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The Seafood Industry
A Self-Study Guide

Sanitation
CLEANING AND SANITATION

Every segment of today's seafood industry is touched by the increased awareness of seafood quality and the need to have a clean and sanitary processing environment. Controlling the environment to assure sanitary conditions is not new, but it is important that processors direct attention toward cleaning and sanitization problems. Improvement of sanitization and seafood quality requires the combined efforts of processing and sanitation personnel. Sanitation is the responsibility of all people in the plant, not just the sanitation crew; the production crew, management and staff must also be involved.

Cleaning and sanitation are two separate processes and are discussed in different sections of this chapter. Cleaning is a process to remove soil and prevent accumulation of food residues, which may decompose and support the growth of disease or spoilage-causing organisms. There are many reasons to clean:

1. To remove soils that will contaminate the next food process.
2. To remove and prevent bacterial buildup.
3. To meet regulatory agency standards.
4. To prevent insect and rodent infestation and harborage.
5. To improve product shelf-life.
6. To reduce chance of off-flavors.
7. To prevent staining and filming of equipment.
8. To increase equipment's thermal efficiency.
9. To lengthen the life of equipment.
10. To improve morale.
11. To increase pride in the processing facility.
12. To remove odor-causing bacteria.
13. To improve safety conditions.

To fully understand principles of cleaning and sanitation, it's necessary to have a knowledge and understanding of:

1. Microbiology and the factors affecting microbial growth.
2. The principles of a Hazard Analysis Critical Control Point (HACCP) program.
3. Chemistry of water.
4. Fundamentals of Cleaning
5. Fundamentals of Sanitation

MICROBIOLOGY AND FACTORS THAT AFFECT GROWTH

All raw foods contain microorganisms, therefore a knowledge of microbiology is essential for food safety, quality control and food preservation. Microorganisms are controlled by slowing down
or stopping their growth and by destroying them. Quality control and sanitation, in a broad sense, are used to control microbial growth. We will explore both microbiology and ways to control growth, including control by reducing initial numbers and control by retarding growth. Finally a brief discussion of pathogens that may be present in seafood will be presented.

Microbiology is the study of living organisms that are so small that they can only be observed with the aid of a powerful microscope. Food microbiologists usually divide organisms of concern into four categories: bacteria, viruses, yeast and molds. With respect to seafood, bacteria and viruses are the organisms of most concern. Molds are often observed in processing plants and most workers are aware of problems with mildew and other fungal problems. Yeast have been found in a number of seafood products. They are usually found in oysters and can be isolated from other seafood products.

Human enteric viruses (these are viruses that shed in the feces) are of concern from a public health stand point. Viruses are the smallest of the microorganisms. They are inert in food systems and are only active inside the host. Only a few viruses can be transmitted through food. These are usually transmitted by the fecal-oral route which includes contamination from human sewage. Enterovirus infections are limited mostly to the intestine, however when the infection goes past the intestine more serious illness, such as hepatitis, may result. When a person becomes infected they may shed viruses in their feces, which may in turn contaminate seafood through pollution or poor personal hygiene habits. Viruses have the potential to contaminate seafood during processing as has happened with other food products.

Bacteria are the microorganisms of most concern to the seafood microbiologist, both from the stand point of quality and safety. All raw seafood contain bacteria, which are the principal agents responsible for spoilage. Of course, some bacteria species can be of public health concern. Bacteria reproduce by fission, where one bacteria divides to form two and those two divide to make four, etc.

Bacteria are described by their Gram reaction, shape, ability to form spores, motility, requirements for oxygen and temperature range for growth. The Gram-reaction is a differential staining reaction that gives an indication of the make up of the cell wall. Gram-positive bacteria have a more rigid cell wall with less lipid whereas Gram-negative bacteria have a more flexible cell wall that contains more lipid. In this technique the bacteria are first stained with crystal violet and the stain is fixed using an iodine solution. The stain and dye form a complex within the cell wall. The cells are then washed with an organic solvent, which removes lipid from the cell wall
and in the case of Gram-negative releases the dye-iodine complex. The cells are then counter stained and examined. Gram-positive bacteria stain purple whereas Gram-negative bacteria stain pink to red.

Bacteria come in a variety of shapes and sizes. To the seafood microbiologist, rod, spherical, and comma shaped bacteria are of the most concern. Also, many bacteria are motile by means of flagella, which is a whip-like appendage. The flagella's location is important for describing the bacterium.

