages of multilayer system.

- Suit all types of shellfish
- Very efficient per square metre floor space.
- Modular system permits great flexibility in running different batches of shellfish through.
- Tanks easily moved and stored (if purification requirements are only seasonal)
- Easily heated or chilled.
- Mechanised loading and unloading for pallet systems.

Disadvantages.

- More expensive than concrete tanks.
- Require higher flow rates, this implies higher pumping costs.

Artificial Seawater.

In several of the plants where the above systems were installed artificial seawater was used due to the unavailability of good quality sea water.

Five salts are used (as defined in MAFF Laboratory leaflet No. 39). The following amounts are mixed in 1000 litre of water to give a final salinity of 27%, (27% is used as it allows for a certain amount of error which most shellfish species can tolerate).

Sodium chloride	Na Cl	21.08 kg.
Magnesium sulphate	Mg $S0_4$	5.18 kg.
Magnesium chloride	Mg Cl_2	4.12 kg.
Calcium chloride	Ca Cl_2	1.06 kg.
Potassium chloride	K Cl	0.50 kg.

The approximate cost to produce 1,000L is £6.00. For the small tanks, because of the high water shellfish ratio only three runs (1 week) can be carried out with the same water (with a 10% replacement after each run) mainly due to the water

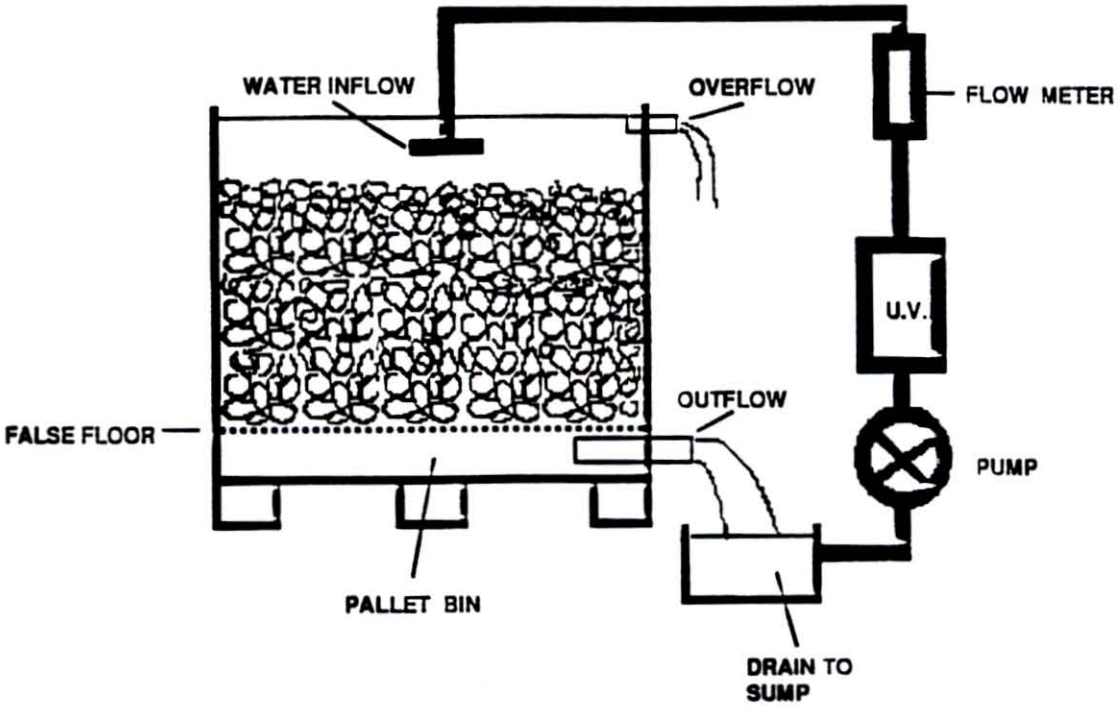


Figure 13. Downwelling bin system.

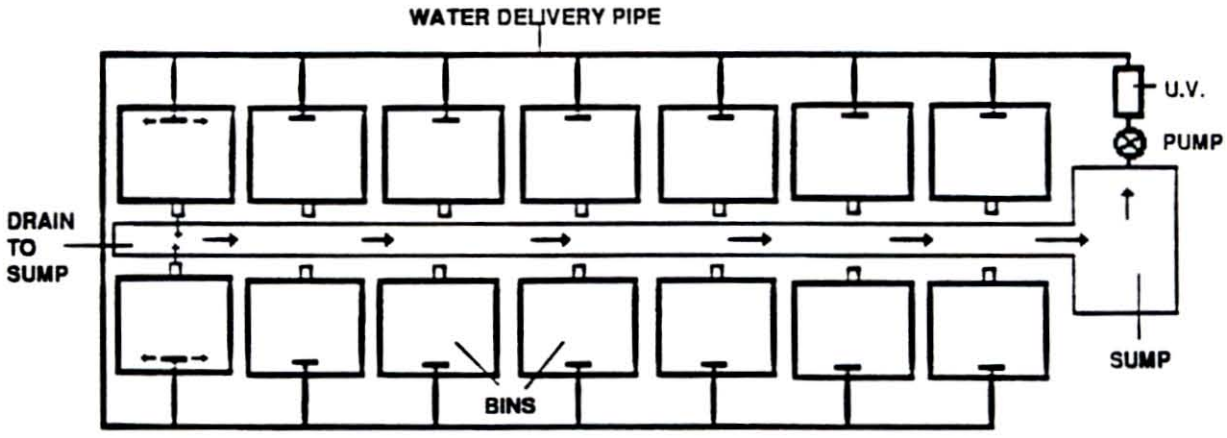


Figure 14. Diagram of linked downwelling bin system.

UPWELLING AND AERATION SYSTEMS

As stated B.I.M. carried out trials on developing an upwelling bulk bin purification system for mussels while Seafish worked on the downwelling system.

Most of the development work was carried out using a prototype system. However, commercially available insulated bins with false floors were readily used for the system (fig. 15).

As with the downwelling system the bins are linked with detachable hoses and over-flow into a common sump. Oxygenation occurs when water cascades over the sides of the tanks. It is very important that the bins and sump hold at least a 3:1 ratio of water to mussels. A lower ratio leads to oxygenation problems etc.

Bins are pallet sized and are loaded with approx. 350 kg. of mussels - to a depth of 40 cm when mussels are closed. Water is pumped via a pipe to the bottom of the tank and rises up through a false floor which acts as a flow screen to evenly circulate the water. Once filtering and open the mussel depth will increase to over 60 cm. The oxygen levels are highest at the bottom of the tank where the water enters and decrease as you go up through the mussel layer. Flow rates of $6M^3$ per hour were required to keep mussels oxygenated sufficiently. As the bins contained approximately 1 M^3 water this equates to six changes of water per hour.

Cooling or heating can take place in the sump if required. Also because the insulated bins can be stacked one on top of the other the system is extremely space effective (fig. 16). This means temperature control of an enclosed room holding the tanks is also easily done.

The system was able to produce a hundred fold reduction of bacteria within 48 hours, giving reduction levels from over 400 f. coliforms/g to under 3 and even 0. Contamination levels in the B classification range were reduced to below detection levels with 24 hours. It was found that best results were obtained by back flushing the tanks twice in 24 hours. This problem occurred because the flow is opposite to the gravity direction and heavier detritus did not flow over the top of the tanks. Back-flushing and washing improved cleaning times and was also incorporated into a training cycle.

As with the downwelling system the ability of the bins to be loaded on boats etc. is very labour efficient. One advantage of the upwelling bins is that they can operate as a holding system with a reduced water flow rate through them.

It should be noted however that bin systems can easily be designed to be interchangeable between upwelling and downwelling.

Advantages of the upwelling system:

- High density per square metre (over 1 tonne per m^2 achievable.)
- Modular system
- Transportable containers

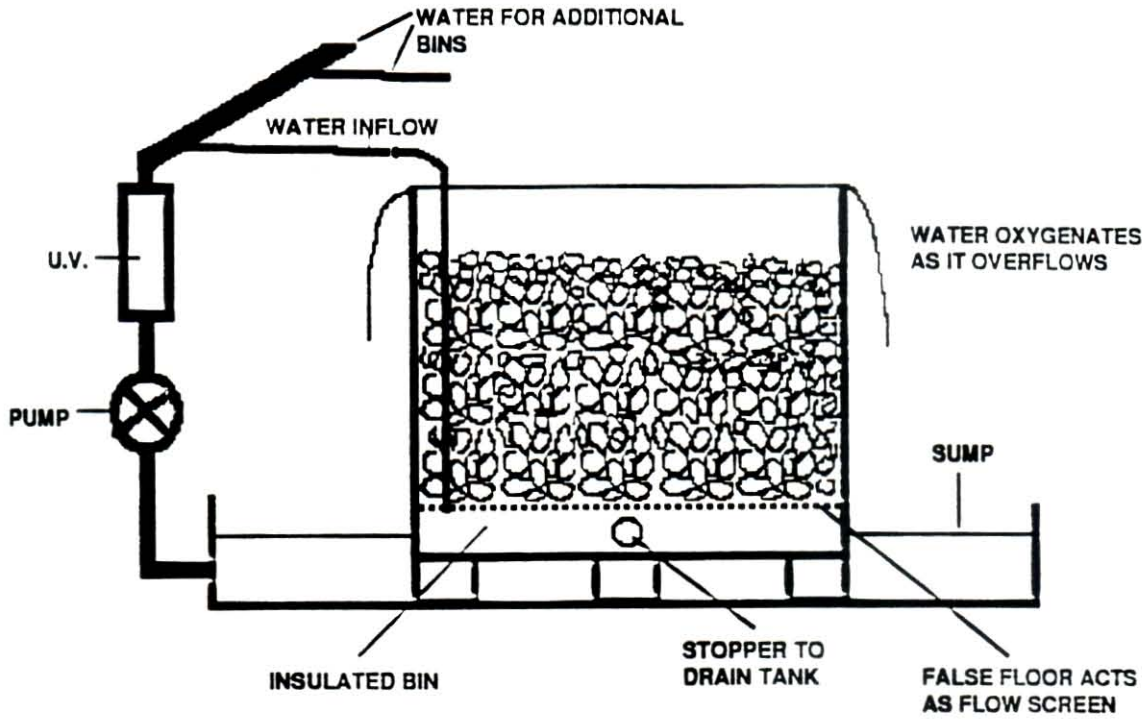


Figure 15. Upwelling system in an insulated bin.

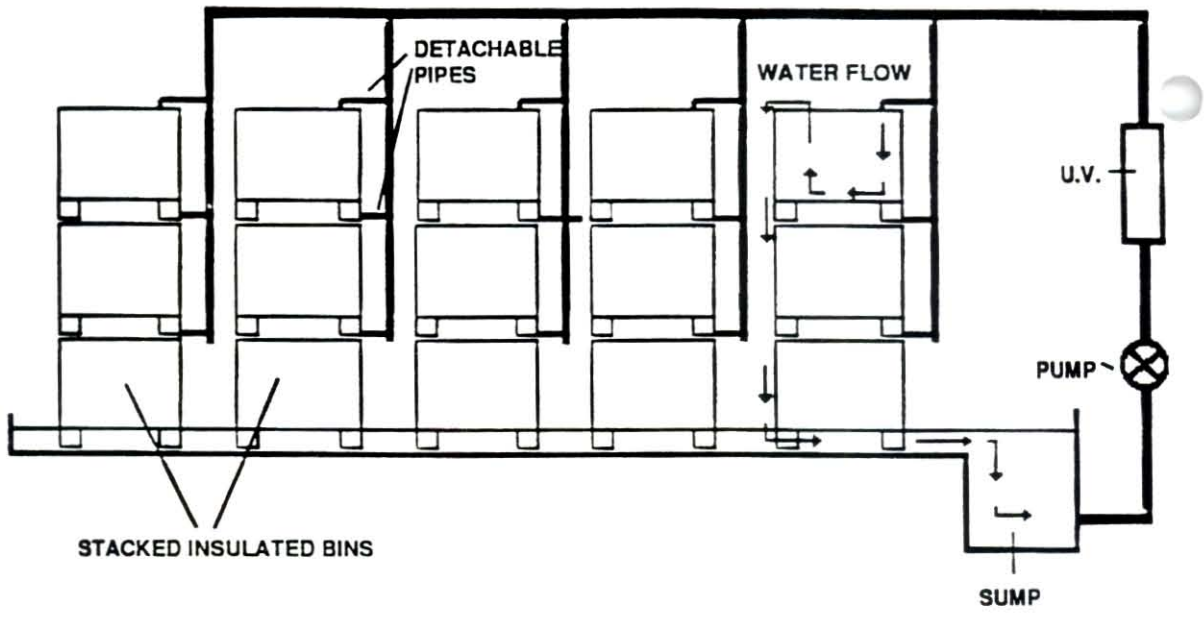


Figure 16. Diagram of stacked upwelling bins.

- Efficient holding system for other species of shellfish and crustacea.
- Well oxygenated water
- Space efficient
- Will hold water and can act as a portable holding system.

Disadvantages

- High pumping cost
- Suitable mainly for mussels (for purification)
- Breakdown of pumping will quickly effect mussels (does not automatically drain down like a downwelling system)
- Detritus removal not as efficient as down welling.

Aeration Systems:

Traditionally aeration systems have been used in areas where the quality of the surrounding seawater has been good and have mainly been used to purify products grown in other areas.

The system is not a pumped recirculating one but a batch system in that tanks are filled (usually at high tide) and the water retained for 10-12 hours and then the system is drained and refilled (twice a day).

The traditional tanks were usually less than one metre deep and shellfish were stacked in one or two basket layers.

Water circulation in the tanks is achieved by aeration. Various types of aerators have been used (fig.17). The most efficient for purification purposes is the venturi system.

The aeration system functions in the following manner. Contaminated shellfish naturally eliminate bacteria etc. (as with other systems). A certain proportion of the bacteria is associated with detritus and falls to the bottom of the tank as silt etc. As the intake water was originally clean it becomes contaminated with eliminated water borne bacteria. The air bubbles produced rising up through the water trap bacteria and other water soluble particles and substances on them (due to physical properties of surface tension etc.) which become trapped in the foam. A certain amount of bacteria is also still present in the water. Natural bacteria mortality and U.V. light from the sun eliminates some of these bacteria.

Once the tanks are flushed and emptied and refilled with clean water a considerable amount of bacteria etc will have been eliminated and the whole process is repeated.

IFREMER have done considerable work on improving these systems (fig. 18). By the use of directed venturi pumps good water circulation has been achieved and the small bubbles are more efficient in foaming and trapping bacteria. A foam drain has also been incorporated into the system so that contaminated foam is continually being removed. Also sterilisation of incoming water by U.V. etc.

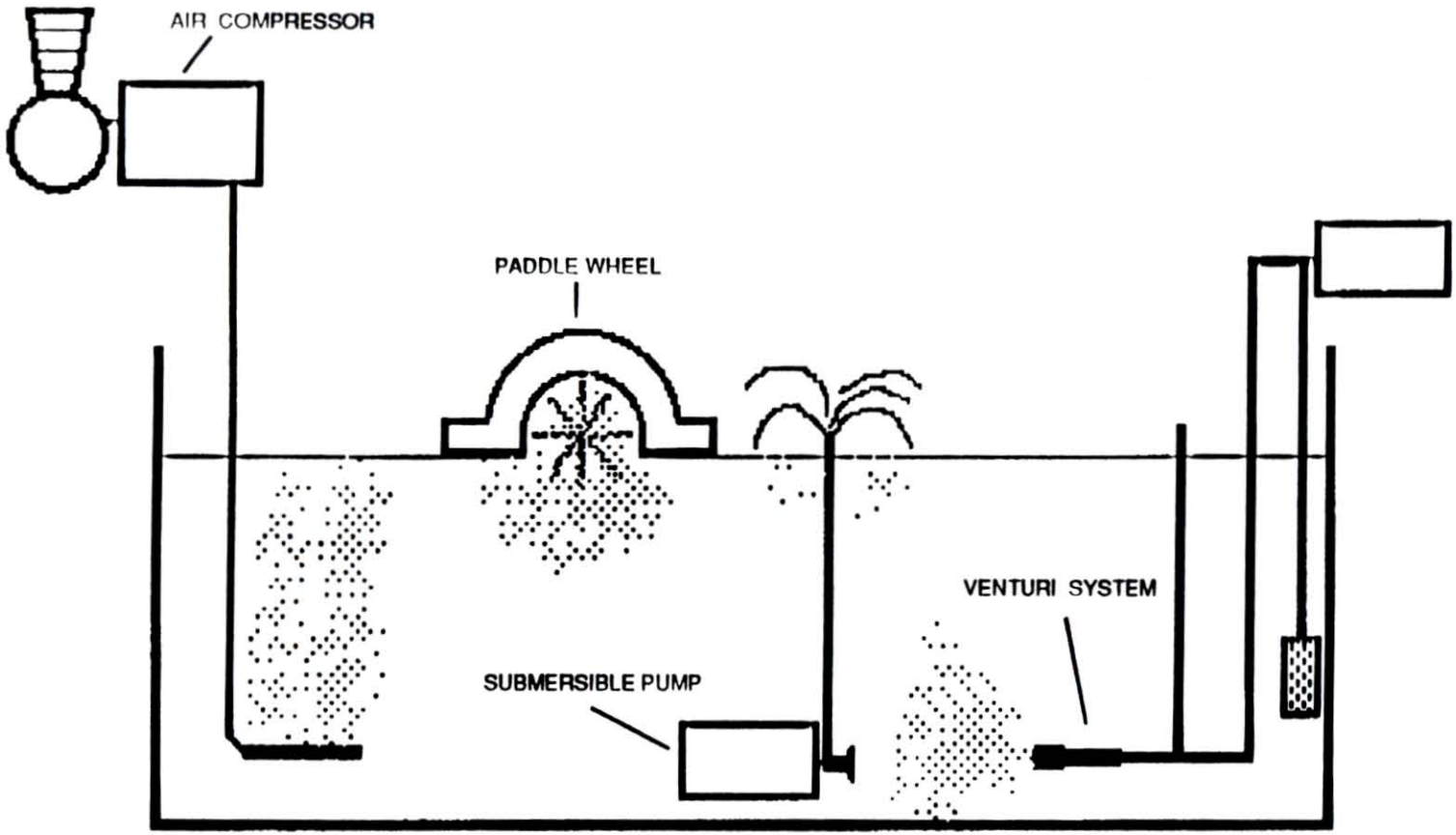
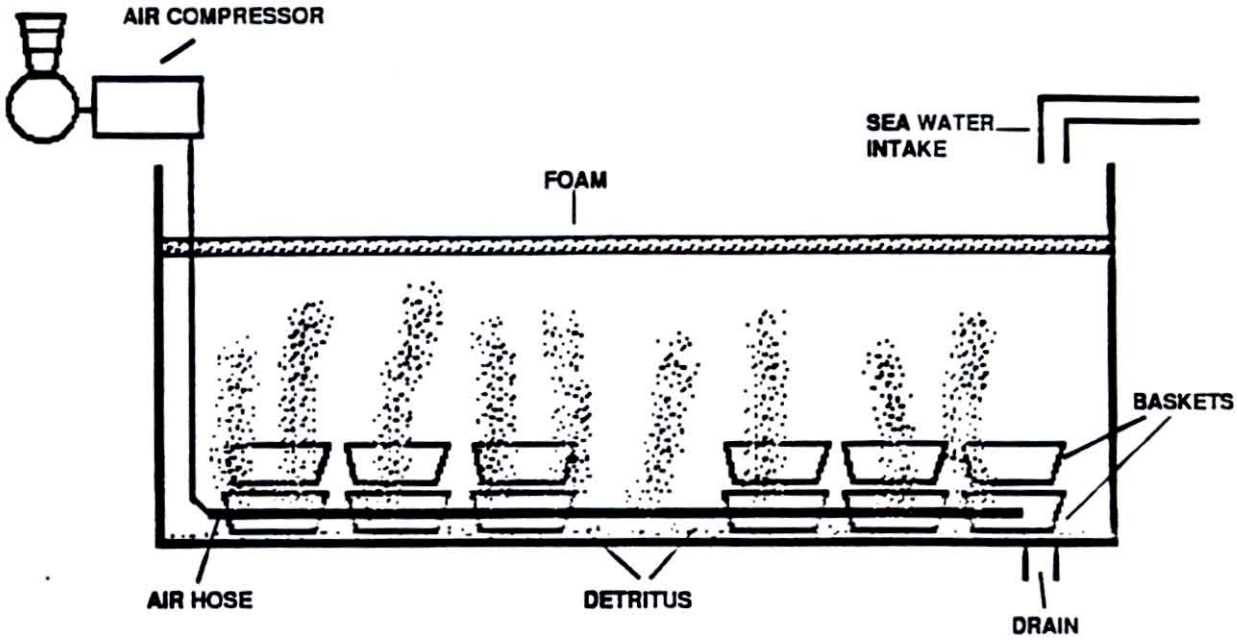


Figure 17. Various types of aerators.



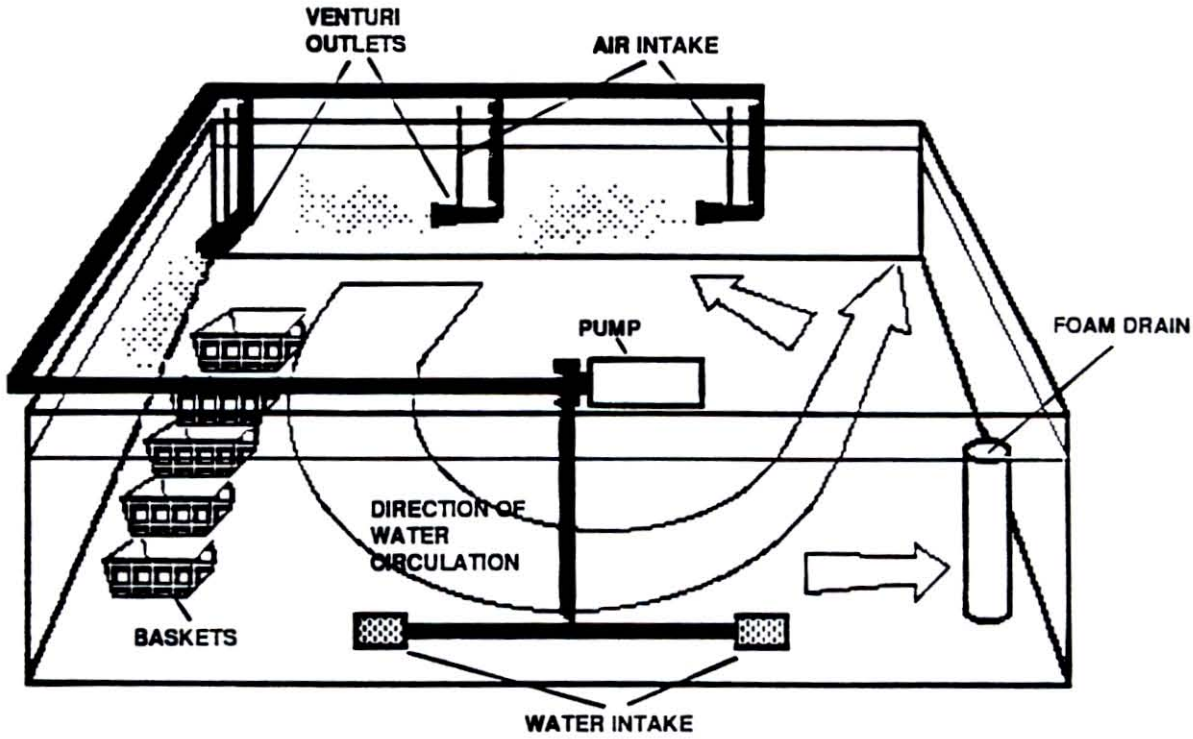


Figure 18. Old aeration system (above) newer aeration system (below).

guarantees the bacteria quality of the water.

Further work is being carried out to use deep tanks and large scale perforated bin holding systems which would improve loading and unloading labour.

Advantages of aeration system:

- Very cheap to run
- Very efficient holding system for all types of shellfish (as new batch of water will contain food particles)
- If new containers prove effective the system will allow high densities per metre squared.

Disadvantages:

- Bacteria and viruses not actively destroyed
- Care needs to be taken to avoid re-contamination by not disturbing silt with aeration
- Water flow in the tanks needs to be checked, incorrect packing and stacking of trays could reduce water flow over shellfish
- Covering tanks and placing them indoors could decrease effectiveness of system (due to less natural U.V.).

Summary of Systems:

In essence all purification systems should do the following:

- Allow shellfish to open and filter
- Supply seawater at correct salinity and temperature range
- Avoid resuspension of detritus
- Avoid physical disturbance of shellfish by water cascades, too turbulent water flow, too vigorous aeration, too close proximity to vibration causing machinery
- Avoid strong sunlight and extreme fluctuations in ambient temperature
- Once bacteria etc. voided by shellfish recontamination avoided. Water sterilisation systems achieve this, but also bacteria being trapped in foam and silt detritus is also effective.

  N.B. If most recirculating systems are run with their sterilisation systems switched off a ten-fold reduction is usually achievable within 24 hours due to foaming, natural die off of bacteria and bacteria being trapped in the detritus.
- Provide sufficient oxygen.

Oxygenation

To be able to actively filter it is recommended that shellfish have oxygen levels no lower than 50% saturation (5mg/L oxygen); so the worst part of the tank should

have at least 50% saturation. At the water input point 100% saturation should be achievable.

There are two ways of providing oxygen in tanks. Aerate the water directly in the tanks (need to avoid vigourous bubbling which would cause shellfish to close) or aerate the water being circulated (i.e. by cascading weirs or spay bars).

At the start of a purification run when the shellfish have just been reimmersed in water there will be a heavy oxygen demand and oxygen levels will drop (even below 50% saturation). This is normal and should settle down after approximately one hour, if it does not settle then something is wrong.

Shellfish use up the oxygen dissolved in the water. If we assume that 1 kg. of shellfish will use 24 mg/kg/hr of oxygen then if we take a tank containing 750 kg. of shellfish they will require 18,000 mg/hr. of oxygen (or 18 g). If water entering the system has approximately 7 mg/L of oxygen (that means 7g per cubic metre) and if only 2 mg/L is easily available to the shellfish (this would bring the dissolved oxygen content of the water down to 5 mg/L, the recommended lowest level. This means that one cubic metre of water would provide 2g of oxygen, to get 18g of oxygen this means a flow of 9 cubic metres per hour would be required in the tank and this is how much water the small multilayer tank requires for a load of 750 kg.