Some bacteria have the ability to form spores, not as a means of reproduction, but as a means of surviving adverse conditions. Spores can withstand adverse conditions far better than vegetative cells. For example, some spores can survive boiling water for several hours and are resistant to many chemical sanitizers. Spore-forming bacteria are of special concern in canning and pasteurization processes because underprocessing may allow survival, which can lead to spoilage or illness.

**Factors Affecting Microorganism Growth**

Microorganisms vary in their growth requirements including their need for oxygen, temperature, moisture etc. The growth of microorganisms can be controlled by changing the factors that affect growth. Temperature control is perhaps the most widely used method for slowing microbial growth in seafood, however other factors, including atmosphere, also are used to slow growth and prolong shelf-life.

**Temperature:** Four major physiological groups of bacteria may be distinguished by their temperature ranges for growth: thermophiles, mesophiles, psychrophiles and psychrotrophs. Thermophiles prefer high growth temperatures. They are usually spore forming bacteria and are of concern in the spoilage of canned foods. Mesophiles, which are often of animal origin, prefer moderate temperatures. Most bacterial pathogens are mesophiles. Psychrophiles prefer cold temperatures and do not grow at temperatures above 20°C. They are very important to their environments, but not as important in food spoilage or safety. Psychrotrophs are organisms which will grow at 0°C, but their optimum temperature is higher than 59° (15°C), usually between 68-86°F (20-30°C). Psychrotrophs are important in food microbiology, being responsible for the spoilage of refrigerated foods.

Temperature has a pronounced effect on the growth of bacteria. Bacteria grow fastest at their optimum temperatures
and even small temperature decreases can greatly slow growth. For example, Ingrahm demonstrated that if the generation time (time it takes to double numbers) of a psychrotrophic bacteria is 30 minutes at 77°F (25°C), it will be 75 minutes at 68°F (20°C), 120 minutes at 15°C, 200 minutes at 50°F (10°C), and 1200 minutes at 68°F (0°C). If one considers that spoilage is to a large extent a function of bacterial growth and that seafood will be spoiled at somewhere between $10^6$-10$^8$ cells per gram, then it is obvious that the initial population and generation time are critical factors in determining shelf-life and quality.

By way of an example, if 8.3 days shelf-life is expected when fillets are held at 32°F, only 2.6 days of shelf-life will be obtained at 41°F and at 50°F the expected shelf-life is only 1.4 days. Of course the shelf-life would have been longer if the initial population were lower. The initial population can be controlled by sanitation. Also, if the temperature was -2°C (28°F) the shelf-life would be considerably longer since the generation time would be much longer.

Fluctuations in temperature also can greatly affect shelf-life. For example, if a product with an initial population of 10,000 organisms per gram is held at 25°C for just 2 hours before cooling to 0°C (32°F) the expected shelf-life will be reduced nearly 45%. The important point is that rapid cooling of seafood products is essential. Seafood should not be left at room temperature during lunch or any other breaks. This practice can greatly reduce the shelf-life and affect safety. Also, the seafood should be cooled to its storage temperature within 2 - 4 hours of processing.

**Oxygen:** Bacteria vary in their requirements for oxygen. Bacteria that require oxygen for growth are called aerobes. Those requiring that oxygen be absent in order to grow are anaerobes. Organisms that can grow in either the presence or absence of oxygen are referred to as facultative anaerobic, and organism that require some oxygen for growth but are inhibited by atmospheric concentrations are called microaerophilic. Vacuum packaging and modified atmosphere packaging have long been used to extend the shelf-life of foods including seafood. Many of the spoilage organisms are aerobes and removing oxygen decreases their growth. However, there are safety concerns which will be discussed in the section dealing with Clostridium botulinum type E.

**Water Activity:** Water in food may exist in either the free or bound form. Bound water is chemically bound to the food molecule and is not available for microbial growth. Free water is water that can be exchanged with the atmosphere and is available for microbial growth. Microorganisms require free
water to grow. The water activity (Aw) of the food is a measure of the free water. Water activity is measured by placing a food in a closed container, allowing the water in the food and air to reach equilibrium and measuring the relative humidity above the food. Water activity is the Equilibrium Relative Humidity divided by 100. Pure water has a water activity of 1.0; the addition of solutes reduces the water activity. Salting a food lowers the water activity.

Microorganisms have minimum water activities for growth. In general molds have the lowest minimum water activities, followed by yeast and bacteria the highest minimum water activity. Bacteria usually do not grow when the water activity is between 0.90 - 0.93 which is equivalent to a 10-12% salt concentration. Dried seafood are slow to spoil because the water activity is low. These foods are not sterile and may actually have high microbial numbers that could quickly spoil the food if the product is allowed to rehydrate. Water activity is also important in plant sanitation. If a surface is kept dry, microorganisms can not grow, even if nutrients are present. After clean up it is important that water not be allowed to pool on equipment and tables.

Acidity and pH: Acidity and low pH have long been used as methods of food preservation. The pH scale goes from 0 - 14 with 7 being neutral. Because pH is measured on a log scale, a pH of 6 is 10 times more acid than a pH of 7 and pH of 5 is 10 times more acid than a pH of 6 and 100 times more acid than a pH of 7 etc. In general, organic acids, such as acetic or citric, are more detrimental to microorganisms than strong acids at the same pH, because organic acids act both internally and externally on the cells. Acidity and pH are important factors in sanitation and cleaning. The effect of different cleaners and the effect of pH will be discussed in later sections.

DEVELOPMENT OF A HACCP PROGRAM

The responsibility for food safety is shared by processors and regulatory agencies. In a broad sense, food safety considerations must start at harvest and continue through all stages of the seafood processing chain to the consumer.

Historically, inspections have been used by regulatory agencies to insure food safety, however, this approach has been shown to have several shortcomings. The laws, which inspectors are charged with upholding, often are not clearly written and there are questions as to what constitutes compliance. Also, it is sometimes difficult to distinguish between factors of critical importance to safety and aesthetics. In addition, the laws are not always specific; for example, the Good Manufacturing
Practices contain phrases like "clean as frequently as necessary". Because of this vagueness the umbrella GMP's do not have the force of law.

Another problem is that the inspector can only observe conditions when he is there and can not know what is happening between inspections.

The Hazard Analysis Critical Control Point (HACCP) approach presents another method to control microbiological hazards in foods. It provides a more rational approach to control of microbiological hazards than traditional inspection. The HACCP concept was first developed in 1971 at the Conference on Food Protection. Then in 1973, the Pillsbury company did a lot of work to refine the program, and the same year HACCP was adopted for low acid canned foods. The concept did not truly catch on with other products, but in 1985 the National Academy of Sciences recommended in a report entitled - "An Evaluation of the Role of Microbiological Criteria for Food and Food Ingredients" that most food processing operations should use HACCP. A HACCP approach is being developed for seafood products.

The HACCP concept is divided into two parts; the Hazard Analysis and determination of Critical Control Points. Hazards include unacceptable levels of either foodborne pathogens or spoilage organisms, to the extent that food is potentially hazardous or has a reduced shelf-life. A hazard analysis evaluates all procedures concerned with production, distribution, and use of raw materials and food products. This requires a thorough knowledge of food microbiology and it is important to know what types of microorganisms might potentially be present and the factors that affect their growth and survival. Food safety and quality problems most often are caused by contaminated raw food or ingredients; improper holding temperatures (time-temperature abuse); improper cooling of foods; improper handling of seafood after processing; cross contamination between products or between raw and processed product; infrequent disassembly and cleaning of equipment; plant design inadequate to allow the separation of raw product from cooked product; poor employee hygiene; and poor sanitation practices. Roughly 25 percent of foodborne illnesses can be traced directly to food-handlers not washing their hands properly.

A food and its raw materials may be classified into hazard categories by means of a two-step procedure: Risk Assessment followed by step two, Assignment of Hazard Categories. Risk Assessment is accomplished by examining the food for three possible hazards.

Hazard 1 - The product contains a sensitive ingredient or ingredients. From a microbiological standpoint, all ingredients have some degree of risk varying from high to
essentially zero. In general, products of animal origin and seafood products are considered most sensitive.

**Hazard 2** - If the manufacturing process does not contain a processing step that effectively destroys harmful bacteria. The processing steps referred to here can be cooking, pasteurization, retorting, irradiation etc. or the development of a food system that, by its composition, destroys harmful microorganisms. An example of the latter is preparation of a seafood product in an acidic tomato based product where the equilibrium pH is below 4.6.

**Hazard 3** - Where there is substantial potential for microbiological abuse in distribution or consumer handling that could render the product harmful when consumed. The principal judgment criterion for abuse potential is whether the food product--in the state in which it is distributed or as normally prepared by the consumer--is a good medium for microbial growth. Consideration must be given to low levels of microbial contamination which have escaped control screening or processing designed to prevent contamination.

The three general food hazard characteristics can be used to develop hazard categories.