The second way of supplying this oxygen is to reduce the flow rate but to provide oxygen directly to the water by aeration. So if the flow rate was halved to 4.5 cubic metres an hour 9g of oxygen would have to be provided to the water by direct aeration in the tank. It must be remembered however that high flow rates may be required by the tank design to ensure that proper and efficient water circulation takes place in the tank.

An important fact to remember about oxygen levels is that shellfish are live animals whose oxygen consumption rate can vary enormously depending on how active they are. Temperature plays an important factor in this. As temperature increases the amount of oxygen dissolved in the water also decreases. To add to this, as temperature increases so does the activity of the shellfish, so they also require more oxygen. This occurs up to a certain temperatures which varies from species to species, once this temperature is reached the animals start to become stressed and their filtration activity starts to decrease. So oxygen levels, avoiding risk of spawning and maintaining optimum filtration temperatures are the reasons temperature control in tanks is important.

SITING LOCATION AND PLANNING REQUIREMENTS FOR ESTABLISHING A SHELLFISH PURIFICATION AND DISPATCH CENTRE.

With the implementation of Council Directive (91/492/EEC) there was a need in Ireland for new purification and dispatch centres to be built and also for older facilities to be modernised.

However, one of the first new centres proposed ran into considerable problems from a planning point of view. To help the shellfish industry and the planning authorities surmount some of the issues raised by this case Bord Iascaigh Mhara commissioned the firm of Brady Shipman Martin (Environmental and Landscape Planning Consultants) to produce "Shellfish Purification and Dispatch Centres - Location and Design Guidelines".

This lecture is just a quick summary of the important points covered in the report which is available from B.I.M. (All course participants will be supplied with a copy).

The report was sent to all coastal County Councils and also to people in the industry planning to build or upgrade a facility. Basically its function was to inform the planners and industry of each of their needs.

The main problem is one of conflict i.e. scenic amenity -v- practical needs of an industry. Planners and indeed many of the general public do not want industry in scenic coastal areas. Planners usually zone industry into discreet areas. However, the shellfish industry does not suit the general industry classification. Indeed the Council Directive (91/492/EEC) tends to force shellfish plants to be built away from such centres of industry because of the need for clean environmental conditions etc. required to meet health standards.

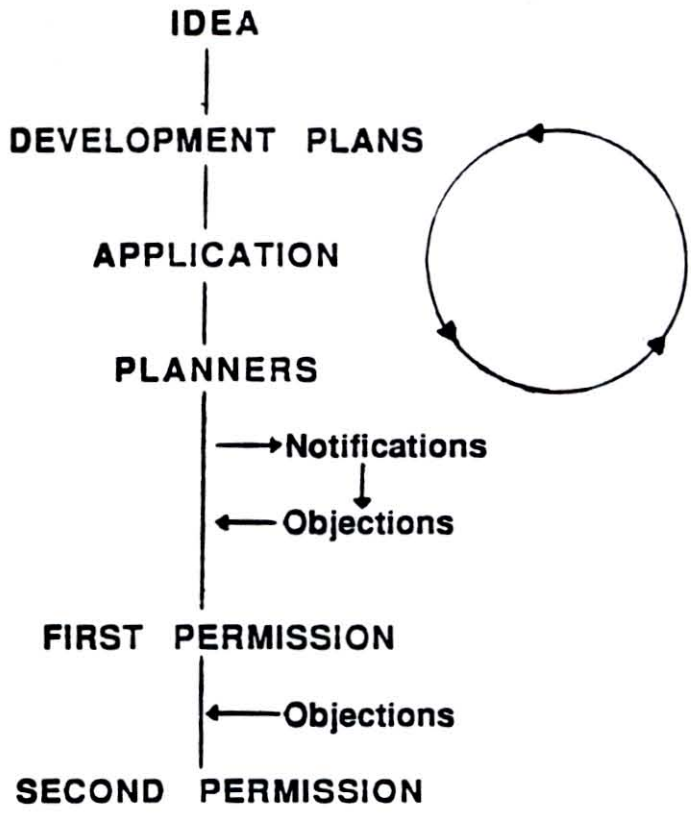
The shellfish industry requires centres -

- close to production areas
- in a clean environment
- with access to suitable sea water
- adequate land and sea access
- and meeting E.U. and National regulations.

This usually means siting the centre on the coast in between the coastal road and the sea. Unfortunately for the Irish Shellfish industry there is a generally accepted planning rule that in scenic coastal areas no building can be built between the road and the sea. In counties Cork and Kerry up to 90% of the coastline is classified as scenic amenity areas. This has caused a severe problem for the Irish rope mussel industry as approximately 80% of the national tonnage is produced in these two counties.

Therefore it is essential that any application for planning is prepared as carefully as possible to avoid unnecessary and expensive complications.

Schematically the planning process can be represented as follows:



It should be noted that in assessing a planning application the planners do not look at the proposer to see if he is a nice person but at what you present to them in the application. In assessing an application the planners look at:

- designation of site in county development plan
- the capacity of existing infrastructure - roads, water, etc.
- risk of traffic hazard - i.e. increased truck movement etc.
- pollution control - effluent - process
  - septic tank
  - solid wastes
  - smells etc.
- visual impact
- existing land use
- relationship of design to the environmental character of the area
- case specific factors

Successful applications usually have good site selection that is consistent with the areas development plan policies and a quality of design that reflects the location of the centre and addresses the environmental issues.

Communication with the planning authorities prior to submitting an application is very important. The planners will be able to tell you where they have problems with

your proposed submission. With a competent design team you should be able to come to a compromise that will suit both parties.

Things such as changing the roof type can make the building look smaller or blend in with the environment. Site screening, good use of plantings and landscaping also can counter many objections. The size of the site should be adequate to take hard standing areas and storage areas. Unutilized portions of the site should be well maintained for a site that is well presented and managed with emphasise the quality of the product.

In summary, basic rules to follow in relation to submitting a planning application for a shellfish purification or dispatch centre are:

1. Keep it simple
2. Take care in site selection
3. Discuss intentions with Planning Officer/Development Officer
4. Appoint a competent design team
5. Answer all questions
6. Deal with ancillary items, outside stores, pump houses etc.
7. Allow for future development
8. Consider and explain site management
9. Anticipate an appeal
10. Keep it simple

BUILDING DESIGN, LAYOUT, GENERAL EQUIPMENT AND COMMISSIONING SYSTEMS.

Before a person should embark on approaching any architects or engineers in relation to designing and building a new plant or modernising an existing one they should answer three questions that only they themselves can answer:

(1) What is your business?
(2) What are your future business plans?
(3) Can you afford the development and will the costs be justified?

All to often a person's answer will be something like "I want a plant that can purify two tonnes of shellfish a week". When they are questioned why they want that size plant they answer is invariably "Oh, I produce about 100 tonnes a year". The assumption is made that divide production by 50 and that will give them what they need to handle a week. This concept is completely wrong. Conversely designing a facility to handle a large tonnage that you may only throughput for one week in the year is also incorrect as the plant may be left idle for most the year.

In an ideal world it would be nice to have a steady weekly stream of shellfish. In reality things such as spawning, "red tides" and market trends dictate supply.

Even considering the above scenario people are not answering the question "what is your business"? Do you grow shellfish or harvest them? Are you an agent? Buying shellfish from growers and fishermen and then on selling to wholesalers. Or are you a wholesaler who buys in shellfish, repacks it and sells it directly to the retail outlets. Your business may be any or all of these.

The shellfish business can be broken down broadly as follows:

Depending on where you currently fit into this scheme and also where you intend to be several years down the road will determine the building and layout that you will have. Usually unless someone is carrying out exactly the same business as yourself in the same type of location, the needs and hence design requirements will be different.

Generally as companies become bigger and more successful there is a natural tendency for growers to expand down the line into direct marketing and conversely for marketing companies to expand backwards into ongrowing in order to consolidate supplies.

Even if you take for example two growers who both decide they wish to be able to purify their product; both may produce 500 tonnes per annum. However, if one grows oysters while the other grows rope mussels their needs and hence the buildings will be completely different, as well as the amount of money they can expect for their respective products. Where it may be financially feasible for the person with the higher value product to build a plant it may not be for the person with the lower value product. However this aspect will be dealt with in more detail later.

The building design and equipment required will also reflect what type of market you intend to supply i.e.

- Bulk tonnage
- Final packing carried out in another plant
- Local markets or export
- Value added
  - wholesalers
  - supermarkets.

Generally the uses of a typical building would need to cater for the following:

- Grading/handling for:
  - Ongrowing. If your business is production based, then a land based work facility for making equipment, grading seed, etc. may be a requirement. This area is basically a work shed and would not need to be up to packing station specifications as the shellfish will be returning to the sea for further ongrowing.
  - The initial wash and grading prior to being loaded into purification tanks.
- Holding tanks. If people are working the tides a simple holding tank where shellfish can be held for several days before they are handled etc. can be very beneficial. These need not be indoors or covered.
- Depuration centre. The area where purification is carried out.
- Packing and dispatch centre. Sterile/clean area where shellfish are washed, graded and packed ready for dispatch.
- Equipment store and workshop. Covered area for machinery and equipment used generally in relation to business.

- Laboratory for product testing.
- Office/reception.
- Canteen and staff facilities

See fig. 19 for a general purpose building design.

The important point to remember when designing a facility is that your product is virtually unique. You are dealing with a food source that is a live animal which usually has to be handled and presented to the consumer alive. Yet you have to conform to the practices and regulations designed for the general food industry. You are also dealing with salt water, sand and shell-grit, which when combined are abrasive and corrosive and therefore the quality of materials that have to be used in the building itself and the equipment have to be able to withstand conditions not usually found in the food processing industry.

It is nearly essential now to adopt the policy that is carried out throughout the food processing sector. That is a one way system, this reduces the risk of contamination of the product.

When you are designing your building it is essential that you contact the relevant local officials who will be inspecting the building to see if it conforms to the E.C. and national directives and regulations. The plans should be discussed in detail with these people and such things as wall and floor finishes, wash room facilities etc. should be agreed if possible for it will be extremely expensive if you go along and build the centre and are then told it will not be certified because they are not happy with the finish on the wall in a particular area.

If however, you feel some of the demands are excessive and to a much higher standard then other plants, you do have a recourse of action. The E.U. directives are meant to establish uniform standards and it could cause an unfair trade advantage if one country is stricter than another if this is the case you would have recourse to European Law to argue your case. (This however should only be worthwhile if major investment costs are in question).

In addition it is very worthwhile for yourself and your architect/engineer to go and visit and talk to people with existing facilities whether at home or abroad who will be able to point out mistakes, etc. they themselves have made or indeed things that worked very well. Visiting many seem to be unnecessary but thousands of pounds can be saved by spending a few hundred at the right stage.

Appointing a competent design team is also usually very worthwhile. For it is not just getting a plan drawn up by a draughtsman who may be a personal friend. Don't forget you have to go through planning permission procedures, the construction phase where work has to be inspected and checked, equipping the facility and finally turning everything on and seeing if it works.

Unfortunately one of the most important aspects of a shellfish facility tends to be given the least thought and in the long run can cause the most problems. This is water supply.

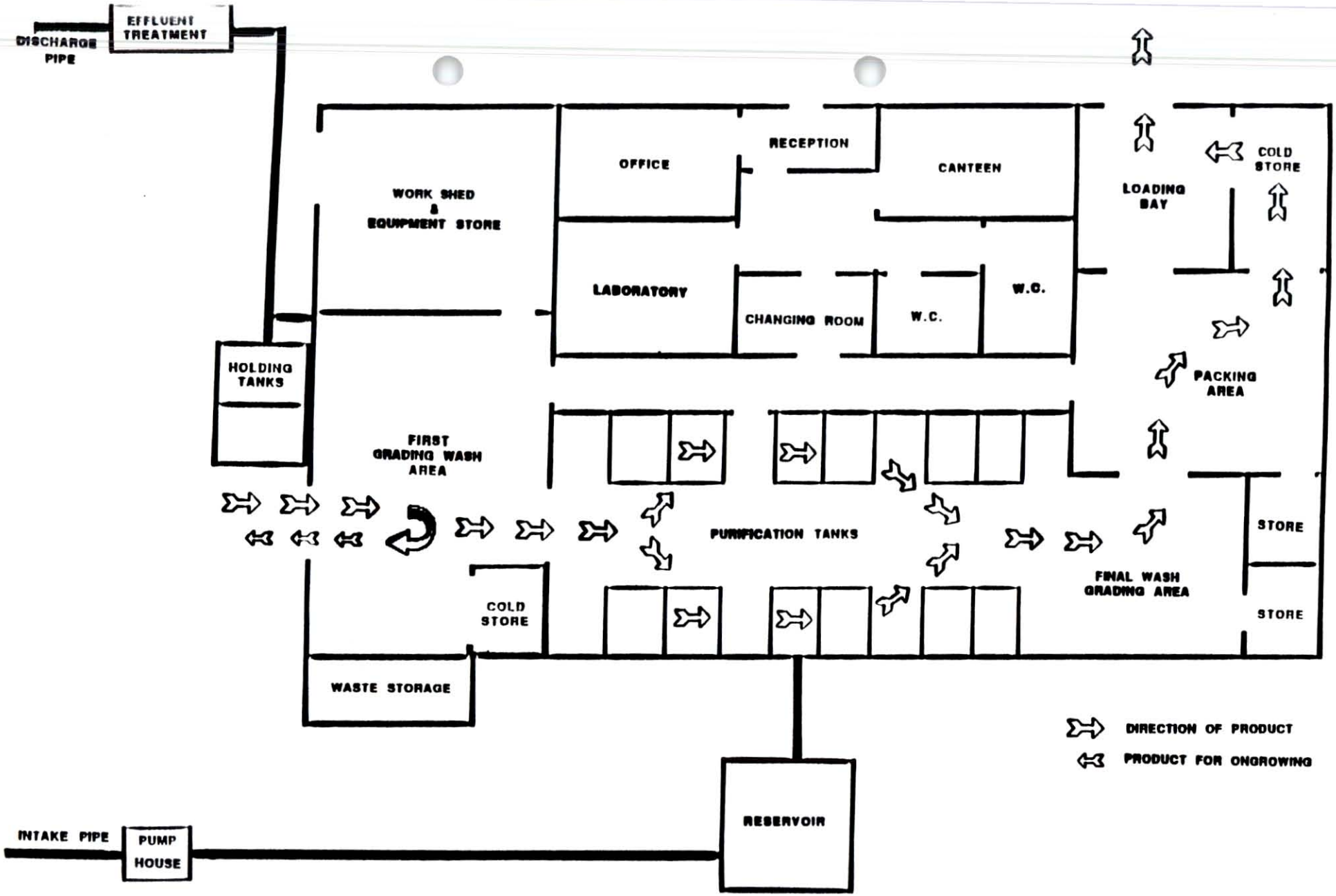


Figure 19. Diagram of a generalised depuration and dispatch centre.

Water requirements need to be calculated carefully, not just for the tanks but also for washing machines (some of which will use 30M3 per hour) and also washing down floors and tanks. Both fresh water and seawater supplies are required. It usually depends on the availability and cost of both which will determine whether fresh water or seawater is used for washing etc.

What the water is used for may also dictate whether or not it needs to pass through settlement tanks or filters etc.

Great care should be taken on siting your sea intake point (this is not relevant if the facility is designed to use artificial seawater). If possible it should be sited so as to satisfy as many of the following as possible:

- Maximum pumping time, making full use of tidal conditions
- Good salinity
- Clear not turbid water
- Temperature - during summer surface water can be 10°C warmer than deep water
- Avoidance of algal blooms
- Low bacteria counts
- Away from sources of contaminations (hydrocarbons etc.)
- Protected from possible storm damage, silting and clogging.

The type of pump used, whether submersible or not will depend on the head height. Submersible pumps are usually best if water has to be lifted any significant height otherwise self priming pumps or foot valves usually have to be used.

If possible the water reservoir should be large enough to hold enough water to last the centre for all its water needs for 24 hours. It should be sited (if possible) so that tanks can be fed by gravity feed. This is important if there is a power failure or equipment breakdown.

The removal of all this water is also very important. Ample drainage should be provided. The drainage scheme should be designed so that it is easily accessible for cleaning and unclogging pipes. As a requirement of planning permission it is not unusual for the effluent water to have to be treated, i.e. passed through a settlement tank, sand filtered and U.V. system. The discharge point should be away from the intake point. In areas where effluent pipes are uncovered at low tide it may be necessary (condition of planning) to retain the water in a holding tank until discharge can be made usually on the outgoing tide.

Whether you are operating in a purpose built facility or converting an existing facility the internal layout and use of space will go a long way in determining how efficient the plant will operate.

The flexibility and variability of your equipment lines may be a crucial factor if your markets and product specifications change. The ability to change quite often can be the deciding factor in the long term viability of a business.

Certain features on the production line will be fixed:

- Raw product entry point
- Final product exit point
- Chill rooms and freezer facilities (though newer types are easily dismantled and moved)
- Tanks, especially if concrete.

Again following a one way flow system will also dictate how a facility is laid out.

Usually before purification tanks there should be ample space to store new product in a protected environment. If purification is not require for every batch of shellfish then a chill room may be required at this point.

Depending on the state of the product, whether graded and washed etc. before reaching the centre, a wash and grading may be required before loading the shellfish into the tanks. It is recommended that strict grading be carried out at this stage for product graded out later will have been wasting space in the tanks, also graded product will have a chance to recover in the tanks which will improve shelf life of the product.

Product should enter the tanks in one direction and leave by another. Purified product must not be stored with unpurified product. After purification a second light wash and grade is recommended prior to packing. This equipment should not be the same washer etc. as being used for unpurified product. For there is a high risk of unpurified product being stuck during the first wash which could be missed when the machine is cleaned and which could contaminate a purified batch.

If different species are being handled then it may be advisable to have separate equipment lines. When laying out a line do not just think in two dimensions. Stacking equipment vertically can improve efficiency and save space. Once packed shellfish should be stored in an area ideally a chill room before being loaded onto the truck for dispatch.

Equipment

When considering equipment you should assess:

Is it needed? how will it improve your business? There are three basic reasons to purchase equipment:

(1) will it increase tonnage throughput; (2) will it improve the quality of the product; (3) will it save labour or just be easier on the workforce. Do not buy a piece of machinery because it looks good or because someone else has it. You would be surprised how often expensive pieces of machinery are hardly ever used once purchased because it didn't do one of the above points.

When purchasing equipment, price, though paramount at first, quite often will be

the least important point in the long run. Take care selecting the supplier. Is he just an agent for a company or is he part of a firm that has the back-up of a design team? Be wary of salesmen. The best course of action is to ask for the names of people who have purchased the same machinery. You should contact them and find out how the machine performed etc. It is also important to have good back-up service and supply of spare parts. Buying a machine from a firm that has a good design team can be important especially if you wish machines modified or customised later on.

Management:

Quite often the most important factor in determining whether a facility operates correctly or not, is the staff. The staff also only tend to be as good as the manager allows them to be. If you are the owner of a facility, especially if it is only a part of an overall business, you need to decide whether or not you are suitable to manage the depuration/dispatch centre. The whole

production process should be broken down into specific jobs and areas of responsibility, with the person doing a specific job being trained adequately to carry out that job.

The best facility in the world will be useless if it is not run correctly. The management and staff influence greatly how successful a venture will be.

Commissioning systems:

Once the expense of building or modifying a facility has been gone through it is vitally important that when all the equipment is switched on that it is tested to see that it is working properly. In addition staff will have to be trained in the use of the facility.

Not only will you need to satisfy yourself that everything is working and being used properly you will also have to satisfy the government officials. Getting an expert in at this stage to test your systems and fine-tune it will save money in the long run.

Seafish Industry Authority provide this service for that industry in the U.K. and they not only bring considerable expertise with them but also sophisticated equipment that can test your system very efficiently and which you yourself could most likely not afford to buy. They will also instruct staff on correct procedures etc.

Depending on tank design, spiking and clearance studies will have to be carried out to satisfy the government officials. These can generally be carried out as part of the service by the person commissioning the system.

OPERATING PRACTICES, QUALITY CONTROL AND HACCP

The recent Council Directive, (94/356/EC) laying down detailed rules for the application of Council Directive 91/493/EEC, as regards own health checks on fishery products, will be implemented over the coming year. This directive makes it mandatory that all premises handling fishery products, which include shellfish from aquaculture and fishing, establish an operational procedure commonly referred to as HACCP (Hazard Analysis Critical Control Points).

A brief overview of this topic will be presented here as it will be nearly essential for every depuration and dispatch manager to attend a specific detailed course in relation to this subject (i.e. a series of two day courses are being run, dealing solely with this topic, at various locations around Ireland for the fish processing and handling sectors).

In the past it has been recommended, but not obligatory, that correct operating procedures were documented and adhered to while running a depuration /dispatch centre. By adopting and recording such procedures, for example, the correct loading, filling and unloading of a purification tank, a manager could be confident that his workforce had adopted practices that would help guarantee and improve the quality of his product. Now however this has been taken a step further under this new directive and will be become mandatory in the near future.

It will be up to the 'persons responsible for the establishment' to design and implement a HACCP system which will be checked and monitored by the competent authority. Appropriate records will have to be kept and staff will have to be adequately trained to carry out the necessary jobs.

HACCP is an American concept whereby a process is broken down into a series of logical steps so that hazards and critical points for the production of any product can be identified and controlled.

A critical point is defined as; any point, step or procedure at which control can be applied and a food safety hazard can be prevented, eliminated or reduced to acceptable levels. All critical points must be identified and your own health checks must be developed and implemented. These will include set observations and/or measurements necessary to ensure that critical points are kept under control.

A sampling programme is not required for every batch but you must validated your own check systems when they are first set up and revalidate them if the product or process changes. Your sampling programme must also be verified at specified intervals.

You must document all information relating to the implementation of your own checks and their verification.

In essence what this all means is that you must:

- Put down in a logical manner, usually by means of a flow chart, the

complete process of what happens to a product, namely your shellfish, from when you first get it until you dispatch it.

- In this process important stages (critical control points) need to be identified which can affect the product quality and increase the risk of contamination; i.e. rough handling which causes shell damage; leaving a newly arrived batch of shellfish outside where it is exposed to strong sunlight and high temperature etc.

- Adopt procedures that reduce risk of product contamination or deterioration.

- Establish correct staff hygiene procedures.

- Checks or tests have to be established. These would be things like; having bacteria counts done on shellfish on arrival; checking the temperature of the chill room or work space; checking oxygen levels in tanks etc.

- Established and maintained a record keeping or documentation process.

- Train staff, so they can not only do a specific job properly but can also carry out and record specified checks etc.

- Establish a verification procedure. This means you have to be certain that if you are carrying out all your checks and handling procedures correctly that your product will meet the correct safety and quality requirements. In other words you have to be able to check the checking procedure to make sure it is working properly.

A sample hazard audit, prepared by Sea Fish Industry Authority (U.K.), for harvesting, purification and dispatch of live bivalve molluscs is included in the appendix. It should be noted that for each individual centre a specific HACCP audit and system will have to be established.

In relation to documentation the directive will require two types for the inspection by the competent authorities.

1 - Description of product.

- Description of the manufacturing process indicating critical points.

- For each critical point; identified hazards, assessment risks and control measures.

- Procedures for monitoring and checking at each such critical point, with indication of critical limits for parameters that need to be controlled and corrective action to be taken in case of loss of control.

- Procedures for verification and review.

2 - Records
  - Results of verification activities.
  - Written accounts of decisions relating to corrective action when taken.
  - Traceable documentation for an identified production batch.

- Flow diagram with sufficient technical data.

These directives and their implementation are not designed to make your life difficult. They are there to try and ensure that the consumer can eat a product and not become ill. It is recommended that if you are not completely sure on how to go about conforming with the above directives that you get advice from a reputable professional consultant or from your national development or regulatory body.

ECONOMICS OF OPERATING A PURIFICATION FACILITY

How economically viable it is to establish and operate a purification facility depends on many factors. e.g. What business are you in? What are your current operating practices and requirements? and do you need to improve to operate under E.U. Directives and national legislation?

If, for example, you were a shellfish dealer who used to operate off the seashore or buying off the pier and selling directly to retailers, you would be out of business if the E.U. Directives are strictly implemented because now shellfish have to pass through registered dispatch centres. Also with the stricter and more widespread classification of waters in respect to bacteria levels you would most likely also need a purification centre. So your choice is simple, either build a depuration/dispatch centre, become an agent for an existing depuration facility or just go out of business.

If you are already an established shellfish dealer with old facilities and need to upgrade to keep in business again you have very little choice in the matter.

The person who has the hardest decision is the grower or fisherman in a B classified area. If they do not build a facility their market options are limited for they usually either have to sell to processors or to people with depuration centres. Many growers in this situation feel they are exploited and it is this that drives them to establish a depuration/dispatch centre. However, the grower that takes that step should realise he is about to enter a whole new aspect of the shellfish business.

In 1992 B.I.M. commissioned a prefeasibility study for the establishment of a depuration/dispatch centre for Irish shellfish in France. The thrust of this was for the sale of rope mussels on the European market (Belgium, Italy, Germany as well as France). The costings used in the report are still not much different than those that would be obtained to-day. This study is interesting in that it is based on the centre buying in shellfish at the pier price in Ireland plus trucking costs and then selling through existing channels.

The study made a basic assumption that only 50 kg/m^2 of shellfish could be loaded into a tank. Two sized centres were costed. A centre with 180m^2 of tanks and an overall building space of 530m^2 which could handle 9 tonne per run giving a weekly output of 27t/week and a yearly capacity over 52 weeks of 1,404 tonnes and a total investment of £253,000. A centre with 280m^2 of tanks and an overall business size of 1,015m^2 could do 14 tonnes per run, 42t/week and 2,184 tonnes in the year and would have a total investment of £676,000.

Assuming 50% grants were available it was found that handling 1,000t a year of rope mussels only was hardly profitable, whereas dealing in 1,000t of mussels and 400t of mixed shellfish, including crabs and lobsters, had good results whereas if the combined tonnage is increased to 1,700t it has very good returns. Improving the density of shellfish per square metre would improve this slightly.

It is interesting to note that the two large scale depuration centres built in Ireland in

recent years will be operating along the above tonnage lines and handling mixed shellfish.

The above scenario was only a general feasibility study and should be taken as that.

If for example you are an oyster grower with a 150 tonne production unit you will need a building to operate your farm, to carry out grading and washing etc. Most farms try and have a small local market and also supply an export market. Adding purification tanks and a packing facility to a something you are going to have to build anyway will be relatively inexpensive and the operating costs of the centre are merged with the production and running costs of the farm. Again, an earlier study commissioned by B.I.M. in relation to oyster farming shows that it is viable and profitable for a farm with a 250t production to build a facility to purify and pack all his production etc. but only if he buys and handles an additional 250 tonnes of oysters giving the plant a total throughput of 500t per annum.