The other step developing a HACCP program is to determine the Critical Control Points (CCP) within the process. It is important to separate the critical from the non-critical. Critical control points are points in the process where if not controlled could lead to a food safety or quality hazard. A practical approach to determining CCPs consists of utilizing a HACCP worksheet with the following headings:

1. Description of the food product and its intended use
2. Flow diagram with the following parts:
   - Raw material handling
   - In-process preparation, processing, and fabrication steps
   - Finished product packaging and handling steps
   - Storage and distribution
   - Point of sale handling

Each step in the process should be examined and a degree of importance assigned. For example, the steps may be graded as control points on a scale of 1 to 5 based on their perceived importance. This will require a knowledge of factors which can lead to safety and quality problems. Hazard categories of raw materials and ingredients, process foods, and finished product are often assigned to help decide on the degree of importance.

As the flow diagram is made, it is easy to identify critical control points which can be a location, practice, procedure or
process, and if controlled, can prevent or minimize contamination. Critical Control Point (CCP) must be monitored to ensure that the steps are under control. Monitoring might include observation, physical measurements (pH, Aw, Cl) or microbiological analysis. Monitoring most often includes visual and physicochemical measurement because microbiological testing is often too time consuming. Exceptions are microbiological analysis of the raw product. When the microbiological status of the raw material is a CCP then microbiological testing is the only acceptable monitoring procedure. Also, when foods are destined for high risk population the finished product is tested. In this case the microbiological testing is part of the process. Actually, I want to emphasize that microbiological testing is usually not necessary, however, knowledge of microbiology is necessary.

If microbial standards are applied within the HACCP program, it should be with careful consideration. Microbial methods can be used to directly determine presence of hazards in raw materials, during processing and in the finished product. They can also be used indirectly to monitor effectiveness of control points such as cooking, cooling, cleaning, and employee hygiene. However, this use of microbiology is a check and may not have to be an ongoing process.

The HACCP concept is being used successfully in the low acid canned food industry where it works because:

1. Monitoring procedures for CCP were developed cooperatively by government and industry.

2. Education of processors is required. All operation are under supervision of someone who has completed a "Better Process Control School".

3. Training of inspectors; FDA inspectors are trained in HACCP.

4. Use of HACCP for low acid can foods is mandated by law.

If HACCP is to work in the seafood industry, the following problems must be overcome.

1. HACCP requires technical sophistication on the part of plant personnel.

2. Regulators in the seafood area must be trained.

3. Food processors must be trained in HACCP.

4. HACCP must be required by law.

5. Trust between regulators and processors, which may be the biggest obstacle to overcome.
The primary constituent of all food processing plant cleaning is water. Basic water requirements are that it must be free from disease-producing organisms, toxic metal ions, and objectionable odors and tastes. Pure water presents no problems, but no food processing establishment has an ideal water supply. Therefore, the cleaning compounds must be tailored to the individual water supply and the processing operation. It is important not to assume the water is good. The water should be routinely checked for bacterial levels and water hardness.

An understanding of the chemistry of water is essential for an understanding of the "Chemistry of Cleaning". Water, the major component in most cleaning solutions, acts as both a solvent and as a carrier for the various ingredients in the cleaning compound. A knowledge of the nature and characteristics of water is invaluable in developing an optimum cleaning and sanitizing program.

Pure water is available only in the laboratory. The source of water used for cleaning may contain a variety of dissolved substances. The substances that are of concern in developing a cleaning and sanitizing program are calcium, magnesium, iron, chlorides, sulfates, manganese, sulfur and carbon dioxide. The presence of these substances define the characteristic of the water and give rise to terms such as hard, soft, red, black, acid etc.

The hardness or softness of water is related to the amount of calcium and magnesium present. The higher the concentration, the "harder" the water. The degree of hardness is measured in parts per million (ppm) or grains per gallon (gpg). One grain per gallon or just one grain hardness is equivalent to 17.1 ppm. There are two types of hardness, temporary and permanent. Hardness is temporary when it can be removed by boiling the water; during boiling, the bicarbonates decompose and react with the calcium and magnesium forming carbonates. Permanent hardness can only be removed by chemical treatment. The sum of the temporary hardness and the permanent hardness is the total hardness of the water.

Using hard water in the cleaning process may result in the formation of a tough adherent scale, which is influenced by the heat of the cleaning solution and the components of the cleaner. If an improper cleaner is selected or if the concentration is not correct, a residue of soil remains and the scale is called a foodstone, (milkstone, beerstone, beetstone, etc.) While most of these scales can be removed, the process is time consuming and expensive.

Red water is caused by a relatively high concentration of
iron. Initially, the iron is present in a soluble, colorless form; however on exposure to air, the colorless form changes to an insoluble form, red in color (rust). Approximately 0.3 ppm iron is required for red color to be evident. The iron will react with chlorine, which is present in many cleaner formulations, to produce a brown precipitate, ferric chloride.

Other problems often encountered in water supplies are the presence of salt and acid. Salt waters, those with a high chloride or sulfate content, are often encountered in coastal areas. Generally, salt waters do not affect the cleaner efficiency; however, they do present the danger of corrosion, which can be significant. Water can be quite acidic in some areas. In general, acid waters do not affect cleaner performance is corrosive, and if appreciable amounts of salt are present, the corrosive nature is compounded.

A water supply can have a number of these characteristics. It may be both hard and contain iron or have both temporary and permanent hardness. The character of water may change from day to day or even from hour to hour because of changes in the raw water supply, changes in municipal treatment, or changes in plant treatment.

FUNDAMENTALS OF CLEANING

The most important factors in controlling microbial growth are reducing the initial numbers and slowing the rate of growth. Cleaning reduces initial numbers of microorganisms. Cleaning can be defined as removal of soil. In addition, the microorganisms that are left after the cleaning step are more susceptible to sanitizer because of the removal of organic matter. It is important that cleaning and sanitization be a two step process. The use of sanitizers, without proper cleaning is usually a waste of money and time. Cleaning compounds allow water to efficiently penetrate, dislodge and remove soil.

Four things happen during cleaning. One the detergent solutions that come into contact with the soil will remove it by means of good wetting and penetrating properties. Two solid and liquid soils are displaced from the surface by saponifying the fat, peptizing the proteins, and dissolving the minerals. Three, dispersion of the soil in the cleaning solution is obtained by deflocculation or emulsification. And four, soil is prevented from being re-deposited on the surface by good rinsing quality of the cleaning compounds used. The following basic procedures should be used for cleaning under most conditions:

1. **Pre-rinse:** Remove excessive soil with a pre-rinse either by Clean In Place or by a rinse hose designed to provide maximum efficiency and minimal energy loss.
2. **Application of detergent solution:** Apply detergent either through the C.I.P. system, high pressure system or foamer.

3. **Post rinse:** Remove the detergent solution with the C.I.P. rinse cycle or moderate high pressure system.

4. **Acid rinse:** Under certain conditions, it is necessary to apply acid solution to aid in the removal of soils. Know the soils and conditions.

5. **Sanitize:** Sanitizers are either applied through automatic dosing (C.I.P.), fogging or flooding. Learn which method is the most effective for each given operation. Some sanitizers should not be used in particular areas of the operation due to the nature of the sanitizer.

6. **Final rinse:** A potable water rinse is sometimes required because of potential problems with worker discomfort, off-flavors, etc. Some are prohibited at particular times by federal regulations. The requirements are specific per sanitizer used. Be Safe! Read the Label!

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**Nature of the soil.**

Soil is defined as matter out of place. For example, grease on a gear box is a lubricant, but that same grease on a food contact surface becomes "soil". Soils vary depending on the nature of the food and processing conditions. Seafoods contain protein, fat and minerals. In addition, sugar may be used in formulation of further processed foods. Proteins and fats are not soluble in water but are soluble in alkali and proteins are slightly soluble in acid. They are difficult to remove and heating increases the difficulty. Monovalents salts such as sodium chloride are water soluble and are easy to remove. Heating does not increase the difficulty of removal. However polyvalent ions such as calcium phosphate are not soluble in water but are soluble in acid. Heating causes these compounds to interact with other constituents in food which makes removal more difficult. Sugars are water soluble and are easy to remove however heating can cause carmelization which increased cleaning difficulty.

The following table outlines types of soil and their ease of removal under different conditions.
<table>
<thead>
<tr>
<th>SOIL</th>
<th>SOLUBILITY</th>
<th>REMOVAL</th>
<th>HEAT CHANGE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fat and oil</td>
<td>Water insoluble, alkali soluble</td>
<td>Difficult</td>
<td>Polymerization more difficult to clean</td>
</tr>
<tr>
<td>Protein</td>
<td>Water insoluble, alkali soluble, Acid</td>
<td>Very</td>
<td>Denaturation, much more difficult to clean</td>
</tr>
<tr>
<td>Carbohydrates</td>
<td>Water soluble</td>
<td>Easy</td>
<td>Carmelization, more difficult to clean</td>
</tr>
<tr>
<td>(sugar)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mineral Salts</td>
<td>Water soluble, acid soluble</td>
<td>Easy</td>
<td></td>
</tr>
<tr>
<td>Monovalent</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Milk-Stone</td>
<td>Water insoluble, Acid soluble</td>
<td>Difficult</td>
<td>Interactions with other constituents, more difficult</td>
</tr>
<tr>
<td>Food-Stone</td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