In reality in Ireland most oyster farmers would only want to purify a small proportion of their production mainly for a local market and still export the majority of their product in bulk for relaying and purification on the continent.

The basic economic question is what is the difference in price for purified and unpurified product. In reality, for most species it is very little, for people with shellfish from B classification areas are competing directly with people with shellfish from A classification areas. If anything putting shellfish through a depuration plant would add an extra cost of approximately £50.00/tonne. Therefore the real benefits of having a depuration and dispatch centre is to be able to by-pass the traditional middle men or develop new markets directly with retail outlets. In this way you obtain a higher price for your product but in return you will most likely have to improve packaging, arrange distribution, guarantee regular supplies and give extended credit along with taking out product insurance etc. In other words, you are taking on a whole new aspect of the shellfish business and as with any other business there will people able to do a better job than others and subsequently make more profit and run their depuration centres more economically.

Market Access for Purified Product.

When your shellfish are purified and dispatched from a properly registered facility it now means you can sell directly to anyone in the E.U., from your neighbour to any of the multiple supermarket stores. However, you must remember the shellfish trade, especially on the continent, is an old established trade that has seen many people come and nearly as many go. Competition in the traditional markets is intense and it is very hard for a new comer to gain proper access.

There are two areas of hope for a new shellfish supplier: One is to find new markets, the second is having a product that is in short supply.

Shellfish is becoming more and more popular so the market demand is generally

good. The main growth area in many countries are the supermarkets. Tying into even a small continental supermarket chain that may be just expanding its shellfish sales will most likely be a large enough market (along with a certain amount going to traditional buyers) for any new venture likely to be established.

Depending on whether you have to go through a general distribution for the supermarkets will affect the price you receive. As a very general rule you can divide the final retail price roughly into three, a third for the supermarket, a third for the distributor and a third for the supplier. If the distributor can be by-passed then a larger portion of the retail price can be obtained. But for this extra you may have to provide daily deliveries to supermarkets over a wide area which can be extremely difficult and costly.

If you have a product that is in short supply, due to a production shortfall by the traditional suppliers. This usually means prices are increased. By carefully bargaining on price you should be able to gain access to a market. If your quality of product and service is good then even if in the next year the regular supplies come back on stream you should be able to retain a share of the market, though it will most likely be at a reduced level.

Being able to offer a range of products also makes establishing a bridgehead in the market place easier.

As prices and demand fluctuate from year to year and indeed month to month it is important that current market prices are obtained when this topic is discussed. B.I.M.'s Paris office can be contacted by businesses based in Ireland for current prices and trends.

INTRODUCTION TO PRACTICALS

By necessity to enable the participants to have as much hands-on experience as possible the number of people carrying out any one session needs to be manageable and reflect the amount of equipment available.

For the laboratory work on this course only half of the group could work in the laboratory while the other half carried out one of the other practicals. This meant that though one practical is listed in the agenda, two or in some cases, three separate practicals were carried out at one time and repeated as necessary to ensure all participants had covered all areas of the course.

In addition, though not scheduled on the agenda, the participants were required to go back to the lab. in groups over the rest of the course (during other practical sessions) to see the progression of their tests and experiments and also to discuss results and ask questions.

The toxic plankton practical took approximately one hour and was carried out in pairs, again this meant people leaving existing practicals which the relevant sections had to be repeated. Various data and results were collected and noted during the practicals. These were discussed and reviewed at appropriate times in the class room.

The equipment used in the practicals was provided by Fastnet Mussels Ltd., B.I.M. and Seafish Industry Authority. Shellfish were sourced from local growers and suppliers.

PRACTICAL: SAMPLING AND BACTERIA TESTING

Part 1 Aseptic Technique

(a) Demonstration of Aseptic Technique and Looping-out Method

(b) "Hands-on" Looping-out

Sample dilutions (1:10), metal loops, bunsens and agar plates are provided.

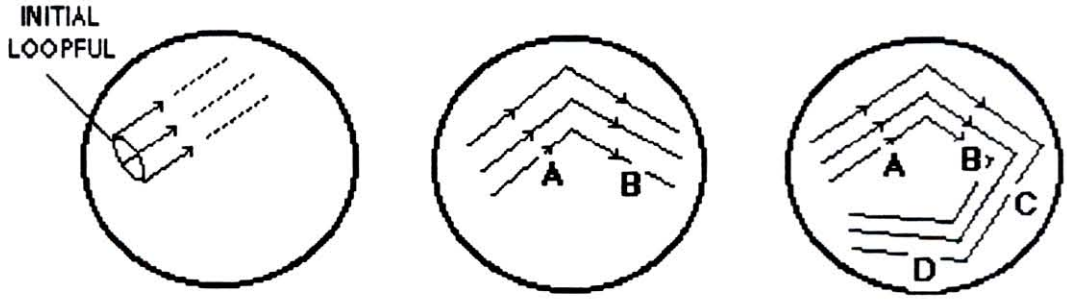
(i) Place one loopful of sample on the agar near the rim of the plate.

(ii) Spread the loopful from area A over area B with parallel streaks, taking care not to let the streaks overlap.

(iii) Flame the loop and repeat with area C and so on, (see diagram). Each looping-out dilutes the inoculum.

(iv) Invert the plates.

(v) Incubate the plates @ 30°C.



Part 2 Environmental Testing

(a) Contact Plates

Each person is provided with a contact plate.

(i) Choose a smooth surface anywhere in the laboratory.

(ii) Label the outside of the lid with your initials and the chosen surface.

(iii) As quickly and carefully as possible, remove the lid (ensuring that you do not touch the agar surface or the inside of the lid with your hands).

(iv) Press the agar surface lightly against your chosen surface and carefully replace the lid.

(v) Invert plates

(vi) Put plates in incubator @ 30°C.

(b) Surface Swabs

Each person is provided with a sterile swab, an agar plate and a universal bottle of buffered peptone water (BPW).

(i) Label the base of the agar plate with your name, the date and the surface swabbed.

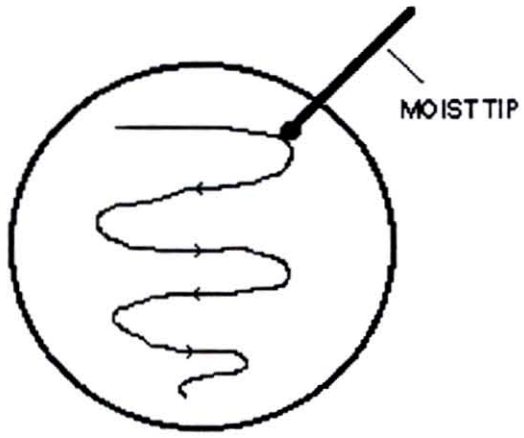
(ii) Remove swab from tube carefully (ensuring that it doesn't touch anything).

(iii) Open the universal bottle by removing lid and flame the top.

(iv) Place the swab into the Buffered Peptone Water and then remove moist tip.

(v) "Roll" tip of swab around a square inch area of the surface chosen.

(vi) Carefully streak this swab tip onto the surface of the agar as in diagram.



(c) Settle Plates

Bacteria and fungi may be suspended in large particles in the are and can settle rapidly and contaminate surfaces. Settle plates are used for assessing potential surface contamination.

Each person is provided with an agar plate.

(i) Label the base of the agar plate with your name, the date, "Settle Plates", and location of plate.

(ii) Remove the lid from the agar base and leave exposed in chosen area.

(iii) After 1 hour replace lid on plate.

(iv) Invert plate.

(v) Incubate plate @ 30°C.

Part 3 Personal Hygiene

A high proportion of the population carry a microorganism on their body surface which, when present in foods in high numbers, can cause food poisoning. In this test you will attempt to isolate their microorganism, Staphylococcus aurous from your hands/nose/face/gums. You are provided with 2 sterile swabs, 2 BP agar plates and 2 universal bottles of Buffered Peptone Water.

Method

(i) Label the base of the BP plate with your name, the date and area of body swabbed.

(ii) Remove swab from tube carefully.

(iii) Remove lid from universal bottle and flame top.

(iv) Place the swab into the BPW in the universal bottle.

(v) "Roll" moist tip of swab around a small area of the body surface chosen.

(vi) Carefully streak this swab tip onto the surface of the BP plate as in Fig 2.

(vii) Invert plate.

(viii) Incubate plates @ 37°C

Demonstrations

1 Pathogen cultivation: salmonella

2 Cultivation of bacteria anaerobically

3 Microscopic analysis of pathogens

PRACTICAL: BACTERIA TESTING

1 Water and Mollusc Sampling Methods

A Water Sampling

a Clean inside and outside of tap nozzle thoroughly and then turn on tap for 2-3 minutes.

b Turn off tap and leave for 1 minute. Sterilise by sponging with methylated spirits or alcohol and flaming.

c Turn on tap for 1 minute and cool tap. Turn off tap.

d Carefully remove the stopper from the bottle, holding the stopper by the outside rim.

e Hold the bottle under the tap and holding the cover in your hand turn on the tap and fill the bottle.

f Turn off tap and replace stopper.

B Mollusc Sampling

a Using sterile gloves or bag, pick molluscs and place in large sterile, plastic bag.

2 Sampling Preparation

Molluscs

(i) Remove mud etc. from shell prior to placing in bag

(ii) Choose animals with intact shells

(iii) Sample size should be about 1kg.

3 Most Probable Number (MPN) Test

a Enumeration of faecal coliforms and E.coli in Shellfish using the 3 tube Most Probable Number (MPN) Technique.

Apparatus and Material

Waterbaths (37°C and 44°C)
Stomacher
Balance
Sterile Bags
Sterile McConkey Broth - 3 x 10mls Double Strength
3 x 10mls Single Strength
3 x 10mls Single Strength

Each bottle contains an inverted durham tube to indicate gas production.

Procedure

- Take 10 mussels, scrub and clean the outside and wash in sterile water.
- Hold with the concave shell down and open with a sterile knife.
- Add the intravalvular fluid to a sterile stomacher bag.
- Using a sterile knife, remove all the mussel flesh and add this to the stomacher bag.
- Aseptically weigh out 10g of the sample into a sterile stomacher bag and add 90mls of sterile diluent. Homogenise for 2 minutes, using a sterile pipette, add 10ml quantities of the homogenate to each of 3 tubes of Double Strength McConkey, 1ml quantities to each of 3 tubes of Single Strength McConkey, 0.1ml quantities to each of 3 tubes of Single Strength McConkey.
- The tubes are incubated in a waterbath for 48 hours at 37°C when testing for coliforms.
- After 48 hours, the bottles are examined for acid and gas production.
- A colour change from purple to yellow is indicative of acid production.
- Gas production can be observed in the durham tubes.
- Acid and gas production is indicative of the presence of coliforms and possible E.coli. Consult MPN tables to give most probable number per gram.

b Enumeration of coliforms and faecal coliforms in water using the 5 tube MPN Technique.

Apparatus and Materials

Waterbaths (37°C and 44°C)
McConkey Broth bottles (containing durham tubes)
Brilliant Green Bile Broth (BGB - containing durham tubes)

- Using sterile pipettes, add 50mls of water to 50mls of broth, 10mls of water to each of five tubes containing 10mls of double strength McConkey Broth.
- 1ml of water to each of five tubes containing 5ml of single strength McConkey Broth.

- The tubes are incubated in a waterbath for 48 hours at 37°C, when testing for coliforms.

- After 48 hours, the bottles are examined for acid and gas production.

4 Membrane Filtration Demonstration

a Pass a 100ml volume of water sample through 47mm sterile membrane filter using a vacuum pump.

Note if the supply is known to contain more than 100 coliform bacilli/100ml. use 10ml of water diluted with 90ml (0.1 peptone).

b Place sterile Whatman No. 17 absorbent pads in sterile petri dishes and pipette 2.5 - 3ml of enriched LST or McConkey Broth over the surface.

c Using sterile forceps remove the filtered membrane from the filtering apparatus.

d Place filtered membrane on Whatman pad containing broth and incubate at 37°C.

Confirmatory tests for Faecal Coliforms

Bottles which have shown acid and gas production for the coliform MPN test are examined.

- 0.1ml quantities are removed and added to bottles of Brilliant Green Bile Broth and Tryptone water. The bottles are incubated in a water bath at 44°C and examined after 24 hours.

- Gas production can be observed in the durham tubes in the Brilliant Green Bile Broth bottles.

- A few drops of Kovac reagent is added to the Tryptone water bottles.

- A pink/red colour after 5 minutes indicates indole production.

- The presence of gas in the BBG bottles and a positive indole test indicates the presence of Faecal Coliforms.

- Consult MPN tables to give most probable number per gram.

PRACTICAL: TOXIC PLANKTON MONITORING

Equipment:

Inverted microscope
Plankton settling chamber
Cell count slide
Sample bottles (100 ml)
Plankton samples (preserved and fresh)

Procedure

In pairs participants are shown plankton samples under the inverted microscope (for approximately one hour). The difference between an inverted and a standard binocular microscope is explained. The use of the microscope is demonstrated. The benefits and use of plankton settling chambers and cell count slides are to be explained and demonstrated especially in relation to carrying out cell counts of toxic algae.

From the samples the following are to be seen and identified:

- Dinophysis sp.
- Gyrodinium aureolum
- Alexandrium sp.
- Nitzchia sp.

Other types of general plankton including zooplankton are to be briefly shown.

The procedure for taking and preserving water samples is to be explained.

PRACTICAL: CARE AND USE OF MONITORING EQUIPMENT

Temperature

Equipment:

Mercury thermometer (0 - 60°C)
Alcohol thermometer (0 - 60°C) with protective sleeve
Digital thermometer with steel probe
Temperature salinity bridge
Maximum/minimum thermometer
Water at three different temperatures (approx. 2°C, 20°C and 40°C)

Procedure

The correct use of each type of thermometer and instrument is demonstrated.

Use the various thermometers and instruments to measure the temperature of the water in the three containers. Record the temperatures obtained. Note:

- The fact that different instruments may give slightly different readings for water at one temperature.
- The time taken for various instruments to reach equilibrium.
- That mercury thermometers should not be used in tanks or where risk of breaking may contaminate shellfish.

Salinity

Equipment:

Hydrometer, clear graduated cylinder, thermometer and specific gravity conversion chart.
Salinity meters (various types).
Refractometer, dropper, and lens tissues.
Four containers, with water at three different salinities (0 ppt, 20 ppt and 33 ppt) at ambient temperature with the fourth container with one of the above salinities at a temperature ten degrees above ambient. Note, the participants should not be told the salinities of the water.

Procedure

The use of each piece of equipment is demonstrated along with how to convert specific gravity to salinity.

Use the various pieces of equipment to take readings from each of the containers, record the readings. Note:

- How some of the salinity meters will automatically compensate for temperature variation.

- If different salinities were obtained for the containers at the same salinity check that the temperature was corrected for.

Oxygen

Equipment:

Oxygen meters (various types).
Membrane kits.
Data logger.

Procedure

The correct use of each piece of equipment is demonstrated along with how to replace the membranes and clean the electrodes on the oxygen probes. The procedure for calibrating each probe is demonstrated.

Using the various oxygen meters record oxygen levels in a water sample. Note:

- Leave the probe stationary and note how the oxygen level starts to decrease. Move the probe gently from side to side or up and down to obtain a steady reading. It is important for most hand held probes to have a good flow of water over the membrane to give an accurate reading.
- The probes of the oxygen data logger when positioned correctly do not need to be moved to obtain a steady reading.
- Some of the meters will automatically compensate for the temperature where others require the use of a conversion table.

Change the membrane and clean the electrode of an oxygen meter. Note:

- Make sure no air bubbles are trapped under the membrane. This can be seen when the probe is held up side down.

Go through the calibration procedures for each type of oxygen meter. Note:

- Certain meters are self calibrating and will also carry out diagnostic checks to indicate whether or not the electrode needs cleaning etc.

Nitrates, ammonia and pH

Equipment:

Various aquaria test kits.
Colorimeter with test kits.
pH meter.
Water samples with varying levels of ammonia etc. loading, i.e. fresh sea water, water from a closed, recirculating purification tank at the end of a run and water from a fish tank.

Procedure

The use of the test kits, colorimeter and pH meter are demonstrated.

Test the water samples for ammonia, nitrates and pH using the various kits and instruments. Note:

- Water samples from fresh sea water and the shellfish tank show very little difference whereas that from the fish tank has relatively high levels.

Turbidity

Equipment:

Secci disc.

Procedure

The use of the secci disc is demonstrated only. During other practicals the use of visual marks on tanks is emphasised.

Equipment care

The correct cleaning, storing and maintenance procedures for each piece of equipment is demonstrated and after use the participant should clean the equipment accordingly.

PRACTICAL: FACTORS AFFECTING PURIFICATION

Species variation

Equipment:

Rope mussels, both unwashed and washed and graded.
Bottom mussels.
Gigas oysters.
Flat oysters.
Native clams.

Procedure

Inspect all the types of shellfish. Note the difference between washed and unwashed product. Check each batch for:

- Broken shell and damaged product.
- General condition of shellfish.
- Fouling on product.
- Presence of crabs and starfish etc.
- Amount of mud on and in product. There is usually more mud on a dredged or dug product.

Filtering activity

Equipment:

Five small aquariums with aerators and air stones etc.
500 g. of washed rope mussels, bottom mussels, gigas oysters, flat oysters and clams.
Stock solution of neutral red dye.
Colorimeter.
Thermometer.

Procedure

Place 500 g of shellfish into each aquarium add four litres of sea water to each tank (sufficient to adequately cover shellfish in tank). Add the same amount of stock neutral red dye to each tank so as to colour the water a strong red colour. Record the time and check degree of transmission from each tank on the colorimeter along with the temperature. Record how long it takes each type of shellfish to open and appear to actively start filtering. Over the next hour periodically check the degree of clearing from each tank by visual inspection and also take readings on the colorimeter. Note:

- Rope mussels generally clear first, followed by the bottom mussels, gigas oysters, flat oysters and then clams.
- Opening and start of filtering activity is approximately the same for

both types of mussels and the rest usually follow as in the above order.

- If the temperature is below 12°C then the clams though open and having syphons extended may not be actively filtering.

After one hour remove shellfish and open several from each tank to see the uptake of dye by the gills. Generally the more active the filtering the darker red the gills will be.

Temperature

Equipment:

Three aerated aquaria with four litres of water maintained at the following temperatures 2°C, 12°C and 20°C.
Three 500 g batches of rope mussels.
Stock solution of neutral red dye.
Colorimeter.
Thermometer.

Procedure

Place the shellfish samples in each of the aquaria to which equal amounts of stock dye solution has been added. Take temperature and transmission readings. Record readings and time. Every five minutes (for 30 minutes) check the colour of the water. Note:

- The aquaria at the highest temperature clears rapidly followed by the tank at 12°C. The tank at 2°C hardly clears at all.

Open several mussels from each tank, check the degree of dye uptake.

Meat yield

Equipment:

1 Kg of rope mussels, bottom mussels, gigas oysters, flat oysters and clams.
Top loading balance.
Absorbent paper towel.
Knives.

Procedure

Remove excess water from the shellfish by rolling them on the paper towel. Weigh out accurately as near to 1 Kg of each of the shellfish as possible. (The shellfish should have been in seawater for at least half an hour immediately before this experiment.) Open all the shellfish and remove the meat from the shell. Decant and discard any fluid. Towel dry shells and meats to remove excess water. Weigh the shells and meats. Divide the meat weight by the total animal weight to obtain

meat yield. Note:

- Variations in meat yields.
- Amount of shell, which is not respiring tissue.

Relate the meat yields to the results of the first dye experiment. It is important to realise that 1 Kg of rope mussels may contain twice as much meat and hence respiring tissue as bottom mussels therefore the stocking densities of tanks should reflect this.

Inducing spawning

Equipment:

Two aerated aquaria, one at 2°C the other at 20°C.
500 g of rope mussels (with meat yields over 30%).

Procedure

Place the mussels in the cold water for 10 minutes, then place them in the warm water for 20 minutes. Repeat as necessary until spawning occurs. Leave animals in warm water with spawn for several hours. Note:

- How cloudy the water with spawn becomes.
- The thickness and colour of the foam.
- The smell as the day progresses.

PRACTICAL: PURIFICATION SYSTEMS 1

Single layer system and general depuration centre layout

The depuration centre, in which this course is based, is designed on the single layer system. Go through all aspects of the centres design pointing out good and bad features.

- Water intake: Submersible pump situated subtidally. Well protected from storms and can pump at all times.

- Pump house: Two large pumps to recirculate water from the sump tank up to the water reservoir.

- Water reservoir: Large capacity, uncovered water storage tank. Can gravity feed purification tanks. Measure salinity and oxygen levels in the reservoir. In theory, because it and the purification tanks are uncovered, during very heavy rainfall the salinity may be lowered.

- U. V. system: The six U. V. systems are vertically mounted in such a way that they cannot air lock. You should be able to isolate all units for easy maintenance. There should be sufficient room to be able to remove tubes and sleeves with ease. Note clocks with reset buttons. Housing made from U. V. stabilised PVC.

- Washing and grading equipment for shellfish entering tanks.

- Purification tanks: Load a tank with mussels, stagger baskets, place baffle to direct water flow. Allow tank to fill. Check salinity. Check oxygen level at water entry point, in the middle of the mussels and at the end of the tank. Check oxygen level after weir. Note the long tanks give oxygenation problems during the summer at this time it is necessary to supply additional aeration.

- Sump tank: Water can be recirculated from sump tank back to the water reservoir.

- Second set of washing and grading machinery for when the shellfish leave the tanks.

- Drains: All drains are large and easily accessible in order to facilitate cleaning of blockages.

- Water discharge/outflow: This is situated well away from intake point and discharges into deep water with a good flushing capacity.

- Buildings: Handling and packaging of shellfish take place indoors. Note, all the outdoor purification tanks will be covered in the near future to comply with the E.C. directive.

PRACTICAL: PURIFICATION SYSTEMS 2

Multilayer system

Go through all aspects of the design and operation of the tank system. This system was set up with an ozone unit, go through all aspects of design and operation of the ozone unit.

Lay out the baskets of mussels for loading on the floor, compare the floor space required to the floor space the tank takes up.

Spiking procedure for checking out the tank is carried out. Either freeze dried bacteria or fresh culture can be used. The various ways of testing a tank are demonstrated. For this run the tank is loaded and filled. Check salinity and oxygen levels throughout the tank, take a mussel sample for bacteria analysis. The circulation pumps are switched on without the sterilising unit. The bacteria culture is added to the tank and circulation is carried out for one hour. A second mussel sample is taken. The sterilisation unit is switched on the run time is recorded.

Record oxygen levels at various points in the system. Switch off circulation pump and note the rapid decrease in oxygen levels indicating the heavy loading of the system. Switch pumps back on and continue with purification run.

Take mussel samples for bacteria analysis after 12 , 24 and 36 hours. Analyse results on last day.

PRACTICAL: PURIFICATION SYSTEMS 3

Stacking system

Go through all aspects of design and operation of the stacking system. Load each layer with shellfish and fill the system. For the purpose of demonstration use different species in each layer. Switch on the system. Check the salinity and oxygen levels throughout the system. Record when each species appears to be open and filtering.

At the end of the evening practical session spike a single layer of shellfish with a bacteria culture. Switch off the sterilising unit and leave run for one hour. Take a shellfish sample for bacterial analysis. Leave the system running overnight with the sterilisation unit switched off. Take sample of shellfish for bacterial analysis after 12 hours. It will be noticed that up to a ten fold reduction in bacterial levels can be obtained without the use of the water sterilising unit.

Ultra violet systems

Several types of low intensity U. V. are stripped down and the correct cleaning procedure is demonstrated. The units are then reassembled.

Water coolers

Various water coolers; in line cooler; direct and indirect water cooler; are demonstrated. The advantages and disadvantages of each system is described.

Flow meters

The use and installation of two types of flow meters are demonstrated.

Pumps

Various types of pumps are demonstrated; submersible; pump with filter; pump without filter. Advantages and disadvantages of each type is discussed.

Materials

Various materials that are used in the construction of purification systems are discussed i.e. the use of GRP of 316 stainless, importance of uPVC piping etc. Cleaning and sterilising of pipe work is also discussed.

PRACTICAL: PURIFICATION SYSTEMS 4

Upwelling and downwelling systems

Go through operation and design of both systems. Load each system with mussels. Place electrodes from the data logger at various positions in the tank. Note the level of the mussels on the side of the tank. Switch on pumps etc. Note data logger recording temperature and oxygen levels. Record when mussels are open and filtering. Note level to which the mussels have increased.

Discuss options of tidal training as part of a purification run and how it is achieved.

Leave tanks running. At a later stage unload tanks. Note how well byssed up the mussels are.

Go through tank cleaning procedures.

Demonstrate downloading of data logger recordings onto the computer and show oxygen profiles obtained.

PRACTICAL: PURIFICATION SYSTEMS 5

Aeration system

Go through design features and operation of the aeration system. Load tank with mussels. Place in data logger probes. Fill tank with water and switch on the venturi pump. Note bubble stream.

Add sufficient neutral red dye to colour the water in the tank. Notice the way in which the dye was circulated. Milk added to the water can also be used to see the circulation pattern of a tank.

Notice the difference in the foam build up once the mussels are actively filtering. Note how the foam is constantly removed via the foam drain. Note oxygen readings on the various probes.

Drain tank and open several of the mussels to see that dye uptake was uniform throughout the tank.

APPENDICES

Cartoon

Course programs

Conversion graph for specific gravity/salinity

Laboratory set up costs

MPN tables

EC Directive 91/492

EC Directive 91/493

EC Directive 94/356

Sample dispatch/purification centre log book

Draft record sheets

Sample HACCP audit (Seafish)

Seafish report list.

DEPURATION CENTRE MANAGEMENT

GERAHIES

BANTRY

CO. CORK.

4th - 8th October 1993.

AGENDA

Monday
Oct. 4th

8.30	Meet at Reception in Bantry Bay Hotel
9.00 - 9.15	Registration

Lectures

9.15 - 9.30	Introduction to the Course *Terence O'Carroll*
9.30 - 10.00	General introduction to shellfish purification *Terence O'Carroll*
10.00 - 10.30	Introduction to bacteria and viruses *Mary Seaver*
10.30 - 10.45	**Coffee**
10.45 - 11.30	Bacteria sampling procedures and testing *Mary Seaver*
11.30 - 1.00	Practical - sampling and bacteria testing
1.00 - 2.00	**LUNCH**

Lecture

2.00 - 3.00	Setting up a laboratory *Mary Seaver*
3.00 - 5.00	Practical - Bacteria testing.

Tuesday		
Oct. 5th	9.00 - 9.45	Monitoring parameters and equipment *Terence O'Carroll/Mark Boulter*
	9.45 - 10.00	Toxic plankton monitoring. *Cillian Roden*
	10.00 - 10.30	Documentation requirements and record keeping *Michael O'Driscoll and Terence O'Carroll*
	10.30 - 10.45	**Coffee**
	10.45 - 1.00	Practical - Care and use of monitoring equipment
	1.00 - 2.00	**LUNCH**
	2.00 - 3.00	Traditional single layer and stacking system *Mary Hannan*
	3.00 - 5.00	Practical - Factors affecting purification (temperature. salinity, turbidity)

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Wednesday		
October. 6th	9.00 - 10.00	Sterilization medium - U.V./Ozone *Peter McKeown*
	10.00 - 10.30	Multilayer and downwelling system *Mark Boulter.*
	10.30 - 10.45	**Coffee**
	10.45 - 1.00	Practical - purification systems **(I)** *(This use of five practicals will cover the use and monitoring of the various purification systems)*
	1.00 - 2.00	**LUNCH**
	2.00 - 3.00	Upwelling and aeration systems *Terence O'Carroll*
	3.00 - 5.00	Practical - purification systems **(II)**

****

Thursday		
Oct. 7th	9.00 - 9.30	Siting, location and planning requirements for establishing a facility. *Terence O'Carroll*
	9.30 - 10.15	Building design, layout, general equipment and commissioning systems. Terence O'Carroll
	10.15 - 10.30	**Coffee**
	10.30 - 1.00	Practical - purification systems **(III)**
	1.00 - 2.00	**LUNCH**

	2.00 - 3.00	Operating practices and quality control H.A.C.C.P. *Mark Boulter.*
	3.00 - 5.00	Practical - purification systems **(IV)**
Friday		
Oct. 8th	9.00 - 9.45	Economics of operating a purification facility *Terence O'Carroll*
	9.45 - 1.00	Practical - purification systems **(V)**
	1.00 - 2.00	**LUNCH**
	2.00 - 3.00	Review of lectures
	3.00 - 3.45	Review of practicals including data
	3.45 - 4.00	**Coffee**
	4.00 - 4.30	Course analysis
	4.30 - 5.00	Closing address

Coffee breaks will be held during the practical sessions.

October 1993.

DEPURATION CENTRE MANAGEMENT

GERAHIES

BANTRY

CO. CORK.

9th - 13th May 1994.

AGENDA

Monday

May 9th

8.30		Meet at Reception in Bantry Bay Hotel
9.00 - 9.15		Registration
		Lectures
9.15 - 9.30		Introduction to the Course *Terence O'Carroll*
9.30 - 10.00		General introduction to shellfish purification *Terence O'Carroll*
10.00 - 10.30		Introduction to bacteria and viruses *Mary Seaver*
10.30 - 10.45		**Coffee**
10.45 - 11.15		Toxic plankton monitoring *Cillian Roden*
11.15 - 1.00	*	**Practical** - Sampling and bacteria testing - Care and use of monitoring equipment - Toxic plankton monitoring
1.00 - 2.00		**LUNCH**
		Lectures
2.00 - 2.30		Bacteria sampling procedures and testing *Mary Seaver*
2.30 - 4.45	*	**Practical** - Sampling and bacteria testing - Care and use of monitoring equipment - Toxic plankton monitoring
4.45 - 5.00		Day's summary - Questions

*** Practicals will be held in concurrent sessions.**

Tuesday

10th May

9.00 - 9.45		Monitoring parameters and equipment *Terence O'Carroll/Mark Boulter*
9.45 - 10.45		Operating practices and quality control H.A.C.C.P. *Mark Boulter*
10.45 - 11.00		***Coffee***
11.00 - 1.00	* **Practical**	- Factors affecting purification (temperature, salinity, turbidity). - Bacteria testing.
1.00 - 2.00		**LUNCH**
2.00 - 3.00		Traditional single layer and stacking system *Terence O'Carroll*
3.00 - 4.45	* **Practical**	- Factors affecting purification temperature, salinity, turbidity) - Bacteria testing
4.45 - 5.00		Day's summary - Questions

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Wednesday

11th May

9.00 - 10.00		Sterilization medium - U.V./Ozone/Ionization *Terence O'Carroll/Mark Boulter.*
10.00 - 10.30		Multilayer and downwelling system *Mark Boulter.*
10.30 - 10.45		**Coffee**
10.45 - 1.00		Practical - purification systems (I) *(This series of five practicals will cover the use and monitoring of the various purification systems)*
1.00 - 2.00		**LUNCH**
2.00 - 2.30		Upwelling systems *Mike Barnett*
2.30 - 3.00		Aeration Systems *Daniel Masson*
3.00 - 4.45	**Practical**	- Purification systems (II)
4.45 - 5.00		Day's summary - Questions.

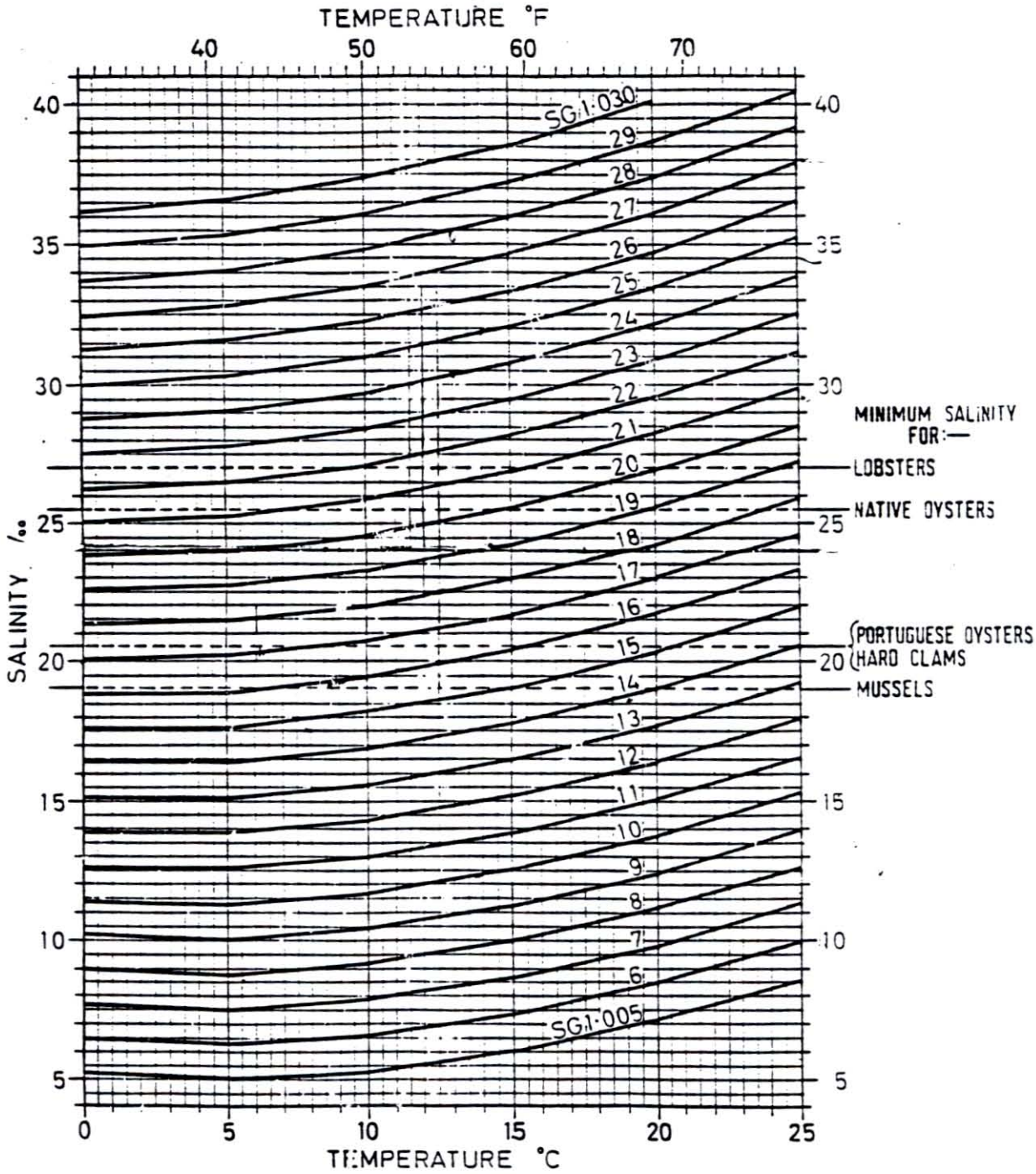
*** Concurrent practical sessions**

Thursday

12th May	9.00 - 9.30	Siting, location and planning requirements for establishing a facility. *Terence O'Carroll*
	9.30 - 10.30	Setting up a laboratory. *Mary Seaver*
	10.30 - 10.45	**Coffee**
	10.45 - 1.00	**Practical** - purification systems **(III)**
	1.00 - 2.00	**LUNCH**
	2.00 - 2.45	Building design, layout, general equipment and commissioning systems. *Terence O'Carroll*
	2.45 - 3.15	Documentation and record keeping *Michael O'Driscoll*
	3.15 - 4.45	**Practical** - Purification systems **(IV)**
	4.45 - 5.00	Day's summary - Questions

Friday

13th May	9.00 - 9.45	Economics of operating a purification facility *Terence O'Carroll*
	9.45 - 10.15	Market Access for purified product *Marc Gerard*
	10.15 - 10.30	**Coffee**
	10.30 - 1.00	**Practical** - Purification systems **(V)**
	1.00 - 2.00	**LUNCH**
	2.00 - 3.30	Overviewing Design and requirements of participants planned and actual facilities
	3.30 - 4.30	Review of lectures and practicals
	4.30 - 5.00	**Course analysis and closing address**



Graph for conversion. Specific gravity ~ salinity.

MICROBIOLOGICAL LABORATORY SET-UP

A. EQUIPMENT REQUIREMENTS

"ESSENTIAL"		UNIT COST	TOTAL COST
Water Distillation Unit		£350.00	£350.00
Incubator (2)		£500.00	£1,000.00
Waterbaths (2)		£500.00	£1,000.00
Stomacher (Lab-Blender)		£1,400.00	£1,400.00
Hotplate Stirrer		£200.00	£200.00
Autoclave (Portable)		£600.00	£600.00
Top-pan Balance		£700.00	£700.00
Fridge		£250.00	£250.00
	TOTAL		£5,500.00
"OPTIONAL"			
Steriling Oven			£450.00
Colony Counter			£400.00
Vortex Mixer			£130.00
Laminar Air Flow Cabinet			
Microwave			£120.00
Dispenser			£150.00
Autoclave (Automatic)			£5,000.000
	TOTAL		£6,250.000

B. GLASSWARE REQUIREMENTS

Universal Bottles (100)		£0.900	£90.000
Durham Tubes (100)		£0.070	£7.000
Glass Rods		£0.400	£4.000
Media Storage Bottles (10)		£5.000	£50.000
Membrane Filtration Units		£120.000	£120.000
	TOTAL		£271.000

C. PLASTICS

Specimen Containers : (250 ml)(10)		£2.300	£23.000
(500 ml)(10)		£2.000	£20.000
Distilled Water Container		£14.000	£14.000
Pipette Fillers (3)		£8.000	£24.000
	TOTAL		£81.000

D. CONSUMMABLES

Plastic Pipettes : (10 ml) (1000)		£0.190	£190.000
: (1 ml)(1000)		£0.080	£80.000
MacConkey Broth (500g)		£17.000	£17.000
Tryptone Broth(500g)		£21.000	£21.000
Brilliant Green Bile Broth (500g)		£38.000	£38.000
Plate Count Agar (500g)		£36.000	£36.000
Methylated Spirits (2.5 l)		£7.500	£7.500
Petri Dishes (1000)		£0.040	£40.000
Autoclave Bags (1000)		£0.010	£100.000
Disposable Gloves (100 Pairs)		£0.050	£5.000
Laboratory Sanitiser (1 l)		£5.000	£5.000
Sterile Membrane Filters (100)		£0.300	£30.000
Sterile Membrane Pads (100)		£0.060	£6.000
	TOTAL		£575.500
TOTAL WITHOUT OPTIONAL EQUIPMENT			£6,427.500
TOTAL WITH OPTIONAL EQUIPMENT			£12,677.500

MPN table for 3 × 1, 3 × 0.1, and 3 × 0.01 g (ml)

Number of positive results			MPN	Category when no. of tests					Confidence limits			
				1	2	3	5	10	≥ 95%	≤ 95%	≥ 99%	≤ 99%
0	0	0	< 0.30						0.00	0.94	0.00	1.40
0	0	1	0.30	3	2	2	2	1	0.01	0.95	0.00	1.40
0	1	0	0.30	2	1	1	1	1	0.01	1.00	0.00	1.60
0	1	1	0.61	0	3	3	3	3	0.12	1.70	0.05	2.50
0	2	0	0.62	3	2	2	2	1	0.12	1.70	0.05	2.50
0	3	0	0.94	0	0	0	0	3	0.35	3.50	0.18	4.60
1	0	0	0.36	1	1	1	1	1	0.02	1.70	0.01	2.50
1	0	1	0.72	2	2	1	1	1	0.12	1.70	0.05	2.50
1	0	2	1.1	0	0	0	3	3	0.4	3.5	0.2	4.6
1	1	0	0.74	1	1	1	1	1	0.13	2.00	0.06	2.1
1	1	1	1.1	3	3	2	2	2	0.4	3.5	0.2	4.6
1	2	0	1.1	2	2	1	1	1	0.4	3.5	0.2	4.6
1	2	1	1.5	3	3	3	3	2	0.5	3.8	0.2	5.2
1	3	0	1.6	3	3	3	3	2	0.5	3.8	0.2	5.2
2	0	0	0.92	1	1	1	1	1	0.15	3.50	0.07	4.60
2	0	1	1.4	2	1	1	1	1	0.4	3.5	0.2	4.6
2	0	2	2.0	0	3	3	3	3	0.5	3.8	0.2	5.2
2	1	0	1.5	1	1	1	1	1	0.4	3.8	0.2	5.2
2	1	1	2.0	2	2	1	1	1	0.5	3.8	0.2	5.2
2	1	2	2.7	0	3	3	3	3	0.9	9.4	0.5	14.2
2	2	0	2.1	1	1	1	1	1	0.5	4.0	0.2	5.6
2	2	1	2.8	3	2	2	2	1	0.9	9.4	0.5	14.2
2	2	2	3.5	0	0	0	0	3	0.9	9.4	0.5	14.2
2	3	0	2.9	3	2	2	2	1	0.9	9.4	0.5	14.2
2	3	1	3.6	0	3	3	3	3	0.9	9.4	0.5	14.2
3	0	0	2.3	1	1	1	1	1	0.5	9.4	0.3	14.2
3	0	1	3.8	1	1	1	1	1	0.9	10.4	0.5	15.7
3	0	2	6.4	3	3	2	2	2	1.6	18.1	1.0	25.0
3	1	0	4.3	1	1	1	1	1	0.9	18.1	0.5	25.0
3	1	1	7.5	1	1	1	1	1	1.7	19.9	1.1	27.0
3	1	2	12	3	2	2	2	1	3	36	2	44
3	1	3	16	0	0	0	3	3	3	38	2	52
3	2	0	9.3	1	1	1	1	1	1.8	36.0	1.2	43.
3	2	1	15	1	1	1	1	1	3	38	2	52
3	2	2	21	2	1	1	1	1	3	40	2	56
3	2	3	29	3	3	3	2	2	9	99	5	152
3	3	0	24	1	1	1	1	1	4	99	3	152
3	3	1	46	1	1	1	1	1	9	198	5	283
3	3	2	110	1	1	1	1	1	20	400	10	570
3	3	3	>110									

Table 9.2 MPN/100 ml, using one tube of 50 ml, five tubes of 10 ml and five tubes of 1 ml

50-ml tubes positive	*10-ml tubes positive*	*1-ml tubes positive*	*MPN/100 ml*
0	0	0	0
0	0	1	1
0	0	2	2
0	1	0	1
0	1	1	2
0	1	2	3
0	2	0	2
0	2	1	3
0	2	2	4
0	3	0	3
0	3	1	5
0	4	0	5
1	0	0	1
1	0	1	3
1	0	2	4
1	0	3	6
1	1	0	3
1	1	1	5
1	1	2	7
1	1	3	9
1	2	0	5
1	2	1	7
1	2	2	10
1	2	3	12
1	3	0	8
1	3	1	11
1	3	2	14
1	3	3	18
1	3	4	20
1	4	0	13
1	4	1	17
1	4	2	20
1	4	3	30
1	4	4	35
1	4	5	40
1	5	0	25
1	5	1	35
1	5	2	50
1	5	3	90
1	5	4	160
1	5	5	180+

Determination of MPN index when the examination is carried out on several samples per batch[1]

Number of positive tubes at the three successive dilutions selected			MPN index	Category[2] when the number of samples (per batch) examined together is				Confidence limits[3]			
								95 %		99 %	
1st	2nd	3rd		2	3	5	10				
0	0	0	< 0,30								
0	0	1	0,30	2	2	2	1	0,01	0,95	0,00	1,40
0	1	0	0,30	1	1	1	1	0,01	1,00	0,00	1,60
0	1	1	0,6	3	3	3	3	0,12	1,70	0,05	2,50
0	2	0	0,6	2	2	2	1	0,12	1,70	0,05	2,50
0	3	0	0,9	0	0	0	3	0,35	3,50	0,18	4,60
1	0	0	0,4	1	1	1	1	0,02	1,70	0,01	2,50
1	0	1	0,7	2	1	1	1	0,12	1,70	0,05	2,50
1	0	2	1,1	0	0	3	3	0,35	3,50	0,18	4,60
1	1	0	0,7	1	1	1	1	0,13	2,00	0,06	2,70
1	1	1	1,1	3	2	2	2	0,35	3,50	0,18	4,60
1	2	0	1,1	2	1	1	1	0,36	3,50	0,19	4,60
1	2	1	1,5	3	3	3	2	0,45	3,80	0,24	5,20
1	3	0	1,6	3	3	3	2	0,45	3,80	0,24	5,20
2	0	0	0,9	1	1	1	1	0,15	3,50	0,07	4,60
2	0	1	1,4	1	1	1	1	0,36	3,50	0,19	4,60
2	0	2	2,0	3	3	3	3	0,45	3,80	0,24	5,20
2	1	0	1,5	1	1	1	1	0,37	3,80	0,20	5,20
2	1	1	2,0	2	1	1	1	0,45	3,80	0,24	5,20
2	1	2	2,7	3	3	3	3	0,87	9,40	0,51	14,20
2	2	0	2,1	1	1	1	1	0,45	4,00	0,24	5,60
2	2	1	2,8	2	2	2	1	0,87	9,40	0,51	14,20
2	2	2	3,5	0	0	0	3	0,87	9,40	0,51	14,20
2	3	0	2,9	2	2	2	1	0,87	9,40	0,51	14,20
2	3	1	3,6	3	3	3	3	0,87	9,40	0,51	14,20
3	0	0	2,3	1	1	1	1	0,46	9,40	0,25	14,20
3	0	1	3,8	1	1	1	1	0,88	10,40	0,52	15,70
3	0	2	6	3	2	2	2	1,60	18,10	1,00	25,00
3	0	3	10	0	0	0	3	3,50	36,00	2,00	43,00
3	1	0	4	1	1	1	1	0,91	18,10	0,53	25,00
3	1	1	7	1	1	1	1	1,70	18,90	1,10	27,00
3	1	2	12	2	2	2	1	3,50	36,00	2,10	44,00
3	1	3	16	0	0	3	3	3,50	38,00	2,50	52,00
3	2	0	9	1	1	1	1	1,80	36,00	1,20	43,00
3	2	1	15	1	1	1	1	3,50	38,00	2,20	52,00
3	2	2	21	1	1	1	1	3,50	40,00	2,50	58,00
3	2	3	29	3	3	2	2	9,00	99,00	4,60	152,00
3	3	0	20	1	1	1	1	3,60	99,00	2,60	152,00
3	3	1	50	1	1	1	1	9,10	198,00	4,70	280,00
3	3	2	110	1	1	1	1	18,20	405,00	11,40	570,00
3	3	3	> 110								

COMMISSION

COMMISSION DECISION

of 20 May 1994

laying down detailed rules for the application of Council Directive 91/493/EEC, as regards own health checks on fishery products

(Text with EEA relevance)

(94/356/EC)

THE COMMISSION OF THE EUROPEAN COMMUNITIES,

Having regard to the Treaty establishing the European Community,

Having regard to Council Directive 91/493/EEC of 22 July 1991 laying down the health conditions for the production and the placing on the market of fishery products (1), and in particular Article 6 (3) thereof,

Whereas, in accordance with Article 6 (3) of the said Directive, rules must be laid down for the application of the principles on which own-checks are based; whereas it is necessary to define what is meant by the identification of critical points and the establishment and implementation of methods for monitoring and checking such critical points;

Whereas laboratories must be approved by the competent authorities on equivalent terms in all the Member States;

Whereas keeping a written record or a record otherwise registered must entail keeping complete documentation containing all information relating to the establishment of own-checks and the results of those checks;

Whereas the design and introduction of own-checks will differ from one establishment to another; whereas it is therefore necessary to propose, in the form of guidelines, a model of a logical approach intended to facilitate the uniform application of Article 6 (1) of Directive 91/493/EEC;

Whereas the measures provided for in this Decision are in accordance with the opinion of the Standing Veterinary Committee,

HAS ADOPTED THIS DECISION:

Article 1

1. 'Own-checks' as referred to in the second subp graph of Article 6 (1) of Directive 91/493/EEC means those actions aimed at ensuring and demonstrating th fishery product satisfies the requirements of that Di tive. Those actions must correspond to an appro internal to the establishment; they must be develo and implemented by the persons responsible for e production unit, or under their management, in cordance with the general principles set out in the Ann hereto.

2. As part of the internal approach referred to in pa graph 1, establishments may use guides of good manu turing practice drawn up by appropriate professional o nizations and acceptable to the competent authoriti

3. The persons responsible for the establishment n ensure that all staff concerned by own-checks rec adequate training in order to effectively participate their implementation.

Article 2

1. 'Critical point' as referred to in the first inden the second subparagraph of Article 6 (1) of Direc 91/493/EEC means any point, step or procedure at wh control can be applied and a food safety hazard can prevented, eliminated or reduced to acceptable levels.

(1) OJ No L 268, 24. 9. 1991, p. 15.

critical points which are useful for ensuring compliance with the hygiene requirements of that Directive must be identified.

For the purpose of identifying these critical points, Chapter I of the Annex hereto shall apply.

2. The critical points are specific to each establishment depending on the raw materials it uses and on its manufacturing processes, structures and equipment, end products and marketing system.

Article 3

'Monitoring and checking such critical points' as referred to in the second indent of the second subparagraph of Article 6 (1) of Directive 91/493/EEC includes all those set observations and/or measurements necessary to ensure that critical points are kept under control. Monitoring and checking critical points does not include verifying that end products conform with the standards laid down in Directive 91/493/EEC.

For the purpose of introducing and implementing monitoring and checking, Chapter II of the Annex hereto shall apply.

Article 4

1. Sampling for laboratory analysis as referred to in the third indent of the second subparagraph of Article 6 (1) of Directive 91/493/EEC is intended to confirm that the own-checks system complies effectively with Articles 1, 2 and 3 of this Decision.

2. The persons responsible for the establishment must make provision for a sampling programme which, though not concerning systematically every production batch, nevertheless allows :

(a) validation of the own-checks system when first set up ;

(b) if necessary, revalidation of the system in case of a change to the characteristics of the product or to the manufacturing process ;

(c) verification, at specified intervals, that all provisions are still appropriate and properly applied.

3. Own-checks system shall be confirmed in accordance with the provisions set out in Chapter III of the Annex.

Article 5

For the approval of laboratories mentioned in the third indent of the second subparagraph of Article 6 (1) of Directive 91/493/EEC, the competent authorities of the Member States shall take into account the requirements of EN 45 001 standards or equivalent requirements. However, for the approval of establishments' internal laboratories, the competent authorities may base themselves on less restrictive principles inspired by the relevant points in Annex B to Council Directive 88/320/EEC (1).

Article 6

1. In order to keep 'a written record or a record registered in an indelible fashion', as referred to in the fourth indent of the second subparagraph of Article 6 (1) of Directive 91/493/EEC, the persons responsible for the establishment must document all information relating to the implementation of own-checks and their verification.

2. The documentation referred to in paragraph 1 must include two types of information to be kept for submission to the competent authority :

(a) a detailed and comprehensive document including :

— description of the product,

— description of the manufacturing process indicating critical points,

— for each critical point, identified hazards, assessment of risks and control measures,

— procedures for monitoring and checking at each such critical point, with indication of critical limits for parameters that need to be controlled and corrective action to be taken in case of loss of control,

— procedures for verification and review.

In the case provided for in Article 1 (2), this document may be the guide of good practice drawn up by the professional organization concerned.

(b) records of the observations and/or measurements referred to in Article 3, results of the verification activities referred to in Article 4, reports and written accounts of decisions relating to corrective action when taken. An appropriate document management system must provide, in particular, for the easy retrieval of all documents relating to an identified production batch.

Article 7

The competent authorities shall ensure appropriate training of inspection staff authorized to perform official

(1) OJ No L 145, 11. 6. 1988, p. 35.

checks to allow them to assess the own-checks system set up by the persons responsible for the establishment on the basis of the documents submitted.

Article 8

Member States shall inform the Commission of any difficulties in the application of this Decision which will be reviewed one year following its adoption, in the light of experience acquired.

Article 9

This Decision is addressed to the Member States.

Done at Brussels, 20 May 1994.

For the Commission

René STEICHEN

Member of the Commission

ANNEX

GENERAL PRINCIPLES

It is recommended that a model of a logical approach be followed, of which the following principles form the essential components :

— identification of hazards, analysis of risks and determination of measures necessary to control them,

— identification of critical points,

— establishment of critical limits for each critical point,

— establishment of monitoring and checking procedures,

— establishment of corrective action to be taken when necessary,

— establishment of verification and review procedures,

— establishment of documentation concerning all procedures and records.

Such a model, or the principles on which it is based, should be used with the flexibility appropriate to each situation.

CHAPTER I

IDENTIFICATION OF CRITICAL POINTS

It is recommended to proceed to the following activities in sequence.