**VARIABLES OF CLEANING**

The variables of time, temperature, concentration and mechanical action affect cleaning regardless of what is to be cleaned. A specific length of time is required to clean a given surface and a given soil. For example, a doctor scrubs for a minimum of twenty minutes before surgery to simply remove soil and contaminants. He can take no shortcuts. In the plant situation time is the most common variable. Production runs are disrupted by mechanical problems or an important sales order has to be filled now. How do we compensate for the loss of necessary time? The variables of temperature, concentration and mechanical action can be altered to compensate for the loss of time; however changing these other variables can create problems. For example, temperature can be increased but there is a point were the increased heat is more of a detriment than a help. Increasing temperature often decreases the strength of the bond between soil and surface. It can also decrease viscosity while increasing solubility of soluble materials. For every 18°F (10°C) temperature increase, the effectiveness of the solution is doubled; however for every increase of 4°F (2.2°C) the protein structure is changed. It has been said that for each 4° increase
the molecule grows additional suction cups. Residue of burned on material is very difficult to clean under the best conditions. Concentration can be increased, but if the solution is saturated with cleaning products it can not clean properly. The correct amount of cleaning solution usually does an excellent job, but too much can greatly decrease effectiveness.

In hand cleaning, mechanical action is applied by "elbow grease"; whereas, fluid flow is used to apply cleaning force in C.I.P. systems. Decreased turbulence provides more effective removal of film from surfaces. However, efficiency is less effected by turbulence as the physical-chemical effectiveness of the detergent increases. C.I.P. cleaning velocities of 5 ft./second are required to ensure adequate turbulence. In foam cleaning the mechanical action is provided by the rinse hose. It is important to rinse from the bottom up. Proper procedures are mandatory to insure a clean surface in any method of cleaning from C.I.P. to hand cleaning. Though the C.I.P. system is automated, it must be observed to assure that the valves do actuate, that the rinse cycle was the proper length and that the sanitizer is automatically dosed at proper concentration.

In selecting cleaning compounds many interrelated factors are involved, including:

1. Quality of water available.
2. Temperature.
3. Type and amount of soil on the surface.
4. Surface to be cleaned.
5. Physical nature of cleaning compound (liquid or powder).
6. Method of cleaning (foaming, C.I.P., soaking, manual cleaning, etc.).
7. Economics
8. Time available.

DETERGENT FUNDAMENTALS

The term detergent is quite broad, referring to any agent that will remove one undesirable substance from another. In the removal of soil, a cleaner functions in various ways involving both physical and chemical reactions. These functions do not occur separately or in any particular sequence but in a complex interrelated manner. All these functions are important and usually all are present in a cleaner to some extent. For cleaning a particular type of soil, certain functions are emphasized more than others to arrive at a balanced product.
Functions of Effective Detergents

An effective detergent must have the following properties:

1. Rapid penetrating and wetting power.
2. Ability to control water hardness.
3. High detergent power to remove soil.
4. Suspending power to keep the removed soil from redepositing on the surface being cleaned.
5. Easy rinsability.
6. Non-corrosiveness to surfaces being cleaned and to cleaning equipment.

In practice these functions are not observed independently but all occur together. No simple chemical—alkali, acid, wetting agent, etc.—can supply all the previously mentioned properties. By combining selected chemicals, cleaners can be prepared having the desired characteristics. No one combination will include all the various functions in the desired amounts to be effective on a given application; different cleaning compounds are required for different cleaning tasks. One group of detergents that work satisfactory in one plant may not be effective in a similar plant because of difference in water supply.

An understanding of terms used in detergents will help understand the proper application and description of the detergents. Terms commonly encountered include:

Alkalinity of a solution is the actual amount of the alkali present. Because many different alkalies are used in formulating detergents, certain standards of expression have been developed. In the cleaning industry, alkalinity is expressed as "the alkalinity equivalent to sodium hydroxide" or the "alkalinity equivalent to sodium oxide". Sodium oxide is 76% that of the alkali expressed as sodium hydroxide. The alkalinity of any product can be expressed in terms of either of these two chemicals regardless of the source of the alkalinity. By this means, different alkaline products can be compared to each other. Alkalinity consists of two parts, the active and the inactive which together comprise the total alkalinity. Active alkalinity is the portion that exists above a pH of 8.4, the point at which a phenolphthalain indicator changes from red to colorless. The term active is used because this is the alkalinity responsible for cleaning. Inactive alkalinity is that portion that exists between a pH of 8.4 and 3.4, the point at which a methyl orange indicator changes from yellow to orange. The term inactive is used because in this pH range little or no cleaning is obtained.