1. Assembly of a multidisciplinary team

This team, which involves all parts of the enterprise concerned with the product, needs to include the whole range of specific knowledge and expertise appropriate to the product under consideration, its production (manufacture, storage, and distribution), its consumption and the associated potential hazards.

Where necessary, the team will be assisted by specialists who will help it to solve its difficulties as regards assessment and control of critical points.

The team may consist of :

— a quality control specialist who understands the biological, chemical or physical hazards connected with a particular product group,

— a production specialist who has responsibility for, or is closely involved with, the technical process of manufacturing the product under study,

— a technician who has a working knowledge of the hygiene and operation of the process plant and equipment,

— any other person with specialist knowledge of microbiology, hygiene and food technology.

One person may fulfil several of these roles, provided all relevant information is available to the team and is used to ensure that the own-checks system developed is reliable. Where expertise is not available in the establishment, advice should be obtained from other sources (consultancy, guides of good manufacturing practices, etc.).

2. Description of the product

The end product should be described in terms of :

— composition (e.g. raw materials, ingredients, additives, etc.),

— structure and physico-chemical characteristics (e.g. solid, liquid, gel, emulsion, Aw, Ph, etc.),

— processing (e.g. heating, freezing, drying, salting, smoking, etc. and to what extent),

— packaging (e.g. hermetic, vacuum, modified atmosphere),

— storage and distribution conditions,

— required shelf life (e.g. sell by date and best before date),

— instructions for use,

— any microbiological or chemical criteria applicable.

3. Identification of intended use

The multidisciplinary team should also define the normal or expected use of the product by the customer and the consumer target groups for which the product is intended. In specific cases, the suitability of the product for particular groups of consumers, such as institutional caterers, travellers, etc. and for vulnerable groups of the population may have to be considered.

4. Construction of a flow diagram (Description of manufacturing process)

Whatever the format chosen all steps involved in the process, including delays during or between steps, from receiving the raw materials to placing the end product on the market, through preparation, processing, packaging, storage and distribution, should be studied in sequence and presented in a detailed flow diagram with sufficient technical data.

Types of data may include but are not limited to:

— plan of working premises and ancillary premises,

— equipment layout and characteristics,

— sequence of all process steps (including the incorporation of raw materials, ingredients or additives and delays during or between steps),

— technical parameters of operations (in particular time and temperature, including delays),

— flow of products (including potential cross-contamination),

— segregation of clean and dirty areas (or high/low risk areas),

— cleaning and disinfection procedures,

— hygienic environment of the establishment,

— personnel routes and hygiene practices,

— product storage and distribution conditions.

5. On-site confirmation of flow diagram

After the flow diagram has been drawn up, the multidisciplinary team should confirm it on site during operating hours. Any observed deviation must result in an amendment of the original flow diagram to make it accurate.

6. Listing of hazards and control measures

Using the confirmed flow diagram as a guide, the team should:

(a) list all potential biological, chemical or physical hazards that may be reasonably expected to occur at each process step (including acquisition and storage of raw materials and ingredients and delays during manufacture).

A hazard is a potential to cause harm to health and is anything covered by the hygiene objectives of Directive 91/493/EEC. Specifically, it can be any of the following:

— unacceptable contamination (or recontamination) of a biological (micro-organisms, parasites), chemical or physical nature of raw materials, intermediate products or final products,

— unacceptable survival or multiplication of pathogenic micro-organisms and unacceptable generation of chemicals in intermediate products, final products, production line or line environment,

— unacceptable production or persistence of toxins or other undesirable products of microbial metabolism.

For inclusion in the list, hazards must be of a nature such that their elimination or reduction to acceptable levels is essential to the to the production of safe food.

(b) consider and describe what control measures, if any, exist which can be applied for each hazard.

Control measures are those actions and activities that can be used to prevent hazards, eliminate them or reduce their impact or occurrence to acceptable levels.

More than one control measure may be required to control an identified hazard and more than one hazard may be controlled by one control measure. For instance, pasteurization or controlled heat treatment may provide sufficient assurance of reduction of the level of both *salmonella* and *listeria*.

Control measures need to be supported by detailed procedures and specifications to ensure their effective implementation. For instance, detailed cleaning schedules, precise heat treatment specifications, maximum concentrations of preservatives used in compliance with the applicable Community rules on additives and in particular Directive 89/107/EEC (1).

7. Methods for identification of criticla points

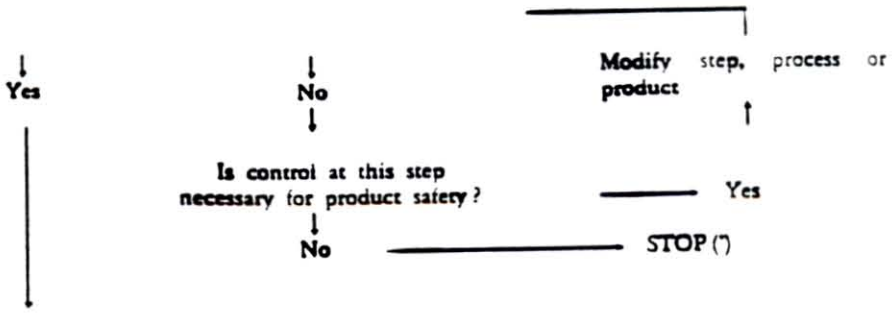
The identification of a critical point for the control of a hazard requires a logical approach. Such an approach can be facilitated by the use of the following decision tree (other methods can be used by the team, according to their knowledge and experience).

Decision tree for the identification of critical points

Answer each question in sequence, at each step and for each identified hazard.

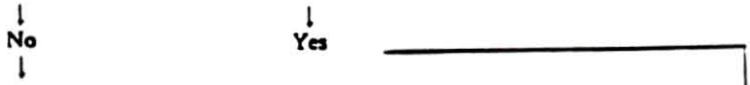
Question 1

Are control measures in place for the hazard?



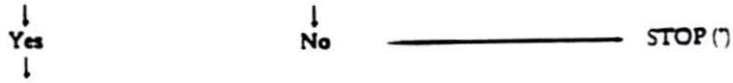
Question 2

Does that step eliminate or reduce the hazard to an acceptable level?



Question 3

Could contamination occur at, or hazard increase to, an unacceptable level?



Question 4

Will a subsequent step eliminate or reduce the hazard to an acceptable level?

(*) The step is not a critical point. Proceed to next step.

(1) OJ No L 40, 11. 2. 1989, p. 27.

For the application of the decision tree, each process step identified in the flow diagram should be considered in sequence. At each step, the decision tree must be applied to each hazard that may be reasonably expected to occur or be introduced and each control measure identified.

Application of the decision tree should be flexible and requires common sense, having consideration for the whole manufacturing process in order to avoid, whenever possible, unnecessary critical points.

8. Action to be taken following identification of a critical point

The identification of critical points has two consequences for the multidisciplinary team which should then :

— ensure that appropriate control measures are effectively designed and implemented. In particular, if a hazard has been identified at a step where control is necessary for product safety and no control measure exists at that step,or at any other, then the product or process should be modified at that step, or at any other, then the product or process should be modified at that step, or at an earlier or later stage, to include a control measure,

— establish and implement a monitoring and checking system at each critical point.

CHAPTER II :

ESTABLISHMENT AND IMPLEMENTATION OF MONITORING AND CHECKING CRITICAL POINTS

An appropriate monitoring and checking system is essential to ensure the effective control of each critical point.

To develop such a system, it is recommended to proceed to the following activities :

1. Establishment of critical limits for each control measure associated with each critical point

Each control measure associated with a critical point should give rise to the specification of critical limits.

Those critical limits correspond to the extreme values acceptable with regard to product safety. They separate acceptability from unacceptability. They are set for observable or measurable parameters which can readily demonstrate that the critical point is under control ; they should be based on substantiated evidence that chosen values will result in process control.

Examples of such parameters include temperature, time, pH, moisture level, additive, preservative or salt level, sensory parameters such as visual appearance or texture, ect.

In some cases, to reduce the risk of exceeding a critical limit due to process variations, it may be necessary to specify more stringent levels (i.e. target levels) to assure that critical limits are observed.

Critical limits may be derived from a variety of sources. When not taken from regulatory standards (e.g. frozen storage temperature) or from existing and validated guides of good manufactoring practices, the team should ascertain their validity relative to the control of identified hazard and critical points.

2. Establishment of a monitoring and checking system for each critical point

An essential part of own-checks is a programme of observations or measurements performed at each critical point to ensure compliance with specified critical limits. The programme should describe the methods, the frequency of observations or measurements and the recording procedure.

Observations or measurements must be able to detect loss of control at critical points and provide information in time for corrective action to be taken.

Observations or measurements can be made continuously or discontinuously. When observations or measurements are not continuous, it is necessary to establish a frequency of observations or measurements which provides reliable information.

The programme of observations or measurements should properly identify for each critical point :

— who is to perform monitoring and checking,

— when monitoring and checking is performed,

— how monitoring and checking is performed.

3. **Establishment of a corrective action plan**

Observations or measurements may indicate:

— that the parameter monitored tends to deviate from its specified critical limits, indicating a trend toward loss of control. Appropriate corrective action to maintain control must be taken before the occurence of hazard,

— that the parameter monitored has deviated from its specified critical limits, indicating a loss of control. It is necessary to take appropriate corrective action to regain control.

Corrective action has to be planned in advance by the multidisciplinary team, for each critical point, so that it can be taken without hesitation when a deviation is observed.

Such corrective action should include:

— proper identification of the person(s) responsible for the implementation of the corrective action,

— description of means and action required to correct the observed deviation,

— action to be taken with regard to products that have been manufactured during the period when the process was out of control,

— written record of measures taken.

CHAPTER III:

VERIFICATION OF OWN-CHECKS SYSTEMS

Own-checks system verification is necessary to ensure that they are working effectively. The multidisciplinary team should specify the methods and procedures to be used.

Usable methods may include in particular random sampling and analysis, reinforced analysis or tests at selected critical points, intensified analysis of intermediate or final products, surveys on actual condition during storage, distribution and sale and on actual use of the product.

Verification procedures may include: inspection of operations, validation of critical limits, review of deviations, corrective action and measures taken with regard to the product, audits of the own-check system and its records.

Verification should provide for confirmation of the suitability of the own-checks system established and ensure, afterwards, with an appropriate frequency, that the provisions laid down are still being properly applied.

In addition, it is necessary to review the system, to ensure that it is (or will be) still valid in case of change.

Examples of change include:

— change in raw material or in product, processing conditions (factory layout and environment, process equipment, cleaning and disinfection programme),

— change in packaging, storage or distribution conditions,

— change in consumer use,

— receipt of any information on a new hazard associated with the product.

Where necessary, such a review must result in the amendment of the provisions laid down.

Any change to the own-checks system arising should be fully incorporated into the documentation and record-keeping system in order to ensure that accurate up-to-date information is available.

Where criteria are specified in regulations, such criteria are to be used as reference values for the verification process.

CORRIGENDA

Corrigendum to Commission Regulation (EC) No 1097/94 of 11 May 1994 on transitional measures concerning the allocation of quotas in the tobacco sector for the 1994 harvest

(Official Journal of the European Communities No L 121 of 12 May 1994)

On page 11 in the Annex:

for: '200 tonnes from Group I, "flue-cured", to Group II, "light flue-cured".',

read: '200 tonnes from Group I, "flue-cured", to Group II, "light air-cured".'

COUNCIL DIRETTIVE

of 22 July 1991

laying down the health conditions for the production and the placing on the market of fishery products

(91/493/EEC)

THE COUNCIL OF THE EUROPEAN COMMUNITIES,

Having regard to the Treaty establishing the European Economic Community, and in particular Article 43 thereof,

Having regard to the proposals from the Commission (1),

Having regard to the opinions of the European Parliament (2),

Having regard to the opinions of the Economic and Social Committee (3),

Whereas, with a view to achieving the internal market and more especially to ensuring the smooth operation of the common organization of the market in fishery products established by Regulation (EEC) No 3796/81 (4), as last amended by Regulation (EEC) No 2886/89 (5), it is essential that the marketing of fish and fish products should no longer be hindered by disparities existing in the Member States in respect of health requirements; whereas this will enable production and placing on the market to be better harmonized and bring about competition on equal terms, whilst ensuring quality products for the consumer;

Whereas the European Parliament in its legislative resolution of 17 March 1989 (6) requested the Commission to come forward with comprehensive proposals on the hygienic production and placing on the market of fishery products, including solutions for the problem of nematodes;

Whereas fishery products freshly caught are in principle free of contamination with micro-organisms; whereas however contamination and subsequent decomposition may occur when handled and treated unhygienically;

Whereas therefore the essential requirements should be laid down for the correct hygienic handling of fresh and processed fishery products at all stages of production and during storage and transport;

Whereas it is appropriate to apply by analogy certain marketing standards which are laid down pursuant to Article 2 of Regulation (EEC) No 3796/81, in order to fix the health quality of these products;

Whereas it is the responsibility primarily of the fisheries industry to ensure that fishery products meet the health requirements laid down in this Directive;

Whereas the competent authorities of the Member States must, by carrying out checks and inspections, ensure that producers and manufacturers comply with the said requirements;

Whereas Community control measures should be introduced to guarantee the uniform application in all Member States of the standards laid down in this Directive;

Whereas, in order to ensure the smooth operation of the internal market, the measures should apply in an identical manner to trade within the Member States and to trade between the Member States;

Whereas in the context of intra-Community trade, the rules laid down in Council Directive 89/662/EEC of 11 December 1989 concerning veterinary checks in intra-Community trade with a view to the completion of the internal market (7) as amended by Directive 90/675/EEC (8) apply to fishery products;

Whereas fishery products from third countries intended to be placed on the market of the Community must not qualify for more favourable arrangements than those applied in the Community; whereas provision should therefore be made for a Community procedure for the inspection in third countries of the conditions of production and placing on the market in order to permit the application of a common import system based on conditions of equivalence;

Whereas the products in question are subject to the rules concerning checks and to safeguard measures covered by Council Directive 90/675/EEC of 10 December 1990 laying down the principles governing the organization of veterinary checks on products entering the Community from third countries;

Whereas, so that account may be taken of particular circumstances, derogations should be granted to some establishments already operating before 1 January 1993 so as to allow them to adapt to all the requirements laid down in this Directive;

(1) OJ No C 66, 11. 3. 1988, p. 2;
OJ No C 282, 8. 11. 1989, p. 7 and OJ No C 84, 2. 4. 1990, p. 56.
(2) OJ No C 96, 17. 4. 1989, p. 29 and OJ No C 183, 15. 7. 1991.
(3) OJ No C 134, 24. 5. 1988, p. 31 and OJ No C 332, 31. 12. 1990, p. 59.
(4) OJ No L 379, 31. 12. 1981, p. 1.
(5) OJ No L 282, 2. 10. 1989, p. 1.
(6) OJ No C 96, 17. 4. 1989, p. 199.
(7) OJ No L 395, 30. 12. 1989, p. 13.
(8) OJ No L 373, 31. 12. 1990, p. 1.

Whereas the Commission should be entrusted with the task of adopting certain measures for implementing this Directive; whereas, to that end, procedures should be laid down introducing close and effective cooperation between the Commission and the Member States within the Standing Veterinary Committee;

Whereas the essential requirements laid down in this Directive may need further specification,

HAS ADOPTED THIS DIRECTIVE

CHAPTER I

General provisions

Article 1

This Directive lays down the health conditions for the production and the placing on the market of fishery products for human consumption.

Article 2

For the purposes of this Directive, the following definitions shall apply:

1. *'fishery products'* means all seawater or freshwater animals or parts thereof, including their roes, excluding aquatic mammals, frogs and aquatic animals covered by other Community acts;

2. *'aquaculture products'* means all fishery products born and raised in controlled conditions until placed on the market as a foodstuff. However seawater or freshwater fish or crustaceans caught in their natural environment when juvenile and kept until they reach the desired commercial size for human consumption are also considered to be aquaculture products. Fish and crustaceans of commercial size caught in their natural environment and kept alive to be sold at a later date are not considered to be aquaculture products if they are merely kept alive without any attempt being made to increase their size or weight;

3. *'chilling'* means the process of cooling fishery products to a temperature approaching that of melting ice;

4. *'fresh products'* means any fishery product whether whole or prepared, including products packaged under vacuum or in a modified atmosphere, which have not undergone any treatment to ensure preservation other than chilling;

5. *'prepared products'* means any fishery product which has undergone an operation affecting its anatomical wholeness, such as gutting, heading, slicing, filleting, chopping, etc.;

6. *'processed products'* means any fishery product which has undergone a chemical or physical process such as the heating, smoking, salting, dehydration or marinating, etc., of chilled or frozen products, whether or not associated with other foodstuffs, or a combination of these various processes;

7. *'preserve'* means the process whereby products are packaged in hermetically sealed containers and subjected to heat treatment to the extent that any micro-organisms that might proliferate are destroyed or inactivated, irrespective of the temperature at which the product is to be stored;

8. *'frozen products'* means any fishery product which has undergone a freezing process to reach a core temperature of −18 °C or lower after temperature stabilization;

9. *'packaging'* means the procedure of protecting fishery products by a wrapper, a container or any other suitable device;

10. *'batch'* means the quantity of fishery products obtained under practically identical circumstances;

11. *'consignment'* means the quantity of fishery products bound for one or more customers in the country of destination and conveyed by one means of transport only;

12. *'means of transport'* means those parts set aside for goods in automobile vehicles, rail vehicles and aircraft, the holds of vessels, and containers for transport by land, sea or air;

13. *'competent authority'* means the central authority of a Member State competent to carry out veterinary checks or any authority to which it has delegated that competence;

14. *'establishment'* means any premises where fishery products are prepared, processed, chilled, frozen, packaged or stored. Auction and wholesale markets in which only display and sale by wholesale takes place are not deemed to be establishments;

15. *'placing on the market'* means the holding or displaying for sale, offering for sale, selling, delivering or any other form of placing on the market in the Community, excluding retail sales and direct transfers on local markets of small quantities by fishermen to retailers or consumers, which must be subject to the health checks laid down by national rules for checking the retail trade;

16. *'importation'* means the introduction into the territory of the Community of fishery products from third countries;

17. *'clean seawater'* means seawater or briny water which is free from microbiological contamination, harmful substances and/or toxic marine plankton in such quantities as may affect the health quality of fishery products and which is used under the conditions laid down in this Directive;

18. *'factory vessel'* means any vessel on which fishery products undergo one or more of the following operations followed by packaging: filleting, slicing, skinning, mincing, freezing or processing.

The following are not deemed to be 'factory vessels':

— fishing vessels in which only shrimps and molluscs are cooked on board;

— fishing vessels on board which only freezing is carried out.

Article 3

1. The placing on the market of fishery products caught in their natural environment shall be subject to the following conditions:

(a) they must have:

(i) been caught and where appropriate handled for bleeding, heading, gutting and the removal of fins, chilled or frozen, on board vessels in accordance with hygiene rules to be established by the Council acting by a qualified majority on a proposal from the Commission. The Commission shall submit proposals to that effect before 1 October 1992;

(ii) where appropriate, been handled in factory vessels approved in accordance with Article 7, and in accordance with the requirements of Chapter I of the Annex.

The cooking of shrimps and molluscs on board must comply with the provisions of Chapter III, section I(5), or Chapter IV, section IV(7), of the Annex. Such vessels shall be specifically registered by the competent authorities;

(b) during and after landing they must have been handled in accordance with Chapter II of the Annex;

(c) they must have been handled and, where appropriate, packaged, prepared, processed, frozen, defrosted or stored hygienically in establishments approved in accordance with Article 7, in compliance with the requirements of Chapters III and IV of the Annex.

The competent authority may, notwithstanding Chapter II, section 2 of the Annex, authorize the transfer of fishery products *ex quay* into containers for immediate delivery to an approved establishment or registered auction or wholesale market to be checked there;

(d) they must have undergone a health check in accordance with Chapter V of the Annex;

(e) they must have been appropriately packaged in accordance with Chapter VI of the Annex;

(f) they must have been given an identification mark in accordance with Chapter VII of the Annex;

(g) they must have been stored and transported under satisfactory conditions of hygiene, in accordance with Chapter VIII of the Annex.

2. Where gutting is possible from a technical and commercial viewpoint, it must be carried out as quickly as possible after the products have been caught or landed.

3. The placing on the market of aquaculture product shall be subject to the following conditions:

(a) they must have been slaughtered under appropriat conditions of hygiene. They must not be soiled wit earth, slime or faeces. If not processed immediately afte having been slaughtered, they must be kept chilled;

(b) they must, in addition, comply with the requiremen laid down under 1 (c) to (g).

4. (a) The placing on the market of live bivalve mollus shall be subject to the requirements laid down Council Directive 91/492/EEC of 15 July 199 laying down the health conditions for the productic and the placing on the market of live bival molluscs (1).

(b) When processed, bivalve molluscs must, in additio to the requirements in point (a), satisfy those c paragraph 1 (c) to (g).

Article 4

Fishery products to be placed on the market alive shall at times be kept under the most suitable survival condition

Article 5

The placing on the market of the following products shall forbidden:

— poisonous fish of the following families: *Tetraodonti Molidae, Diodontidae, Canthigasteridae,*

— fishery products containing biotoxins such as ciguat toxins or muscle-paralysing toxins.

Detailed requirements concerning the species covered by Article and concerning methods of analysis shall be down in accordance with the procedure prescribed Article 15.

(1) See page 1 of this Official Journal.

Article 6

1. Member States shall ensure that persons responsible for establishment take all necessary measures, so that, at all stages of the production of fishery products, the specifications of this Directive are complied with.

To that end, the said persons responsible must carry out their own checks based on the following principles:

— identification of critical points in their establishment on the basis of the manufacturing processes used;

— establishment and implementation of methods for monitoring and checking such critical points;

— taking samples for analysis in an approved laboratory by the competent authority for the purpose of checking cleaning and disinfection methods and for the purpose of checking compliance with the standards established by this Directive;

— keeping a written record or a record registered in an indelible fashion of the preceding points with a view to submitting them to the competent authority. The results of the different checks and tests will in particular be kept for a period of at least two years.

2. If the results of own checks or any information at the disposal of the persons responsible referred to in paragraph 1 reveal the risk of a health risk or suggest one might exist and without prejudice to the measures laid down in the fourth subparagraph of Article 3 (1) of Directive 89/662/EEC, the appropriate measures shall be taken, under official supervision.

3. Rules for the application of the second subparagraph of paragraph 1 shall be established in accordance with the procedure laid down in Article 15.

Article 7

1. The competent authorities shall approve establishments once they have verified that these establishments meet the requirements of this Directive, with regard to the nature of the activities they carry out. The approval must be renewed if an establishment decides to carry out activities other than those for which it has received approval.

The competent authorities shall take the necessary measures if the requirements cease to be met. To this end, they shall take particular account of the conclusions of any check carried out in accordance with Article 8.

The competent authority shall register those auction and wholesale markets which are not subject to approval after verifying that such installations comply with the provisions of this Directive.

2. However, subject to the express condition that products coming from factory-vessels and establishments, auction and wholesale markets meet the hygiene standards set by this Directive, Member States may, for the requirements relating to equipment and structures laid down in Chapters I to IV to the Annex, grant to factory-vessels and establishments, auction and wholesale markets a further period expiring on 31 December 1995 within which to comply with the conditions of approval set out in Chapter IX. Such derogations may be granted only to factory-vessels and establishments, auction and wholesale markets, already operating on 31 December 1991, which have, before 1 July 1992, submitted a duly justified application for derogation to the competent national authority. This application must be accompanied by a work plan and programme indicating the period within which it would be possible for them to comply with the requirements in question. Where financial assistance is requested from the Community, only requests in respect of projects complying with the requirements of this Directive can be accepted.

3. The competent authorities shall draw up a list of their approved establishments, each of which shall have an official number.

Each Member State shall notify the Commission of its list of approved establishments and of any subsequent amendment thereof. The Commission shall forward this information to the other Member States.

4. The inspection and monitoring of establishments shall be carried out regularly under the responsibility of the competent authority, which shall at all times have free access to all parts of establishments, in order to ensure compliance with the requirements of this Directive.

If such inspections and monitoring reveal that the requirements of this Directive are not being met, the competent authority shall take appropriate action.

5. Paragraphs 1, 3 and 4 shall also apply in respect of factory vessels.

6. Paragraphs 3 and 4 shall also apply to wholesale and auction markets.

Article 8

1. Experts from the Commission may, in cooperation with the competent authorities of the Member States, make on-the-spot checks insofar as this is necessary to ensure the uniform application of this Directive. They may in particular verify whether establishments are in effect complying with the requirements of this Directive. A Member State in whose territory a check is being carried out shall give all necessary assistance to the experts in carrying out their duties. The Commission shall inform the Member States of the results of the investigations.

2. The arrangements for implementing paragraph 1 shall be adopted in accordance with the procedure laid down in Article 15.

Article 9

1. The rules laid down in Directive 89/662/EEC, as regards fishery products intended for human consumption, shall apply, in particular as regards the organization of and the action to be taken following the inspections to be carried out by the Member States of destination, and the protective measures to be implemented.

2. Directive 89/662/EEC shall be amended as follows:

(a) in Annex A the following indent shall be added:

'— Council Directive 91/493/EEC of 22 July 1991 laying down the health conditions for the production and placing on the market of fishery products (OJ No L 268, 24. 9. 1991, p. 15);'

(b) In Annex B the following indent shall be deleted:

'— fishery products intended for human consumption'.

CHAPTER II

Imports from third countries

Article 10

Provisions applied to imports of fishery products from third countries shall be at least equivalent to those governing the production and placing on the market of Community products.

Fishery products caught in their natural environment by a fishing vessel flying the flag of a third country must undergo the checks laid down in Article 18 (3) of Directive 90/675/EEC.

Article 11

1. For each third country or group of third countries, fishery products must fulfil the specific import conditions fixed in accordance with the procedure laid down in Article 15, depending on the health situation in the third country concerned.

2. In order to allow the import conditions to be fixed, and in order to verify the conditions of production, storage and dispatch of fishery products for consignment to the Community, inspections may be carried out on the spot by experts from the Commission and the Member States.

The experts of the Member States who are to be entrusted with these inspections shall be appointed by the Commission acting on a proposal from the Member States.

These inspections shall be made on behalf of the Community, which shall bear any expenditure incurred.

The frequency of and procedure for these inspections shall be determined in accordance with the procedure laid down in Article 15.

3. When fixing the import conditions of fishery products referred to in paragraph 1, particular account shall be taken of:

(a) the legislation of the third country;

(b) the organization of the competent authority of the third country and of its inspection services, the powers of such services and the supervision to which they are subject, as well as their facilities for effectively verifying the implementation of their legislation in force;

(c) the actual health conditions during the production, storage and dispatch of fishery products intended for the Community;

(d) the assurances which a third country can give on the compliance with the standards laid down in Chapter V of the Annex.

4. The import conditions referred to in paragraph 1 shall include:

(a) the procedure for obtaining a health certificate which must accompany consignments when forwarded to the Community;

(b) the placing of a mark identifying the fishery products, in particular with the approval number of the establishment of origin, except in the case of frozen fishery products, landed immediately for canning and bearing the certificate provided for under (a);

(c) drawing up a list of approved establishments and auction or wholesale markets registered and approved by the Commission in accordance with the procedure laid down in Article 15;

For that purpose, one or more lists of such establishments shall draw up on the basis of a communication from the competent authorities of the third country to the Commission. An establishment may not appear on a list unless it is officially approved by the competent authority of the third country exporting to the Community. Such approval shall be subject to observance of the following requirements:

— compliance with requirements equivalent to those laid down in this Directive,

— monitoring by an official inspection service of the third country.

5. The conditions referred to in paragraph 4 (a) and (b) may be modified in accordance with the procedure laid down in Article 15.

The list referred to in paragraph 4 (c) may be amended by the Commission, in accordance with the rules established by Commission Decision 90/13/EEC (1).

6. To deal with specific situations and in accordance with the procedure laid down in Article 15, imports may be authorized direct from an establishment or factory vessel of a third country where the latter is unable to provide the guarantees laid down in paragraph 3, provided that the establishment or factory vessel in question has received special approval following an inspection carried out in accordance with paragraph (2). The authorization decision shall fix the specific import conditions to be followed for products coming from that establishment or factory vessel.

7. Pending the fixing of the import conditions referred to in paragraph 1, the Member States shall ensure that the conditions applied to imports of fishery products from third countries shall be at least equivalent to those governing the production and placing on the market of Community products.

Article 12

1. The rules and principles laid down by Directive 90/675/EEC shall apply, notably as regards the organization of and follow up to the inspections to be carried out by the Member States.

2. Without prejudice to compliance with the rules and principles referred to in paragraph 1 of this Article and pending implementation of the decisions provided for in Article 8 (3) and Article 30 of Directive 90/675/EEC, and in Article 11 of this Directive the relevant national rules for applying Article 8 (1) and (2) of the said Directive shall continue to apply.

CHAPTER III

Final provisions

Article 13

The Annexes shall be amended by the Council, acting by a qualified majority on a proposal from the Commission.

Article 14

The Commission, after consulting the Member States, shall by 1 July 1992 submit a report to the Council concerning the minimum structural and equipment requirements to be met by small establishments which distribute on the local market and are situated in regions subject to particular supply constraints, together with any proposals, on which the Council, acting under the voting procedure laid down in Article 43 of the Treaty, shall act before 31 December 1992.

Article 15

1. Where the procedure laid down in this Article is to be followed, the Chairman shall refer the matter to the Standing Veterinary Committee set up by Decision 68/361/EEC (2) hereafter referred to as the Committee, either on his own initiative or at the request of a Member State.

2. The representative of the Commission shall submit to the committee a draft of the measures to be taken. The committee shall deliver its opinion on the draft within a time limit which the chairman may lay down according to the urgency of the matter. The opinion shall be delivered by the majority laid down in Article 148 (2) of the Treaty in the case of decisions which the Council is required to adopt on a proposal from the Commission. The votes of the representatives of the Member States within the committee shall be weighted in the manner set out in that Article. The chairman shall not vote.

3. (a) The Commission shall adopt the measures envisaged if they are in accordance with the opinion of the committee.

(b) If the measures envisaged are not in accordance with the opinion of the committee, or if no opinion is delivered, the Commission shall, without delay, submit to the Council a proposal relating to the measures to be taken. The Council shall act by a qualified majority.

If, on the expiry of a period of three months from the date of referral to the Council, the Council has not acted, the proposed measures shall be adopted by the Commission, save where the Council has decided against the said measures by a simple majority.

Article 16

In order to take into account the possible failure to take a decision on the detailed rules for applying this Directive by 1 January 1993, necessary transitional measures may be adopted in accordance with the procedure laid down in Article 15 for a period of two years.

Article 17

The provisions of this Directive shall be re-examined before 1 January 1998 by the Council, acting on proposals from the Commission, on the basis of experience gained.

(1) OJ No L 8, 11. 1. 1990, p. 70.

(2) OJ No L 255, 18. 10. 1968, p. 23.

Article 18

The Member States shall bring into force the laws, regulations and administrative provisions necessary to comply with this Directive before 1 January 1993. They shall notify the Commission thereof.

When Member States adopt these measures, they shall contain a reference to this Directive or shall be accompanied by such reference on the occasion of their official publication. The methods of making such a reference shall be laid down by the Member States.

Article 19

This Directive is addressed to the Member States.

Done at Brussels, 22 July 1991.

For the Council

The President

P. DANKERT

ANNEX

CHAPTER I

CONDITIONS APPLICABLE TO FACTORY VESSELS

I. **Conditions concerning design and equipment**

1. The minimum requirements for factory vessels are as follows:

(a) a reception area set aside for taking fishery products on board, designed and arranged into pounds or pens that are large enough to allow each successive catch to be separated. The reception area and its movable parts must be easy to clean. It must be designed in such a way as to protect the products from the sun or the elements and from any source of dirt or contamination;

(b) a system for conveying fishery products from the reception area to the work area that conforms with rules of hygiene;

(c) work areas that are large enough for the preparation and processing of fishery products in proper conditions of hygiene. They must be designed and arranged in such a way as to prevent any contamination of the products;

(d) storage areas for the finished products that are large enough and designed so that they are easy to clean. If a waste processing unit operates on board, a separate hold must be designated for the storage of these by-products;

(e) a place for storing packaging materials that is separate from the product preparation and processing areas;

(f) special equipment for pumping waste or fishery products that are unfit for human consumption either directly into the sea or, where circumstances so require, into a watertight tank reserved for that purpose. If waste is stored and processed on board with a view to cleaning, separate areas must be allocated for that purpose;

(g) equipment providing a supply of potable water within the meaning of Council Directive 80/778/EEC of 15 July 1980 relating to the quality of water intended for human consumption [1] or pressurized clean seawater. The seawater intake must be situated in a position where it is not possible for the water being taken in to be affected by discharges into the sea of waste water, waste and engine coolant outlets;

(h) a suitable number of changing rooms, wash basins and toilets, the latter not opening directly onto areas where fishery products are prepared, processed or stored. The wash basins must be equipped with appliances for washing and drying the hands that comply with hygiene requirements; the wash-basin taps must not be hand-operable.

2. Areas used for the preparation and processing or freezing/quick-freezing of fishery products must have:

(a) a non-slip floor that is also easy to clean and disinfect and equipped for easy drainage of water. Structures and fixtures must have timber holds that are large enough not to be obstructed by fish waste and to allow water to drain freely;

(b) walls and ceilings that are easy to clean, particularly where there are pipes, chains or electricity conduits;

(c) the hydraulic circuits must be arranged or protected in such a way as to ensure that it is not possible for any leakage of oil to contaminate fishery products;

(d) adequate ventilation and, where necessary, proper vapour extraction;

(e) adequate lighting;

(f) appliances for cleaning and disinfecting tools, equipment and fittings;

(g) appliances for cleaning and disinfecting the hands with taps that are not hand-operable and with single use towels.

[1] OJ No L 229, 30. 9.1980, p. 11. Directive last amended by the 1985 Act of Accession (OJ No L 302, 15. 11. 1985, p. 218).

3. Equipment and tools such as cutting benches, containers, conveyors, gutting or filleting machines, etc., must be resistant to seawater corrosion, easy to clean and disinfect and well-maintained.

4. Factory vessels which freeze fishery products must have:

   (a) a refrigeration plant sufficiently powerful to lower the temperature rapidly so as to achieve a core temperature that complies with the specifications of this Directive;

   (b) refrigeration plants sufficiently powerful to keep fishery products in the storage holds at a temperature that complies with the specifications of this Directive. The storage holds must be equipped with a temperature recording system placed so that it can easily be consulted.

II. Conditions of hygiene relating to on-board handling and storage of fishery products

1. A qualified person on board the factory vessel must be responsible for applying good fishery products manufacturing practices. That person shall have the authority to ensure that the provisions of this Directive are applied and shall make available to inspectors the programme for inspecting and checking critical points as applied on board, a register containing that person's comments and the temperature recordings that may be required.

2. The general conditions of hygiene applicable to areas and equipment shall be those laid down in Chapter III, section II (A), of this Annex.

3. The general conditions of hygiene applicable to staff shall be those laid down in Chapter III, section II (B), of this Annex.

4. Heading, gutting and filleting must be carried out under the conditions of hygiene laid down in Chapter IV, section I (2), (3) and (4) of this Annex.

5. On-board processing of fishery products must be carried out under the conditions of hygiene laid down in Chapter IV, sections III, IV and V of this Annex.

6. Fishery products must be wrapped and packaged under the conditions of hygiene laid down in Chapter VI of this Annex.

7. On-board storage of fishery products must be carried out under the conditions of hygiene laid down in Chapter VIII, points 1 and 2, of this Annex.

CHAPTER II

REQUIREMENTS DURING AND AFTER LANDING

1. Unloading and landing equipment must be constructed of material which is easy to clean and disinfect and must be kept in a good state of repair and cleanliness.

2. During unloading and landing, contamination of fishery products must be avoided. It must in particular be ensured that:

   — unloading and landing operations proceed rapidly;

   — fishery products are placed without unnecessary delay in a protected environment at the temperature required on the basis of the nature of the product and, where necessary, in ice in transport, storage or market facilities, or in an establishment;

   — equipment and handling practices that cause unnecessary damage to the edible parts of the fishery products are not authorized.

3. Parts of auction or wholesale markets where fishery products are displayed for sale must:

   a) be covered and have walls which are easy to clean;

   (b) have waterproof flooring which is easy to wash and disinfect and laid in such a way as to facilitate the drainage of water and have a hygienic waste water disposal system;

(c) be equipped with sanitary facilities with an appropriate number of wash basins and flush lavatories. Wash basins shall be supplied with materials for cleaning the hands and single use hand towels;

(d) be well lit to facilitate the inspection of fishery products provided for in Chapter V of this Annex;

(e) when they are used for display or storage of fishery products, not be used for other purposes: vehicles emitting exhaust fumes which may impair the quality of the fishery products not be admitted to markets; undesirable animals must not be admitted;

(f) be cleaned regularly and at least after each sale; crates must, after each sale, be cleaned and rinsed inside and outside with drinking water or clean seawater; where required, they must be disinfected;

(g) have displayed in a prominent position signs prohibiting smoking, spitting, eating and drinking;

(h) be closeable and be kept closed when the competent authority considers it necessary;

(i) have facilities to provide adequate water supplies satisfying the conditions laid down in Chapter III, section I, point 7 of this Annex;

(j) have special watertight receptacles made of corrosion-resistant materials for fishery products which are unfit for human consumption;

(k) insofar as they do not have their own premises on-the-spot or in the immediate vicinity on the basis of the quantities displayed for sale, have, for the purposes of the competent authority, an adequately equipped lockable room and the equipment necessary for carrying out inspections.

4. After landing or, where appropriate, after first sale, fishery products must be transported without delay, under the conditions laid down in Chapter VIII, of this Annex, to their place of destination.

5. However, if the conditions laid down in point 4 are not fulfilled, the markets in which fishery products may be stored before being displayed for sale or after being sold and pending transport to their place of destination must have sufficiently large cold rooms which satisfy the conditions laid down in Chapter III, section I, point 3 of this Annex. In such cases, fishery products must be stored at a temperature approaching that of melting ice.

6. The general conditions of hygiene laid down in Chapter III, section II — with the exception of point B 1(a) — of this Annex shall apply *mutatis mutandis* to the markets in which fishery products are displayed for sale or stored.

7. The wholesale markets in which fishery products are displayed for sale or stored shall be subject to the same conditions as those laid down in points 3 and 5 of this Chapter and to those set out in points 4, 10 and 11 of Chapter III, section I of this Annex.

The general conditions of hygiene laid down in Chapter III, section II of this Annex shall apply *mutatis mutandis* to wholesale markets.

CHAPTER III

GENERAL CONDITIONS FOR ESTABLISHMENTS ON LAND

I. General conditions relating to premises and equipment

Establishment shall afford at least the following facilities:

1. working areas of sufficient size for work to be carried out under adequate hygienic conditions. Their design and layout shall be such as to preclude contamination of the product and keep quite separate the clean and contaminated parts of the building;

2. in areas where products are handled, prepared and processed:

(a) waterproof flooring which is easy to clean and disinfect and laid down in such a way as to facilitate the drainage of the water or provided with equipment to remove water;

(b) walls which have smooth surfaces and are easy to clean, durable and impermeable;

(c) ceilings or roof linings which are easy to clean;

(d) doors in durable materials which are easy to clean;

(e) adequate ventilation and, where necessary, good steam and water-vapour extraction facilities;

(f) adequate natural or artificial lighting;

(g) an adequate number of facilities for cleaning and disinfecting hands. In work rooms and lavatories taps must not be hand-operable. These facilities must be provided with single use hand towels;

(h) facilities for cleaning plant, equipment and utensils;

3. in cold rooms where fishery products are stored:

— the provisions set out under point 2 (a), (b), (c), (d) and (f);

— where necessary, a sufficiently powerful refrigeration plant to keep products at temperatures prescribed in this Directive;

4. appropriate facilities for protection against pests such as insects, rodents, birds, etc.;

5. instruments and working equipment such as cutting tables, containers, conveyor belts and knives made of corrosion-resistant materials, easy to clean and disinfect;

6. special watertight, corrosion-resistant containers for fishery products not intended for human consumption and premises for the storage of such containers if they are not emptied at least at the end of each working day;

7. facilities to provide adequate supplies of drinking water within the meaning of Directive 80/778/EEC, or alternatively of clean seawater or seawater treated by an appropriate system, under pressure and in sufficient quantity. However, by way of exception, a supply of non-drinking water is permissible for the production of steam, fire-fighting and the cooling of refrigeration equipment, provided that the pipes installed for the purpose preclude the use of such water for other purposes and present no risk of contamination of the products. Non-drinking-water pipes must be clearly distinguished from those used for drinking water or clean seawater;

8. hygienic waste water disposal system;

9. an adequate number of changing-rooms with smooth, water-proof, washable walls and floors, wash basins and flush lavatories. The latter may not open directly onto the work rooms. The wash basins must have materials for cleaning the hands and disposable towels; the wash basin taps must not be hand-operable;

10. if the volume of products treated requires regular or permanent presence an adequately equipped lockable room for the exclusive use of the inspection service;

11. adequate facilities for cleaning and disinfecting means of transport. However, such facilities are not compulsory if there is a requirement for the means of transport to be cleaned and disinfected at facilities officially authorized by the competent authority;

12. establishments keeping live animals such as crustaceans and fish must have appropriate fittings ensuring the best survival conditions provided with water of a quality such that no harmful organisms or substances are transferred to the animals.

II. General conditions of hygiene

A. *General conditions of hygiene applicable to premises and equipment*

1. Floors, walls and partitions, ceilings or roof linings, equipment and instruments used for working on fishery products must be kept in a satisfactory state of cleanliness and repair, so that they do not constitute a source of contamination for the products.

2. Rodents, insects and any other vermin must be systematically exterminated in the premises or on the equipment; rodenticides, insecticides, disinfectants and any other potentially toxic substances must be stored in premises or cupboards which can be locked; their use must not present any risk of contamination of the products.

3. Working areas, instruments and working equipment must be used only for work on fishery products. However, following authorization by the competent authority they may be used at the same time or other times for work on other foodstuffs.

4. Drinking water, within the meaning of Directive 80/778/EEC, or clean seawater must be used for all purposes. However, by way of an exception, non-drinking waster may be used for steam production, fire-fighting and the cooling of refrigeration equipment, provided that the pipes installed for the purpose preclude the use of such water for other purposes and present no risk of contamination of the products.

5. Detergents, disinfectants and similar substances must be approved by the competent authority and used in such a way that they do not have adverse effects on the machinery, equipment and products.

B. *General conditions of hygiene applicable to staff*

1. The highest possible standard of cleanliness is required of staff. More specifically:

(a) staff must wear suitable clean working clothes and headgear which completely encloses the hair. This applies particularly to persons handling exposed fishery products;

(b) staff assigned to the handling and preparation of fishery products must be required to wash their hand at least each time work is resumed; wounds to the hands must be covered by a waterproof dressing;

(c) smoking, spitting, eating and drinking in work and storage premises of fishery products must be prohibited.

2. The employer shall take all the requisite measures to prevent persons liable to contaminate fishery products from working on and handling them, until there is evidence that such persons can do so without risk.

When recruited, any person working on and handling fishery products shall be required to prove, by a medical certificate, that there is no impediment to such employment. The medical supervision of such a person shall be governed by the national legislation in force in the Member State concerned or in the case of third countries by specific guarantees to be fixed under the procedure set out in Article 15.

CHAPTER IV

SPECIAL CONDITIONS FOR HANDLING FISHERY PRODUCTS ON SHORE

I. Conditions for fresh products

1. Where chilled, unpackaged products are not dispatched, prepared or processed immediately after reaching the establishment, they must be stored or displayed under ice in the establishment s cold room. Re-icing must be carried out as often as is necessary; the ice used, with or without salt, must be made from drinking water or clean seawater and be stored under hygienic conditions in receptacles provided for the purpose; such receptacles must be kept clean and in a good state of repair. Prepacked fresh products must be chilled with ice or mechanical refrigeration plant creating similar temperature conditions.

2. If they are not carried out on board, operations such as heading and gutting must be carried out hygienically. The products must be washed thoroughly with drinking water or clean seawater immediately after such operations.

3. Operations such as filleting and slicing must be carried out in such a way as to avoid the contamination or spoilage of fillets and slices, and in a place other than that used for heading and gutting operations. Fillets and slices must not remain on work tables any longer than is necessary for their preparation. Fillets and slices to be sold fresh must be chilled as quickly as possible after preparation.

4. Guts and parts that may constitute a danger to public health must be separated from and removed from the vicinity of products intended for human consumption.

5. Containers used for the dispatch or storage of fresh fishery products must be designed in such a way as to ensure both their protection from contamination and their preservation under sufficiently hygienic conditions and, more particularly, they must provide adequate drainage of melt water.

6. Unless special facilities are provided for the continuous disposal of waste, the latter must be placed in leakproof, covered containers which are easy to clean and disinfect. Waste must not be allowed to accumulate in working areas. It must be removed either continuously or as soon as the containers are full and at least at the end of each working day in the containers or to the premises referred to in Chapter III, section I, paragraph 6 of this Annex. The containers, receptacles and/or premises set aside for waste must always be thoroughly cleaned and, if appropriate, disinfected after use. Waste stored there must not constitute a source of contamination for the establishment or of pollution of its surroundings.

II. Conditions for frozen products

1. Plants must have:

(a) freezing equipment sufficiently powerful to achieve a rapid reduction in the temperature so that the temperatures laid down to in this Directive can be obtained in the product;

(b) freezing equipment sufficiently powerful to keep products in storage rooms at a temperature not exceeding those laid down in this Directive, whatever the ambient temperature may be.

However, for technical reasons related to the method of freezing and to the handling of such products, for whole fish frozen in brine and intended for canning, higher temperatures than those laid down in this Directive are acceptable although they may not exceed − 9 °C.

2. Fresh products to be frozen or quick-frozen must comply with the requirements of section I of this Chapter.

3. Storage rooms must have a temperature recording device in a place where it can easily be read. The temperature sensor of the recorder must be located in the area furthest away from the cold source, i.e. where the temperature in the storage room is the highest.

Temperature charts must be available for inspection by the supervisory authorities at least during the period in which the products are stored.

III. Conditions for thawing products

Establishments that carry out thawing operations must comply with the following requirements:

1. fishery products must be thawed under hygienic conditions; their contamination must be avoided and there must be adequate drainage for any melt water produced.

During thawing, the temperature of the products must not increase excessively;

2. after thawing, fishery products must be handled in accordance with the requirements of this Directive. When they are prepared or processed, these operations must be carried out without delay. If they are put directly onto the market, particulars as to the thawed state of the fish must be clearly marked on the packaging in accordance with Article 5 (3) of Council Directive 79/112/EEC of 18 December 1978 on the approximation of the laws of the Member States relating to the labelling, presentation and advertising of foodstuffs (1).

IV. Conditions for processed products

1. Fresh, frozen and thawed products used for processing must comply with the requirements of sections I or II of this Chapter.

2. Where the processing treatment is carried out to inhibit the development of pathogenic micro-organisms, or if it is a significant factor in the preservation of the product, the treatment must be scientifically recognized by the law in force, or in the case of a treatment of products referred to in Chapter I Section 1 (b) and (c) of Directive 91/492/EEC which have not been relayed or purified, such treatment must be approved, in accordance with the procedure laid down in Article 15 of this Directive, within four months of receipt of a request from a Member State.

The person responsible for an establishment must keep a register of the processing carried out. Depending on the type of process employed, heating time and temperature, salt content, pH, water content, etc., must be monitored and controlled. Records must be kept at least for the expected storage life of the products and be available to the competent authority.

(1) OJ No L 33, 8. 2. 1979, p. 1. Directive last amended by Directive 91/72/EEC (OJ No L 42, 16. 1. 1991, p. 22).

3. For products which are preserved for a limited period by a treatment such as salting, smoking, drying or marinading, the appropriate conditions for storage must be clearly marked on the packaging, in accordance with Directive 79/112/EEC.

In addition, the following conditions shall be complied with..

4. *Canning*

In the case of fishery products which have been subjected to sterilization in hermetically sealed containers:

(a) the water used for the preparation of cans must be drinking water;

(b) the process used for the heat treatment must be appropriate, having regard to such major criteria as the heating time, temperature, filling, size of containers, etc., a record of which must be kept; the heat treatment must be capable of destroying or inactivating pathogenic organisms and the spores of pathogenic micro-organisms. The heating equipment must be fitted with devices for verifying whether the containers have in fact undergone appropriate heat treatment. Drinking water must be used to cool containers after heat treatment, without prejudice to the presence of any chemical additives used in accordance with good technological practice to prevent corrosion of the equipment and containers;

(c) further checks must be carried out at random by the manufacturer to ensure that the processed products have undergone appropriate heat treament, *viz.*:

— incubation tests: incubation must be carried out at 37 °C for seven days or at 35 °C for ten days, or at any other equivalent combination;

— microbiological examination of contents and containers in the establishment's laboratory or in another approved laboratory;

(d) samples must be taken of production each day at predetermined intervals, to ensure the efficacy of sealing. For that purpose, appropriate equipment must be available for the examination of cross-sections of the can-seams;

(e) checks are carried out in order to ensure that containers are not damaged;

(f) all containers which have undergone heat treatment under practically identical conditions must be given a batch identification mark, in accordance with Council Directive 89/396/EEC of 14 June 1989 on indications or marks identifying the lot to which a foodstuff belongs (1).

5. *Smoking*

Smoking must be carried out in separate premises or a special place equipped, if necessary, with a ventilation system to prevent the smoke and heat from the combustion from affecting other premises or places where fisnery products are prepared, processed or stored.

(a) Materials used to produce smoke for the smoking of fish must be stored away from the place of smoking and must be used in such a way that they do not contaminate the products.

(b) Materials used to produce smoke by burning wood that has been painted, varnished, glued or has undergone any chemical preservation treatment must be prohibited.

(c) After smoking, products must be cooled rapidly to the temperature required for their preservation before being packaged.

6. *Salting*

(a) Salting operations must take place in different premises and sufficiently removed from the premises where the other operations are carried out.

(b) Salt used in the treatment of fishery products must be clean and stored in such a way as to preclude contamination. It must not be re-used.

(c) Any container used for salting or brining must be constructed in such a way as to preclude contamination during the salting or brining process.

(d) Containers or areas used for salting or brining must be cleaned before use.

(1) OJ No L 186, 30. 6. 1989, p. 21.

7. *Cooked crustacean and molluscan shellfish products*

Crustaceans and molluscan shellfish must be cooked as follows:

(a) any cooking must be followed by rapid cooling. Water used for this purpose must be drinking water or clean seawater. If no other method of preservation is used, cooling must continue until the temperature approaching that of melting ice is reached;

(b) shelling or shucking must be carried out under hygienic conditions avoiding the contamination of the product. Where such operations are done by hand, workers must pay particular attention to the washing of their hands and all working surfaces must be cleaned thoroughly. If machines are used, they must be cleaned at frequent intervals and disinfected after each working day.

After shelling or shucking, cooked products must immediately be frozen or kept chilled at a temperature which will preclude the growth of pathogens, and be stored in appropriate premises;

(c) every manufacturer must carry out micro-biological checks on his production at regular intervals, complying with the standards to be fixed in accordance with Chapter V, Section 4 of this Annex.

8. *Mechanically recovered fish flesh*

The mechanical recovery of fish flesh must be carried out under the following conditions:

(a) mechanical recovery of gutted fish must take place without undue delay after filleting, using raw materials free of guts. Where whole fish are used, they must be gutted and washed beforehand;

(b) the machinery must be cleaned at frequent intervals and at least every two hours;

(c) after recovery, mechanically recovered flesh must be frozen as quickly as possible or incorporated in a product intended for freezing or stabilizing treatment.

V. Conditions concerning parasites

1. During production and before they are released for human consumption, fish and fish products must be subject to a visual inspection for the purpose of detecting and removing any parasites that are visible.

Fish or parts of fish which are obviously infested with parasites, and which are removed, must not be placed on the market for human consumption.

The detailed rules for this inspection shall be adopted in accordance with the procedure laid down in Article 15 of this Directive, on a proposal from the Commission to be submitted before 1 October 1992.

2. The fish and fish products referred to in point 3 which are to be consumed as they are must, in addition, be subjected to freezing at a temperature of not more than – 20 °C in all parts of the product for not less than 24 hours. Products subjected to this freezing process must be either raw or finished.

3. Fish and products subject to the conditions in point 2:

(a) fish to be consumed raw or almost raw, e.g. raw herring 'maatje';

(b) the following species, if they are to undergo a cold smoking process at which the internal temperature of the fish is less than 60 °C:

— herring,

— mackerel,

— sprat,

— (wild) Atlantic and Pacific salmon;

(c) marinated and/or salted herring where this process is insufficient to destroy the larvae of nematodes.

This list may be amended, in the light of scientific data, in accordance with the procedure laid down in Article 15 of this Directive. In accordance with the same procedure, criteria will be laid down which must enable the processes which are deemed sufficient or insufficient to destroy nematodes to be defined.