Acidity of acid detergents is the amount of acid present in the solution. We do not specify an inactive and active form as we do with alkalinity. The importance of acidity is in relation
to the amount of mineral salt that can be dissolved in the solution. Above a pH of 3.9 for phosphoric acid products, the mineral dissolving capability of the solution decreases rapidly. A standard of acidity to compare various acid solutions has not been established. Therefore, to compare acid cleaner solutions by titration of the acidity present requires a knowledge of the actual acid present.

*Saponification* is the chemical conversion by alkali of water insoluble fatty acids soils into more soluble substances - soaps.

*Emulsification* is the action of breaking up fats and soils and colloidally dispersing them throughout the cleaning solution. The emulsion thus formed must be stable enough to prevent these soils from redepositing on equipment surfaces.

*Dispersion* is the action of breaking up solid aggregates of soil into smaller particles down to colloidal size. This is accomplished through the action of the chemical media and mechanical agitation.

*Peptization* strictly speaking occurs only by chemical action without agitation and can be considered as spontaneous dispersion of the solid soil throughout the cleaning solution. Peptization is usually associated with the removal of protein soils.

*Solubilization* occurs in two ways, physically or chemically. Lactose, found in milk solids, is soluble in water and therefore easily removed. Mineral salts found in stone deposits are solubilized by acid cleaning solutions through chemical alteration into soluble substances. Various insoluble oils are easily solubilized by surface active agents by the action of the micellar structure in aqueous media.

*Suspension* is the act of holding divided particulate matter in the liquid phase. Suspensions can be stabilized by polyelectrolytes in solution which maintains a positive or negative charge on the dispersed phase. A stable suspension of soils is particularly important in preventing re-deposition of the soils.

Wetting and Penetration are complex phenomena and depend on diffusion rates, surface tension, concentration, and roughness of the surface. Surface active agents are clearly superior in lowering the surface tension of the cleaning solution allowing the displacement of soils and the penetration into cracks and holes of solid deposits.

*Rinsability* of detergents cannot be over-emphasized. The last step of the cleaning process, it is the result of all the properties previously discussed. An acceptable rinsed surface is one free of all particulate matter and detergent film.
Water softening is the function of rendering the hardness of water unavailable for reaction with certain components of the cleaning solution. For example, caustic soda forms an adherent film of calcium and magnesium carbonates in hard water. Softening can be accomplished by precipitating the hard water elements as insoluble salts. A second method is by sequestration. Certain chemicals, notably the polyphosphates and ethylenediaminetera acetic acid (EDTA), can form soluble complexes with calcium and magnesium salts. The calcium and magnesium are so firmly held in this complex that they are unaffected by the addition of substances normally reacting with these hardness elements.

Corrosiveness is a property which must be closely controlled to protect equipment from deterioration due to the aggressiveness of the detergent and protect the equipment from the corrosive nature of accumulated soils.

The ideal detergent, then, would contain all these properties to the maximum extent. This, of course, is not possible. For instance, the most efficient chemical for saponification is caustic soda which is also the most aggressive towards aluminum and precipitates calcium and magnesium as an adherent film. The addition of glassy silicates will lessen the corrosive effect at a significant loss of alkali available for saponification. Polyphosphates will soften the water and provide other benefits but, again, alkali is decreased. Consequently, each detergent is geared to a specific application or to several very closely related applications. There can be no "all-purpose" cleaner; at best, we can only build in a degree of versatility.

DETERGENT INGREDIENTS

The components of a detergent exhibiting the listed properties can be categorized as follows:

1. Hydroxides
2. Carbonates
3. Silicates
4. Phosphates
5. Chelates
6. Oxidants
7. Acids
8. Surfactants

Sodium hydroxide, commonly called caustic soda is the strongest alkaline material employed. It is excellent for the saponification of fatty soils such as those found in milk and cooking oils. Caustic soda has some serious limitations. It has
only fair emulsifying and deflocculating properties, exhibits poor rinsing characteristics and is corrosive to some of the metals used in food processing equipment. Under the proper conditions—a 1% to 4% concentration at 140°F to 160°F for 5 to 10 minutes—caustic soda has shown bactericidal properties.