4. Manufacturers must ensure that fish and fish products listed in point 3 or the raw materials for use in their manufacture are subjected to the treatment described in point 2, prior to their release for consumption.

5. The fishery products listed in point 3 must, when they are placed on the market, be accompanied by a document from the manufacturer stating the type of process they have undergone.

CHAPTER V

HEALTH CONTROL AND MONITORING OF PRODUCTION CONDITIONS

I. General monitoring

Arrangements for checking and monitoring must be made by the competent authorities in order to establish whether the requirements laid down in this Directive are complied with.

Such arrangements will include, in particular:

1. a check on the fishing vessels, on the understanding that such a check may be carried out during the stay in port;

2. a check on the conditions of landing and first sale;

3. an inspection at regular intervals of establishments to check, in particular:

   (a) whether the conditions for approval are still fulfilled;

   (b) whether the fishery products are handled correctly;

   (c) the cleanliness of the premises, facilities and instruments and staff hygiene;

   (d) whether identification marks are put on correctly;

4. an inspection of the wholesale and auction markets;

5. a check on storage and transport conditions.

II. Special checks

1. *Organoleptic checks*

Without prejudice to the derogations provided for by Council Regulation (EEC) No 103/76 of 19 January 1976 laying down common marketing standards for certain fresh or chilled fish (1), each batch of fishery products must be submitted for inspection by the competent authority at the time of landing or before first sale to check whether they are fit for human consumption. This inspection comprises an organoleptic check carried out by sampling.

Fishery products complying, as far as the freshness criteria are concerned, with the common marketing standards already laid down pursuant to Article 2 of Regulation (EEC) No 3796/81 are considered to fulfil the organoleptic requirements necessary for compliance with the provisions of this Directive.

The Commission may, where necessary, in accordance with the procedure referred to in Article 15 of this Directive, lay down specific organoleptic requirements for fishery products not harmonized under Regulation (EEC) No 3796/81.

The organoleptic examination must be repeated after the first sale of fishery products, if it is found that the requirements of this Directive have not been complied with or when considered necessary. After the first sale, fishery products must at least comply with the minimum freshness requirements of the aforementioned Regulation.

If the organoleptic examination reveals that the fishery products are not fit for human consumption, measures must be taken to withdraw them from the market and denature in such a way that they cannot be re-used for human consumption.

If the organoleptic examination reveals any doubt as to the freshness of the fishery products, use may be made of chemical checks or microbiological analyses.

2. *Parasite checks*

Before they are released for human consumption, fish and fish products must be subject to a visual inspection, by way of sample, for the purpose of detecting any parasites that are visible.

(1) OJ No L 20, 28. 1. 1976, p. 29. Regulation last amended by Regulation (EEC) No 33/89 (OJ No L 5, 7. 1. 1989, p. 18).

Fish or parts of fish which are obviously infested with parasites, and which are removed, must not be placed on the market for human consumption.

The detailed rules for this inspection shall be established in accordance with the procedure laid down in Article 15.

3. *Chemicals checks*

A. Samples must be taken and subjected to laboratory analysis for the control of the following parameters:

(a) TVB-N (Total Volatile Basic Nitrogen) and TMA-N (Trimethylamine-Nitrogen)

The levels of these parameters must be specified for each category of species in accordance with the procedure laid down in Article 15 of this Directive.

(b) Histamine

Nine samples must be taken from each batch. These must fulfil the following requirements:

— the mean value must not exceed 100 ppm;

— two samples may have a value of more than 100 ppm but less than 200 ppm;

— no sample may have a value exceeding 200 ppm.

These limits apply only to fish species of the following families: Scombridae and Clupeidae. However, fish belonging to these families which have undergone enzyme ripening treatment in brine may have higher histamine levels but not more than twice the above values. Examinations must be carried out in accordance with reliable, scientifically recognized methods, such as high-performance liquid chromatography (HPLC).

B. Contaminants present in the aquatic environment

Without prejudice to the Community rules concerning water protection and management, and in particular those concerning pollution of the aquatic environment, fishery products must not contain in their edible parts contaminants present in the aquatic environment such as heavy metals and organochlorinated substances at such a level that the calculated dietary intake exceeds the acceptable daily or weekly intake for humans.

A monitoring system must be established by the Member States to check the level of contamination of fishery products.

C. In accordance with the procedure laid down in Article 15 of this Directive, the following shall be decided on by not later than 31 December 1992:

(a) the methods of analysis to be used to check the chemical parameters, as well as the sampling plans;

(b) the acceptable levels for the chemical parameters.

4. *Microbiological analyses*

In accordance with the procedure laid down in Article 15 of this Directive, microbiological criteria, including sampling plans and methods of analysis, may be laid down when there is a need to protect public health. The Commission will to this end submit appropriate proposals for measures by 1 October 1992.

CHAPTER VI

PACKAGING

1. Packaging must be carried out under satisfactory conditions of hygiene, to preclude contamination of the fishery products.

2. Packaging materials and products liable to enter into contact with fishery products must comply with all the rules of hygiene, and in particular:

— they must not be such as to impair the organoleptic characteristics of the fishery products;

— they must not be capable of transmitting to the fishery products substances harmful to human health;

— they must be strong enough to protect the fishery products adequately.

3. With the exception of certain containers made of impervious, smooth and corrosion-resistant material which are easy to clean and disinfect, which may be re-used after cleaning and disinfecting, packaging materials may not be re-used. Packaging materials used for fresh products held under ice must provide adequate drainage for melt water.

4. Unused packaging materials must be stored in premises away from the production area and be protected from dust and contamination.

CHAPTER VII

IDENTIFICATION MARKS

Without prejudice to the requirements laid down in Directive 79/112/EEC, it must be possible to trace for inspection purposes the establishment of dispatch of consignments of fishery products, by means of either labelling or the accompanying documents. For that purpose, the following information must appear on the packaging or in the accompanying documents:

— the country of dispatch;

— identification of the establishment by its official approval number or, in the case of separate registering of auction or wholesale markets as laid down in Article 7 (1), third subparagraph of this Directive, the registration number of the auction or wholesale market.

CHAPTER VIII

STORAGE AND TRANSPORT

1. Fishery products must, during storage and transport, be kept at the temperatures laid down in this Directive and in particular:

   — fresh or thawed fishery products and cooked and chilled crustacean and molluscan shellfish products must be kept at the temperature of melting ice; .

   — frozen fishery products, with the exception of frozen fish in brine intended for the manufacture of canned foods, must be kept at an even temperature of – 18 °C or less in all parts of the product, allowing for the possibility of brief upward fluctuations of not more than 3 °C, during transport;

   — processed products must be kept at the temperatures specified by the manufacturer, when the circumstances so require, prescribed in accordance with the procedure laid down in Article 15 of this Directive.

2. Where frozen fishery products are transported from a cold-storage plant to an approved establishment to be thawed on arrival for the purposes of preparation and/or processing and where the distance to be covered is short, not exceeding 50 km or one hour's journey, the competent authority may grant a derogation from the conditions laid down in point 1, second indent.

3. Products may not be stored or transported with other products which may contaminate them or affect their hygiene, unless they are packaged in such a way as to provide satisfactory protection.

4. Vehicles used for the transport of fishery products must be constructed and equipped in such a way that the temperatures laid down in this Directive can be maintained throughout the period of transport. If ice is used to chill the products, adequate drainage must be provided in order to ensure that water from melted ice does not stay in contact with the products. The inside surfaces of the means of transport must be finished in such a way that they do not adversely affect the fishery products. They must be smooth and easy to clean and disinfect.

5. Means of transport used for fishery products may not be used for transporting other products likely to impair or contaminate fishery products, except where the fishery products can be guaranteed uncontaminated as a result of such transport being thoroughly cleaned and disinfected.

6. Fishery products may not be transported in a vehicle or container which is not clean or which should have been disinfected.

7. The transport conditions of fishery products to be placed on the market alive must not adversely affect the products.

CHAPTER IX

POINTS OF ANNEX I WHICH MAY BE SUBJECT TO DEROGATIONS AND POSSIBLE CONDITIONS APPLICABLE IN THE CASE OF DEROGATIONS

Re Chapter I Part I of the Annex

1. *Point 1 (a)*

   provided products are sheltered from the sun and the elements and from any source of dirt or contamination.

2. *Point 1 (c)*

   provided any contamination of the products is prevented.

3. *Point 1 (d), first sentence*

   provided the finished products are stored on board at the required temperature.

4. *Point 1 (g), last sentence*

   provided products cannot be contaminated by waste water, waste or engine coolant.

5. *Point 1 (h)*

   provided staff handling fishery products can wash their hands after using the toilet.

6. *Point 2 (a)*

   provided floors are properly cleaned and disinfected.

7. *Point 2 (b), (c) and (d)*

8. *Point 2 (g) on taps and towels*

9. *Point 3*

   provided equipment and tools are well maintained.

Re Chapter II of the Annex

10. *Point 3 (a)*

    provided the walls are kept clean.

11. *Point 3 (b)*

    provided the flooring is kept clean after every sale.

12. *Point 3 (c), first sentence*

13. *Point 3 (e): vehicles emitting exhaust fumes*

    provided products contaminated by exhaust fumes are withdrawn from the market.

14. *Point 3 (j)*

    provided that products which are not fit for human consumption cannot contaminate or be mixed with fishery products.

15. *Point 3 (k)*

16. *Point 7*

insofar as it refers to point 3 of the same Chapter and point 10 of Chapter III, section I.

Re Chapter III Part I of the Annex

17. *Point 1*

provided finished products cannot be contaminated by raw materials or waste.

18. *Point 2 (a)*

provided the flooring is cleaned and disinfected accordingly.

19. *Point 2 (b)*

provided the walls are kept clean.

20. *Point 2 (c)*

provided the ceiling is not a source of contamination.

21. *Point 2 (d)*

22. *Point 2 (e)*

provided products cannot be spoilt or contaminated by the steam.

23. *Point 2 (g)*

provided there are facilities available for staff to wash their hands.

24. *Point 3*

25. *Point 5*

insofar as it relates to corrosion-resistant materials provided instruments and working equipment are kept clean.

26. *Point 6*

provided products cannot be contaminated by waste or leakage therefrom.

27. *Point 10*

Re Chapter IV of the Annex

28. *Part I, point 1*

in respect of the requirement for products being held over to be put in the establishment's cold room provided the products are re-iced as often as necessary during a period not in excess of 12 hours or that a nearby cold room not belonging to the establishment can be used.

29. *Part I, point 6*

in respect of the requirement for waste to be put in leakproof covered containers provided products cannot be contaminated by waste or leakage therefrom.

30. *Part IV, point 5, first paragraph*

provided that every precaution is taken to prevent fishery products that are being prepared or stored from being affected by the smoke.

31. *Part IV, point 6 (a)*

provided fishery products that are being prepared or stored are not affected by salting operations.

II

(Acts whose publication is not obligatory)

COUNCIL

COUNCIL DIRECTIVE

of 15 July 1991

laying down the health conditions for the production and the placing on the market of live bivalve molluscs

(91/492/EEC)

THE COUNCIL OF THE EUROPEAN COMMUNITIES,

Having regard to the Treaty establishing the European Economic Community, and in particular Article 43 thereof,

Having regard to the proposal from the Commission (1),

Having regard to the opinion of the European Parliament (2),

Having regard to the opinion of the Economic and Social Committee (3),

Whereas, with a view to achieving the internal market and more especially to ensure the smooth operation of the common organization of the market in fishery products established by Regulation (EEC) No 3796/81 (4) as last amended by Regulation (EEC) No 2886/89 (5), it is essential that the placing on the market of live bivalve molluscs should no longer be hindered by disparities existing in the Member States in respect of health requirements; whereas this will enable production and placing on the market to be better harmonized and bring about competition on equal terms while ensuring quality products for the consumer.

Whereas Council Directive 79/923/EEC of 30 October 1979 on the quality required of shellfish waters (6) lays down that it is necessary to establish the health requirements to be observed for shellfish products;

Whereas these requirements should be laid down for all stages during harvesting, handling, storage, transport and distribution of live bivalve molluscs in order to safeguard the public health of consumers; whereas these requirements shall apply equally to echinoderms, tunicates and marine gastropods;

Whereas it is important, should a health problem occur after the placing on the market of live bivalve molluscs to be able to trace back the establishment of dispatch and the harvesting area of origin; whereas it is therefore necessary to introduce a registration and labelling system which will enable the route of a batch after harvesting to be followed;

Whereas it is important that the public health standards for the final product must be specified; whereas, however, scientific and technological knowledge is not always advanced enough to lay down definitive solutions for certain health problems and whereas it is therefore necessary, in order to guarantee optimal protection of public health, to set up a Community system to ensure rapid adoption and where necessary reinforcement of the health standards to safeguard human health from virus contamination or other hazards;

Whereas live bivalve molluscs obtained from harvesting areas which do not permit direct, safe consumption may be rendered safe by submitting them to a purification process or by relaying in clean water over a relatively long period; whereas it is therefore necessary to define production areas from which molluscs can bet gathered for direct human consumption, or from which they have to be purified or relayed;

Whereas it is primarily the responsibility of the producers to ensure that the bivalve molluscs are produced and placed on the market in compliance with the health requirements prescribed; whereas the competent authorities must, by

(1) OJ No C 84, 2. 4. 1990, p. 29.
(2) OJ No C 183, 15. 7. 1991.
(3) OJ No C 332, 31. 12. 1990, p. 1.
(4) OJ No L 379, 31. 12. 1981, p. 1.
(5) OJ No L 282, 2. 10. 1989, p. 1.
(6) OJ No L 281, 10. 11. 1979, p. 47.

carrying out checks and inspections, ensure that producers comply with those requirements; whereas the competent authorities must in particular submit harvesting areas to a regular control to ensure that molluscs from these harvesting areas do not contain microorganisms and toxic substances in quantities which are considered to be dangerous to human health;

Whereas control measures organized on a Community level must be introduced to guarantee the uniform application in all Member States of the standards laid down in this Directive;

Whereas the rules, principles and safeguard measures established by Council Directive 90/675/EEC of 10 December 1990 laying down the principles governing the organization of veterinary checks on products entering the Community from third countries (1), should apply to the case in question;

Whereas in the context of trade between the Member States, the rules laid down in Council Directive 89/662/EEC of 11 December 1989 concerning veterinary checks in intra-Community trade, with a view to the completion of the internal market (2) as amended by Directive 90/675/EEC should also be applied;

Whereas live bivalve molluscs produced in a third country and intended to be placed on the market in the Community must not qualify for more favourable conditions than those applied in the Community; whereas provision must be made for a Community procedure for checking the conditions in third countries of production and of the placing on the market, in order to allow the Community to apply a common import system based on conditions of equivalence;

Whereas, so that account may be taken of particular circumstances, derogations should be granted to certain establishments already operating before 1 January 1993 so as to allow them to adapt to all the requirements laid down in this Directive;

Whereas, in the case of living animals that are edible whilst they are alive, a derogation should be made, with regard to the durability date, to the rules of Council Directive 79/112/EEC of 18 December 1978 on the approximation of the laws of the Member States relating to the labelling, presentation and advertising of foodstuffs for sale (3) as last amended by Directive 91/72/EEC (4);

Whereas provision should be made for the possibility of adopting transitional measures in order to cover the absence of certain implementing rules;

Whereas the Commission should be entrusted with the task of adopting certain measures for implementing this Directive; whereas to that end, procedures should be laid down introducing close and effective cooperation between the Commission and the Member States within the Standing Veterinary Committee,

(1) OJ No L 373, 31. 12. 1990, p. 1.
(2) OJ No L 395, 30. 12. 1989, p. 13.
(3) OJ No L 33, 8. 2. 1979, p. 1.
(4) OJ No L 42, 16. 1. 1991, p. 27.

HAS ADOPTED THIS DIRECTIVE:

CHAPTER I

General provisions

Article 1

This Directive lays down health conditions for the production and placing on the market of live bivalve molluscs which are intended for immediate human consumption or for further processing before consumption.

With the exception of the provisions on purification, this Directive applies to echinoderms, tunicates and marine gastropods.

Article 2

For the purposes of this Directive, the following definitions shall apply:

1. *'bivalve molluscs'* means filter-feeding lamellibranch molluscs;

2. *'marine biotoxins'* means poisonous substances accumulated by bivalve molluscs feeding on plankton containing toxin;

3. *'clean sea water'* means sea water or brackish water which is to be used under the conditions laid down in this Directive and which is free from microbiological contamination and toxic and objectionable substances occurring naturally or after discharge in the environment such as those listed in the Annex to Directive 79/923/EEC, in such quantities as may adversely affect the health quality of bivalve molluscs or to impair their taste;

4. *'competent authority'* means the central authority of a Member State competent to carry out veterinary checks or any authority to which it has delegated that competence;

5. *'conditioning'* means the storage of live bivalve molluscs, whose quality does not indicate the need for relaying or treatment in a purification plant, in tanks or any other installation containing clean sea water or in natural sites to remove sand, mud or slime;

6. *'gatherer'* means any natural or legal person who collects live bivalve molluscs by any means from a harvesting area for the purpose of handling and placing on the market;

7. *'production area'* means any sea, estuarine or lagoon area containing natural deposits of bivalve molluscs or sites used for cultivation of bivalve molluscs from which live bivalve molluscs are taken;

8. *'relaying area'* means any sea, estuarine or lagoon area approved by the competent authority, with boundaries clearly marked and indicated by buoys, posts or any other fixed means, and used exclusively for the natural purification of live bivalve molluscs;

— representative numbers of samples for laboratory examination are regularly taken and analysed in order to establish an historical record on the basis of the areas where batches come from and of the health quality of the live bivalve molluscs both before and after handling at a dispatch centre or purification centre.

— a register is kept for the permanent record of the results of the various checks and kept for presentation to the competent authority.

Article 5

1. (a) The competent authority shall approve dispatch centres and purification centres once it is satisfied that they meet the requirements of this Directive. The competent authority shall take the necessary measures if the requirements cease to be met. In so doing, it shall take account of, in particular, the outcome of any check carried out in accordance with Article 6 (1).

However, subject to the express condition that live molluscs coming from such centres meet the hygiene standards set by this Directive, Member States may, for the requirements relating to equipment and structures laid down in Chapter IV of the Annex, to be specified before 1 October 1991 in accordance with the procedure laid down in Article 12, grant to dispatch and purification centres, a further period expiring on 31 December 1995 within which to comply with the conditions of the approval set out in the abovementioned Chapter. Such derogations may be granted only to establishments, already operating on 31 December 1991, which have, before 1 July 1992, submitted a duly substantiated application for derogation to the competent national authority. This application must be accompanied by a work plan and programme indicating the period within which it would be possible for the establishments to comply with the requirements in question. Where financial assistance is requested from the Community, only requests in respect of projects complying with the requirements of this Directive can be accepted.

The competent authority shall draw up a list of approved dispatch centres and purification centres, each of which shall have an official number.

The list of approved dispatch centres and purification centres, and any subsequent amendments thereto, must be communicated by each Member State to the Commission, which shall pass such information on to the other Member States.

(b) The inspection and monitoring of these centres shall be carried out regularly under the responsibility of the competent authority, which shall have free access to all parts of the centres, in order to ensure compliance with the provisions of this Directive.

If such inspections and monitoring reveal that the requirements of this Directive are not being met, the competent authority shall take appropriate action.

2. (a) The competent authority shall establish a list of production and relaying areas, with an indication of their location and boundaries, from which live bivalve molluscs may be taken in accordance with the requirements of this Directive and, in particular, with Chapter I of the Annex.

This list must be communicated to those affected by this Directive, such as gatherers and operators of purification centres and dispatch centres.

(b) The monitoring of the production and relaying areas shall be carried out under the responsibility of the competent authority in accordance with the requirements of this Directive.

If such monitoring reveals that the requirements of this Directive are no longer being met, the competent authority shall close the production or relaying area concerned until the situation has been restored to normal.

3. The competent authority may prohibit any production and harvesting of bivalve molluscs in areas considered unsuitable for these activities for health reasons.

Article 6

1. Experts from the Commission may, in cooperation with the competent authorities of the Member States, make on-the-spot checks insofar as is necessary to ensure the uniform application of this Directive. They may, in particular, check whether centres, production and relaying areas are in effect complying with the requirements of this Directive. A Member State in whose territory a check is being carried out shall give all necessary assistance to the experts in carrying out their duties. The Commission shall inform the Member States of the results of such checks.

2. The arrangements for implementing paragraph 1 shall be adopted in accordance with the procedure laid down in Article 12.

3. The Commission, may draw up recommendations containing guidelines on good manufacturing practices applicable at the different stages of production and placing on the market.

Article 7

1. The rules laid down in Directive 89/662/EEC as regards live bivalve molluscs, echinoderms, tunicates and marine gastropods intended for human consumption, shall apply, in particular as regards the organization of and the action to be taken following the checks to be carried out by the Member State of destination, and the safeguard measures to be implemented.

9. *'dispatch centre'* means any approved on-shore or off-shore installation for the reception, conditioning, washing, cleaning, grading and wrapping of live bivalve molluscs fit for human consumption;

10. *'purification centre'* means an approved establishment with tanks fed by naturally clean sea water or sea water that has been cleaned by appropriate treatment, in which live bivalve molluscs are placed for the time necessary to remove microbiological contamination, so making them fit for human consumption;

11. *'relaying'* means an operation whereby live bivalve molluscs are transferred to approved sea or lagoon areas or approved estuarine areas under the supervision of the competent authority for the time necessary to remove contamination. This does not include the specific operation of transferring bivalve molluscs to areas more suitable for further growth or fattening;

12. *'means of transport'* means those parts set aside for goods in automobile vehicles, rail vehicles and aircraft, the holds of vessels and containers for transport by land, sea or air;

13. *'wrapping'* means an operation whereby live bivalve molluscs are placed in packaging material adequate for the purpose;

14. *'consignment'* means a quantity of live bivalve molluscs handled in a dispatch centre or treated in a purification centre and subsequently intended for one or more customers;

15. *'batch'* means a quantity of live bivalve molluscs collected from a production area and subsequently intended for delivery to an approved dispatch centre, purification centre, relaying area or processing plant as appropriate;

16. *'placing on the market'* means the holding or displaying for sale, offering for sale, selling, delivering or any other form of placing on the market of live bivalve molluscs for human consumption either raw or for the purpose of processing in the Community, excluding the direct transfer on the local market in small quantities by the coastal fisherman to the retailer or the consumer which must be subject to the health checks laid down by national rules for checking on retail business;

17. *'importation'* means the introduction of live bivalve molluscs into the territory of the Community from third countries;

18. *'faecal coliform'* means facultative, aerobic, gram-negative, non-sporeforming, cytochrome oxidase negative, rod-shaped bacteria that are able to ferment lactose with gas production in the presence of bile salts, or other surface active agents with similar growth-inhibiting properties, at 44 °C ± 0,2 °C within 24 hours at least;

19. *'E. coli'* means faecal coliforms which also form indole from tryptophan at 44 °C ± 0,2 °C within 24 hours.

CHAPTER II

Provisions for Community production

Article 3

1. The placing on the market of live bivalve molluscs for immediate human consumption shall be subject to the following conditions:

(a) they must originate from production areas which comply with the requirements laid down in Chapter I of the Annex; however, in the case of pectinidae, this provision shall apply only to aquaculture products as defined in Article 2 (2) of Council Directive 91/493/EEC of 22 July 1991 laying down the health conditions for the production and placing on the market of fishery products [1];

(b) they must have been harvested and transported from production area to a dispatch centre, purification centre, relaying area or processing plant under the conditions laid down in Chapter II of the Annex;

(c) where provided for in this Directive, they must have been relaid in suitable areas approved for that purpose and complying with the conditions laid down in Chapter III of the Annex;

(d) they must have been handled hygienically, and where appropriate, they must have been purified in establishments approved for that purpose and complying with the requirements of Chaper IV of the Annex;

(e) they must comply with the criteria set out in Chapter V of the Annex;

(f) health controls must have been carried out in accordance with Chapter VI of the Annex;

(g) they must have been appropriately wrapped in accordance with Chapter VII of the Annex;

(h) they must have been stored and transported under satisfactory conditions of hygiene in accordance with Chapters VIII and IX of the Annex;

(i) they must bear a health mark as provided for in Chapter X of the Annex.

2. Live bivalve molluscs intended for further processing must comply with the relevant requirements of paragraph 1 and be processed in accordance with the requirements of Council Directive 91/493/EEC.

Article 4

Member States shall ensure that persons handling live bivalve molluscs during their production and placing on the market shall adopt all measures necessary to comply with the requirements of this Directive.

Persons responsible for dispatch and purification centres shall in particular ensure that:

[1] See page 15 of this Official Journal.

of live bivalve molluscs from third countries shall be at least equivalent to those governing the production and placing on the market of Community products.

Article 10

The rules and principles laid down in Directive 90/675/EEC shall apply, with particular reference to the organization of and follow up to the inspections to be carried out by the Member States and the safeguard measures to be implemented.

Without prejudice to compliance with the rule and principles referred to in the first subparagraph of this Article and pending implementation of the decisions provided for in Article 8 (3) and Article 30 of Directive 90/675/EEC, the relevant national rules for applying Article 8 (1) and (2) of the said Directive shall continue to apply.

CHAPTER IV

Final provisions

Article 11

The chapters of the Annex may be amended by the Council, acting by a qualified majority on a proposal from the Commission.

The Commission shall, before 1 January 1994, submit to the Council, after receiving the opinion of the Scientific Veterinary Committee, a report on Chapters I and V of the Annex, accompanied by any proposed amendments to those Chapters.

Article 12

1. Where the procedure laid down in this Article is to be followed, the Chairman shall refer the matter to the Standing Veterinary Committee hereafter referred to as the committee, either on his own initiative or at the request of a Member State.

2. The representative of the Commission shall submit to the committee a draft of the measures to be taken. The committee shall deliver its opinion on the draft within a time limit which the chairman may lay down according to the urgency of the matter. The opinion shall be delivered by the majority laid down in Article 148 (2) of the Treaty in the case of decisions which the Council is required to adopt on a proposal from the Commission. The votes of the representatives of the Member States within the committee shall be weighted in the manner set out in that Article. The chairman shall not vote.

3. (a) The Commission shall adopt the measures envisaged if they are in accordance with the opinion of the committee.

(b) If the measures envisaged are not in accordance with the opinion of the committee, or if no opinion is delivered, the Commission shall, without delay, submit to the Council a proposal relating to the measures to be taken. The Council shall act by a qualified majority.

If, on the expiry of a period of three months from the date of referral to the Council, the Council has not acted, the proposed measures shall be adopted by the Commission save where the Council has decided against the said measures by a simple majority.

Article 13

In order to take into account the possible failure to take a decision on the detailed rules for applying this Directive by 1 January 1993, necessary transitional measures may be adopted in accordance with the procedure laid down in Article 12 for a period of two years.

Article 14

The Commission shall, after consulting the Member States, submit, before 1 July 1992, a report to the Council on the minimum requirements to be met with regard to structure and equipment by small dispatch centres or by small establishments ensuring distribution on the local market and situated in areas subject to particular constraints with respect to their supply, possibly accompanied by proposals, on which the Council will take a decision, acting in accordance with the voting procedure laid down in Article 43 of the Treaty, before 31 December 1992.

The provisions of this Directive shall be re-examined before 1 January 1998 by the Council, acting on a Commission proposal, in the light of the experience gained.

Article 15

The Member States shall bring into force the laws, regulations and administrative provisions necessary to comply with this Directive before 1 January 1993. They shall notify the Commission thereof.

When Member States adopt these measures, they shall contain a reference to this Directive or shall be accompanied by such reference on the occasion of their official publication. The methods of making such a reference shall be laid down by the Member States.

Article 16

This Directive is addressed to the Member States.

Done at Brussels, 15 July 1991.

For the Council

The President

P. BUKMAN

2. Directive 89/662/EEC shall be amended as follows:

(a) In Annex A, the following indent shall be added:

'— Council Directive 91/492/EEC of 15 July 1991 laying down the health conditions for the production and placing on the market of live bivalve molluscs, (OJ No L 268, 24. 9. 1991, p. 1.);

(b) in Annex B, the following indent shall be deleted:

'— live bivalve molluscs intended for human consumption'.

CHAPTER III

Imports from third countries

Article 8

Provisions applied to imports of live bivalve molluscs from third countries shall be at least equivalent to those governing the production and placing on the market of Community products.

Article 9

In order to ensure the uniform application of the requirement imposed in Article 8, the following procedure shall apply:

1. inspections shall be carried out on the spot by experts from the Commission and the Member States to verify whether the conditions of production and placing on the market can be considered as being equivalent to those applied in the Community.

   The experts from the Member States who are to be entrusted with these inspections shall be appointed by the Commission, acting on a proposal from the Member States.

   These inspections shall be made on behalf of the Community, which shall bear the cost of any expenditure in this connection.

   The frequency and the procedure for these inspections shall be determined in accordance with the procedure laid down in Article 12;

2. in deciding whether the conditions of production and placing on the market of live bivalve molluscs in a third country can be deemed equivalent to those of the Community, particular account shall be taken of:

   (a) the legislation of the third country;

   (b) the organization of the competent authority of the third country and of its inspection services, the powers of such services and the supervision to which they are subject, as well as their facilities for monitoring the implementation of their legislation in force;

   (c) the actual health conditions during the production and placing on the market of live bivalve molluscs and in particular the monitoring of production areas in relation to microbiological and environmental contamination, and to the presence of marine biotoxins;

   (d) the regularity and the rapidity of the information provided by the third country on the presence of plankton containing toxin in the production areas and, in particular, of species not occurring in Community waters, and risks that such presence may signify for the Community;

   (e) the assurances which a third country can give on the compliance with the standards laid down in Chapter V of the Annex;

3. the Commission, following the procedure laid down in Article 12, shall decide on:

   (a) the list of third countries fulfilling the conditions of equivalence referred to in paragraph 2;

   (b) for each third country, the specific conditions for the importation of live bivalve molluscs. These conditions must include:

      (i) the procedure for obtaining a health certificate which must accompany consignments when forwarded to the Community;

      (ii) the demarcation of the production areas from which live bivalve molluscs may be harvested and imported;

      (iii) the obligation to notify the Community of any possible change in the approval of production areas;

      (iv) any purification after arrival in the territory of the Community;

   (c) a list of establishments from which the importation of live bivalve molluscs is authorized. For this purpose, one or more lists of such establishments shall be established. An establishment may not appear on a list unless it is officially approved by the competent authority of the third country exporting to the Community. Such approval shall be subject to observance of the following requirements:

      — compliance with requirements equivalent to those laid down in this Directive,

      — monitoring by an official inspection service of the third country;

4. the decisions referred to in paragraph 3 may be amended in accordance with the procedure laid down in Article 12.

   These decisions and the amendments thereto shall be published in the *Official Journal of the European Communities*, L series;

5. pending the decisions referred to in paragraph 3, the conditions which Member States shall apply to imports

— the shellfish species and quantity indicated in as precise detail as is practicable,

— the approval number and place of destination for wrapping, relaying, purification or processing.

The registration documents must be numbered permanently in sequence. The competent authority must keep a register indicating numbers of registration documents together with the names of the persons collecting live bivalve molluscs and to whom the documents were issued. The registration document for each batch of live bivalve molluscs must be date-stamped upon delivery of a batch to a dispatch centre, purification centre, relaying area or processing plant and must be kept by operators of such centres, areas or establishments for at least 60 days.

However, if gathering is carried out by the same staff operating the dispatch centre, purification centre, relaying area or processing plant of destination, the registration document may be replaced by a permanent transport authorization granted by the competent authority.

7. If a production or relaying area is closed temporarily, the competent authority must refrain from issuing registration documents for that area and immediately suspend the validity of all registration documents already issued.

CHAPTER III

CONDITIONS FOR RELAYING LIVE BIVALVE MOLLUSCS

The following conditions must be met:

1. live bivalve molluscs must be gathered and transported in accordance with the requirements of Chapter II of this Annex;

2. techniques for handling live bivalve molluscs intended for relaying must permit the resumption of filter-feeding activity after immersion in natural waters;

3. live bivalve molluscs must not be relaid at a density which does not permit purification;

4. live bivalve molluscs must be immersed in seawater at the relaying area for an appropriate period which must exceed the time taken for levels of faecal bacteria to become reduced to the levels permitted by this Directive taking account of the fact that the standards of Chapter V of this Annex must be met;

5. the minimum water temperature for effective relaying must, where necessary, be determined and announced by the competent authority for each species of live bivalve mollusc and approved relaying area;

6. areas for relaying live bivalve molluscs must be approved by the competent authority. The boundaries of the sites must be clearly identified by buoys, poles or other fixed means; there must be a minimum distance of 300 metres between relaying areas, and also between relaying areas and production areas;

7. sites within a relaying area must be well separated to prevent mixing of batches; the 'all in, all out' system must be used, so that a new batch cannot be brought in before the whole of the previous batch has been removed;

8. permanent records of the source of live bivalve molluscs, relaying periods, relaying areas and subsequent destination of the batch after relaying must be kept by the operators of relaying areas for inspection by the competent authority;

9. after harvesting from the relaying area, batches must, during transport from the relaying area to the approved dispatch centre, purification centre or processing plant, be accompanied by the registration document referred to in Chapter II, section 6 of this Annex, except in the case where the same staff operates both the relaying area and the dispatch centre, purification centre or processing plant.

CHAPTER IV

CONDITIONS FOR THE APPROVAL OF DISPATCH OR PURIFICATION CENTRES

I. **General conditions relating to premises and equipment**

Centres must not be located in areas which are close to objectionable odours, smoke, dust and other contaminants. The location must not be subject to flooding by ordinary high tides or run-off from surrounding areas.

ANNEX

CHAPTER I

CONDITIONS FOR PRODUCTION AREAS

1. The location and the boundaries of production areas must be fixed by the competent authority in such a way as to identify the areas from which live bivalve molluscs:

   (a) can be collected for direct human consumption. Live bivalve molluscs taken from these areas must meet the requirements set out in Chapter V of this Directive;

   (b) can be collected but only placed on the market for human consumption after treatment in a purification centre, after relaying. Live bivalve molluscs from these areas must not exceed the limits of a five-tube, three-dilution MPN-test of 6 000 faecal coliforms per 100 g of flesh or 4 600 *E. Coli* per 100 g of flesh in 90 % of samples.

   After purification or relaying, all the requirements set out in Chapter V of this Annex must be met;

   (c) can be collected but placed on the market only after relaying over a long period (at least two months), whether or not combined with purification, or after intensive purification for a period to be fixed in accordance with the procedure provided for in Article 12 of this Directive, so as to meet the requirements under (a). Live bivalve molluscs from these areas must not exceed the limits of a five-tube, three-dilution MPN-test of 60 000 faecal coliforms per 100 g of flesh.

2. Any change in the demarcation of production areas and the temporary or definitive closure thereof must be immediately announced by the competent authority to those affected by this Directive and in particular to producers and operators of purification and dispatch centres.

CHAPTER II

REQUIREMENTS FOR HARVESTING AND TRANSPORTATION OF BATCHES TO A DISPATCH OR PURIFICATION CENTRE, RELAYING AREA OR PROCESSING PLANT

1. Harvesting techniques must not cause excessive damage to the shells or tissues of live bivalve molluscs.

2. Live bivalve molluscs must be adequately protected from crushing, abrasion or vibration after harvesting and must not be exposed to extremes of hot or cold temperature.

3. Techniques for harvesting, transporting, landing and handling live bivalve molluscs must not result in additional contamination of the product, nor in a significant reduction in the quality of the product, nor in any changes significantly affecting their ability to be treated by purification, processing or relaying.

4. Live bivalve molluscs must not be re-immersed in water which could cause additional contamination between harvesting and landing.

5. The means of transport used for transporting live bivalve molluscs must be used under conditions which protect the latter from additional contamination and crushing of shells. They must permit adequate drainage and cleaning.

   In the event of bulk transport over long distances of live bivalve molluscs to a dispatch centre, purification centre, relaying area or processing plant, the means of transport must be equipped in such a way as to ensure the best survival conditions possible, and in particular must comply with the requirements laid down in Chapter IX, Section 2 of this Annex.

6. A registration document for the identification of batches of live bivalve molluscs during transport from the production area to a dispatch centre, purification centre, relaying area or processing plant is issued by the competent authority upon request by the gatherer. For each batch, the gatherer must complete legibly and indelibly the relevant sections of the registration document which must contain the following information:

   — the gatherer's identity and signature,

   — the date of harvesting,

   — the location of the production area in as precise detail as is practicable,

2. live bivalve molluscs must be washed free of mud with pressurized clean sea water or potable water before purification. The initial washing may also be carried out in the purification tanks before purification commences, the drainage pipes being kept open during the entire initial washing and sufficient time being allowed thereafter for the system to be flushed clean before the purification process begins;

3. the purification tanks must be supplied with a sufficient flow of sea water per hour and per tonne of live bivalve molluscs treated;

4. clean sea water or sea water cleaned by treatment must be used for purifying live bivalve molluscs; the distance between the sea water intake point and the waste water outlets must be sufficient to avoid contamination; if treatment of the sea water is necessary, the process shall be authorized once its effectiveness has been verified by the competent authority; potable water used to prepare sea water from its major constituent chemicals must comply with the requirements laid down in Directive 80/778/EEC;

5. operation of the purification system must allow live bivalve molluscs to rapidly resume filter feeding activity, remove sewage contamination, not to become recontaminated and be able to remain alive in a suitable condition after purification for wrapping, storage and transport before being placed on the market;

6. the quantity of live bivalve molluscs to be purified must not exceed the capacity of the purification centre; the live bivalve molluscs must be continuously purified for a period sufficient to allow the microbiological standards laid down in Chapter V of this Annex to be met. This period starts from the moment at which the live bivalve molluscs in the purification tanks are adequately covered by the water until the moment when they are removed.

   The purification centre must take account of the data relating to the raw materials (the type of bivalve mollusc, its area of origin, microbe content, etc.) in case it is necessary to extend the purification period so as to ensure that the live bivalve molluscs meet the bacteriological requirements of Chapter V of this Annex;

7. should a purification tank contain several batches of molluscs, they must be of the same species and come from the same production area or different areas conforming to the same health conditions. The length of the treatment must be based on the time required by the batch needing the longest period of purification;

8. containers used to hold live bivalve molluscs in purification systems must have a construction which allows sea water to flow through; the depth of layers of live bivalve molluscs should not impede the opening of shells during purification;

9. no crustaceans, fish or other marine species must be kept in a purification tank in which live bivalve molluscs are undergoing purification;

10. after completion of purification, the shells of live bivalve molluscs must be washed thoroughly by hosing with potable water or clean sea water; this may take place in the purification tank if necessary; the washing water must not be recirculated;

11. purification centres must have their own laboratories or secure the services of a laboratory equipped with the necessary facilities for checking the efficiency of purification by use of microbiological specifications. Laboratory facilities outside the centres must be recognized by the competent authority;

12. purification centres must regularly keep a record of the following data:

   — results of microbiological tests on purification system water entering the purification tanks;

   — results of microbiological tests on unpurified live bivalve molluscs;

   — results of microbiological tests on purified live bivalve molluscs;

   — dates and quantities of live bivalve molluscs delivered to the purification centre and corresponding registration document numbers;

   — the times of filling and emptying of purification systems (purification times);

   — dispatch details of consignments after purification.

   These records must be complete and accurate, legible and recorded in a permanent ledger book which must be available for inspection by the competent authority;

13. purification centres must accept only those batches of live molluscs which are accompanied by the registration document referred to in Chapter II of this Annex;

   Purification centres dispatching batches of live bivalve molluscs to dispatch centres must provide the registration document referred to in Chapter II, section 6 of this Annex.

14. every package containing purified live bivalve molluscs must be provided with a label certifying that all molluscs have been purified.

Centres must have at least:

1. on premises where live bivalve molluscs are handled or stored:

   (a) buildings or facilities of sound construction, designed and maintained adequately for the purpose of preventing contamination of live bivalve molluscs by any type of waste, dirty water, fumes, dirt or by the presence of rodents or other animals;

   (b) flooring which is easy to keep clean and is laid in such a way as to facilitate drainage;

   (c) adequate working space to allow for satisfactory performance of all operations;

   (d) durable walls which are easy to clean;

   (e) adequate natural or artificial lighting;

2. access to an appropriate number of changing rooms, wash basins and lavatories; there must be a sufficient number of wash basins close to the lavatories;

3. adequate equipment for washing tools, containers and equipment;

4. facilities for the supply and, where appropriate, storage of exclusively potable water within the meaning of Council Directive 80/778/EEC of 15 July 1980 relating to the quality of water intended for human consumption [1] or facilities for the supply of clean sea water.

Facilities supplying non-potable water may be authorized. The water concerned may not come into direct contact with live bivalve molluscs or be used for cleaning or disinfecting containers, plant or equipment which come into contact with live bivalve molluscs. Pipes and outlets carrying non-potable water must be clearly distinguished from those carrying potable water;

5. equipment and instruments or their surfaces which are intended to come into contact with live bivalve molluscs must be made of corrosion-resistant material which is easy to wash and clean repeatedly.

II. General hygiene requirements

A high degree of cleanliness and hygiene must be required of staff, premises, equipment and working conditions:

1. staff who treat or handle live bivalve molluscs must in particular wear clean working clothes and, where appropriate, gloves which are suitable for the work in which the person is engaged;

2. staff are obliged to refrain from personal behaviour, such as spitting, which could result in contamination of live bivalve molluscs; any person suffering from an illness which can be transmitted by live bivalve molluscs must be temporarily prohibited, until recovery, from working with or handling these products;

3. any rodents, insects or other vermin found must be destroyed and further infestation prevented. Domestic animals must not enter the facilities;

4. premises, equipment and instruments used for handling live bivalve molluscs must be kept clean and in a good state of repair; equipment and instruments must be thoroughly cleaned at the end of the day's work and at such other times as may be appropriate;

5. premises, instruments and equipment must not be used for purposes other than the handling of live bivalve molluscs without authorization by the competent authority;

6. waste products must be stored hygienically in a separate area and, where appropriate, in covered containers suitable for the purpose intended. Waste material must be removed from the vicinity of the establishment at appropriate intervals;

7. the finished products must be stored under cover and must be kept away from the areas where animals other than live bivalve molluscs, such as crustaceans, are handled.

III. Requirements for purification centres

In addition to the requirements under Sections I and II, the following conditions must be met:

1. the floors and walls of the purification tanks and any water storage containers must have a smooth, hard and impermeable surface and be easy to clean by scrubbing or use of pressurized water. The base of the purification tanks must be sufficiently sloped and be equipped with drainage sufficient for the volume of work;

[1] OJ No L 229, 30. 8. 1980, p. 11. Directive last amended by the 1985 Act of Accession (OJ No L 302, 15. 11. 1985, p. 218).

5. The upper limits as regards the radionuclide contents must not exceed the limits for foodstuffs as laid down by the Community.

6. The total Paralytic Shellfish Poison (PSP) content in the edible parts of molluscs (the whole body or any part edible separately) must not exceed 80 microgrammes per 100 g of mollusc flesh in accordance with the biological testing method — in association if necessary with a chemical method for detection of Saxitoxin — or any other method recognized in accordance with the procedure laid down in Article 12 of this Directive.

   If the results are challenged, the reference method shall be the biological method.

7. The customary biological testing methods must not give a positive result to the presence of Diarrhetic Shellfish Poison (DSP) in the edible parts of molluscs (the whole body or any part edible separately).

8. In the absence of routine virus testing procedures and the establishment of virological standards, health checks must be based on faecal bacteria counts.

Examinations for checking compliance with the requirements of this Chapter must be carried out in accordance with proven methods which are scientifically recognized.

For the uniform application of this Directive sampling plans as well as the methods and analytical tolerances to be applied in order to check compliance with the requirements of this Chapter must be established in accordance with the procedure laid down in Article 12 of this Directive.

The effectiveness of the faecal indicator bacteria and their numerical limits as well as the other parameters laid down in this Chapter must be kept under constant review and, where scientific evidence proves the need to do so, be revised following the procedure laid down in Article 12 of this Directive.

When there is scientific evidence indicating the need to introduce other health checks or to amend the parameters in this Chapter for the purpose of protecting public health, such measures must be adopted in accordance with the procedure laid down in Article 12.

CHAPTER VI

PUBLIC HEALTH CONTROL AND MONITORING OF PRODUCTION

A public health control system must be established by the competent authority in order to verify whether the requirements laid down in this Directive are complied with. This control system must include:

1. periodic monitoring of live bivalve mollusc relaying and production areas in order to:

   (a) avoid any malpractice with regard to the origin and destination of the live bivalve molluscs;

   (b) check the microbiological quality of the live bivalve molluscs in relation to the production and relaying areas;

   (c) check the possible presence of toxin-producing plankton in production and relaying waters and biotoxins in live bivalve molluscs;

   (d) check the possible presence of chemical contaminants, the maximum authorized level of which will be fixed, in accordance with the procedure laid down in Article 12 of this Directive, by 31 December 1992.

   For the purposes of points (c) and (d), sampling plans must be established by the competent authorities for checking such possible presence at regular intervals or on a case-by-case basis in the event of irregular periods of harvesting.

2. Sampling plans as provided for in point 1, must in particular take account of:

   (a) likely variations in faecal contamination at each production and relaying area;

   (b) possible variations in production at relaying areas in the presence of plankton containing marine biotoxins. The sampling must be carried out as follows:

      (i) monitoring: periodic sampling organized to detect changes in the composition of the plankton containing toxins and the geographical distribution thereof. Information leading to a suspicion of accumulation of toxins in mollusc flesh must be followed by intensive sampling;

IV. **Requirements for dispatch centres**

1. In addition to the requirements under Sections I and II, the following conditions must be met:

   (a) conditioning must not cause any contamination of the product; conditioning facilities must be used in accordance with procedures recognized by the competent authorities, with special regard to the bacteriological and chemical quality of the sea water used in those facilities;

   (b) equipment and containers in the conditioning facilities must not constitute a source of contamination;

   (c) procedures for calibration of live bivalve molluscs must not result in additional contamination of the product or in any changes affecting the ability of the product to be transported and stored after wrapping;

   (d) any washing or cleaning of live bivalve molluscs must be carried out using pressurized clean sea water or potable water; cleaning water may not be recycled.

2. Dispatch centres must accept only those batches of live bivalve molluscs which are accompanied by the registration document referred to in Chapter II, section 6 of this Annex and coming from an approved production area, relaying area or purification centre.

3. Dispatch centres must have their own laboratories or secure the services of a laboratory equipped with the necessary facilities for checking, *inter alia*, whether the molluscs comply with the microbiological standards of Chapter V of this Annex. Laboratory facilities outside the centres must be recognized by the competent authority.

   However, these requirements do not apply to dispatch centres obtaining their molluscs exclusively and directly from a purification centre where they have been examined after purification.

4. Dispatch centres must keep the following data at the disposal of the competent authority:

   — results of microbiological tests on live bivalve molluscs from an approved production area or relaying area;

   — dates and quantities of live bivalve molluscs delivered to the dispatch centre and corresponding registration document numbers;

   — dispatch details.

   These data must be classified chronologically and preserved for a period to be laid down by the competent authority, but not less than three months.

5. Dispatch centres situated aboard vessels shall be subject to the conditions laid down in point 1 (b), (c) and (d) and in points 3 and 4. The conditions laid down in I and II shall apply *mutatis mutandis* to such dispatch centres although special conditions may be laid down in accordance with the procedure laid down in Article 12 of this Directive.

CHAPTER V

REQUIREMENTS CONCERNING LIVE BIVALVE MOLLUSCS

Live bivalve molluscs intended for immediate human consumption must comply with the following requirements:

1. The possession of visual characteristics associated with freshness and viability, including shells free of dirt, an adequate response to percussion, and normal amounts of intravalvular liquid.

2. They must contain less than 300 faecal coliforms or less than 230 *E. Coli* per 100 g of mollusc flesh and intravalvular liquid based on a five-tube, three-dilution MPN-test or any other bacteriological procedure shown to be of equivalent accuracy.

3. They must not contain salmonella in 25 g of mollusc flesh.

4. They must not contain toxic or objectionable compounds occurring naturally or added to the environment such as those listed in the Annex to Directive 79/923/EEC in such quantities that the calculated dietary intake exceeds the permissible daily intake (PDI), or that the taste of the molluscs may be impaired.

   In accordance with the procedure laid down in Article 12 of this Directive, the Commission shall determine the testing methods for checking the chemical criteria and the limit values applicable.

(ii) intensive sampling:

— monitoring plankton in the growing and fishing waters by increasing the number of sampling points and the number of samples, and

— toxicity tests using the molluscs from the affected area which are most susceptible to contamination.

Placing on the market of molluscs from that area may not be re-authorized until new sampling has provided satisfactory toxicity test results;

(c) possible contamination of the molluscs in the production and relaying area;

If the result of a sampling plan shows that placing on the market of live bivalve molluscs may constitute a hazard to human health, the competent authority must close the production area, as regards molluscs concerned, until the situation has been restored.

3. Laboratory tests in order to check compliance with the requirements for the end product as laid down in Chapter V of this Annex. A control system must be established to verify that the level of marine biotoxins does not exceed safety limits.

4. An inspection of establishments at regular intervals. These inspections must include in particular checks:

(a) to verify whether the approval conditions are still being complied with;

(b) on the cleanliness of the premises, facilities, equipment and on staff hygiene;

(c) to verify whether the live bivalve molluscs are handled and treated correctly;

(d) on the correct application and functioning of purification or conditioning systems;

(e) on the ledger books referred to in Chapter IV section III, 12 of this Annex,

(f) on the correct use of health marks.

These checks may include the taking of samples for laboratory tests; the results of these tests are notified to the persons responsible for the establishments.

5. Checks on the storage and transport conditions for consignments of live bivalve molluscs.

CHAPTER VII

WRAPPING

1. Live bivalve molluscs must be wrapped under satisfactory conditions of hygiene.

The wrapping material or container must:

— not impair the organoleptic characteristics of the live bivalve molluscs,

— not be capable of transmitting substances harmful to human health to the live bivalve molluscs,

— be strong enough to give adequate protection to the live bivalve molluscs.

2. Oysters must be wrapped with the concave shell downwards.

3. All wrappings of live bivalve molluscs must be sealed and remain sealed from the dispatch centre until delivery to the consumer or retailer.

CHAPTER VIII

PRESERVATION AND STORAGE

1. In any storing rooms, live bivalve molluscs must be kept at a temperature which does not adversely affect their quality and viability; the wrapping must not come into contact with the floor of the store room, but must be placed on a clean, raised surface.

2. Reimmersion in or spraying with water of live bivalve molluscs must not take place after they have been wrapped and have left the dispatch centre except in the case of retail sale at the dispatch centre.

CHAPTER IX

TRANSPORT FROM THE DISPATCH CENTRE

1. Consignments of live bivalve molluscs intended for human consumption must be transported wrapped as sealed parcels from the dispatch centre until offered for sale to the consumer or retailer.

2. The means of transport used for consignments of live bivalve molluscs must have the following characteristics:

   (a) their interior walls and any other parts which might come into contact with the live bivalve molluscs must be made of corrosion-resistant materials; the walls must be smooth and easy to clean;

   (b) they must be suitably equipped to provide efficient protection of the live bivalve molluscs against extremes of hot and cold, contamination with dirt or dust, and damage to the shells from vibration and abrasion;

   (c) the live bivalve molluscs must not be transported with other products which might contaminate them.

3. Live bivalve molluscs must be transported and distributed using closed vehicles or containers which maintain the product at a temperature which does not adversely affect their quality and viability.

   The parcels containing live bivalve molluscs must not be transported in direct contact with the floor of the vehicle or container but must be supported on raised surfaces or by some other means which prevents contact.

   Where ice is used in transporting consignments of live bivalve molluscs, it must have been made from potable water or clean sea water.

CHAPTER X

MARKING OF CONSIGNMENTS

1. All parcels in a consignment of live bivalve molluscs must be provided with a health mark so that the original dispatch centre may be identified at all times during transport and distribution until retail sale. Without prejudice to Directive 79/112/EEC, the mark must contain the following information:

   — the country of dispatch,

   — the species of bivalve mollusc (common name and scientific name),

   — the identification of the dispatch centre by the approval number issued by the competent authority,

   — the date of wrapping, comprising at least the day and the month.

   By way of derogation from Directive 79/112/EEC the date of durability may be replaced by the entry 'these animals must be alive when sold'.

2. The health mark may be printed on the wrapping material or be put on a separate label which is then affixed to the wrapping material or put inside the wrapping. It may also be of a twist-tie or staple design; self-adhesive health marks must not be used, unless they are not detachable. All types of health mark must be for single use only and may not be transferred.

3. The health mark must be durable and waterproof, and the information presented must be legible, indelible and in easily decipherable characters.

4. The health mark attached to consignments of live bivalve molluscs which are not wrapped in individual consumer-size parcels must be kept for at least 60 days by the retailer after splitting up the contents of the consignment.