Sodium carbonate, commonly called soda ash, is a moderately strong alkali that contributes to the total alkalinity of the composition and is more easily inhibited against attack on soft metals than is sodium or potassium hydroxide. Soda has a relatively high absorption capacity for liquids, thus providing a base for the addition of liquid detergents while maintaining free flowing characteristics. Its major disadvantage in hard water is its property of forming a hard scale of calcium carbonate and similar insoluble salts. Sodium bicarbonate, commonly called baking soda, is used primarily in conjunction with soda ash as a buffering agent in mild, moderately alkaline systems.

The silicates exist in several crystalline forms providing a range of useful properties. Sodium orthosilicate is next to casuistic soda as a saponification agent; however, it also provides much the same undesirable corrosive characteristics as caustic soda. Sodium sequesilicate is lower in alkali content than the orthosilicate and provides good emulsifying and soil suspending properties, and although not as corrosive as the orthosilicate, it is nonetheless aggressive towards soft metals. Sodium metasilicate has much to recommend its use. Its wetting ability is good, emulsification and deflocculation high and it possesses definite anticorrosive properties. The glassy silicates, those having a silica to sodium oxide ratio greater than 1:1, have excellent corrosion inhibiting properties. Their major disadvantages are a lower solubility rate and greater tendency for moisture absorption which has an adverse effect on the free flowing characteristics of the cleaning composition.

Phosphates, as a group, provide the best means of maximizing all the characteristics of a good detergent with the exception of providing alkali for saponification. Trisodiumphosphate was the most outstanding of all basic alkalies in water softening prior to the advent of polyphosphates. Although its softening process occurs by precipitation, the precipitate is very finely divided, non-adherent and easily rinsed. Its properties of emulsification, dispersion and wetting are only slightly less than those of sodium metasilicate. A disadvantage is its corrosive effect on aluminum. Polyphosphates have the ability to modify the natural behavior of hard water. As a result, many uses for polyphosphates as water conditioners have been developed. The threshold treatment of hard water involves the addition of a few parts per million of polyphosphates, which inhibit the precipitation of hard water salts in many cases. The threshold treatment is limited to additions up to 20 parts per million. Polyphosphate is also a sequestrant. Sequestration is
the term applied to the mechanism whereby calcium and magnesium are held in a soluble complex which is not affected by the addition of substances normally reacting with and precipitating these hardness elements. The amount of polyphosphate required to function as a sequestering agent in any given specific application depends on several factors including the polyphosphate used, the water hardness, the nature of the cleaner and the conditions of the cleaning operation. Tetrasodium polyphosphate, although a poor calcium sequestrant, is an excellent magnesium sequestrant and is widely used because it is the most stable under conditions of high alkalinity and temperature. Sodium tripolyphosphate has good overall properties, making it perhaps the most widely used of the polyphosphates. It is an excellent sequestrant for both calcium and magnesium and provides good detergency, wetting, deflocculation, and rinsing properties. Sodium hexametaphosphate is used primarily as a calcium sequestrant when hardness alone is considered. It is unstable in hot alkali, reverting to the ortho and pyrophosphate forms. Sodium acid pyrophosphate is an excellent buffering and peptizing agent; however, it is deficient in its sequestering and wetting ability. Note phosphates are banned in many areas.

The chelates are a class of specialty chemicals of limited versatility but nonetheless important. They function similarly to the polyphosphates in forming a water soluble complex with the hardness elements, calcium and magnesium. They also will chelate iron and other metals that cause staining. The most widely used chelates are ethylenediaminetetra acetic acid (EDTA) and sodium gluconate.

The only oxidants commonly used in detergents are peroxides and those chlorine bearing compounds capable of forming hypochlorous acid. Their primary functions are as bactericides and bleaches for food stains. Detergent solutions containing available chlorine are particularly effective in the removal of proteinaceous soils.

Although the majority of detergents today are alkaline based, the acid-based detergents are alkaline necessary to "correct" some deficiencies of products and to perform functions specific to their nature. In general two types of acid detergents are used on food processing equipment.

1. Waterstone/foodstone removers
2. General purpose acid cleaners.

Waterstone and foodstone are calcareous deposits composed of calcium and magnesium solids reacting with components of alkaline cleaners and food residues. The deposits are for the most part insoluble in alkali and over a period of time will form a hard, tenaciously adherent deposit. Accumulations of these deposits
will provide harboring areas for bacteria, will significantly reduce heat transfer efficiency and cause corrosion. The use of strong mineral acids such as muriatic acid, while quite effective, have limited applicability due to the handling hazards and corrosion problems, even when they contain corrosion inhibitors. The most widely used acids are phosphoric, glycolic, sulfuric, and sodium bisulfite. They are rarely used in concentrated form but rather contain wetting agents and inhibitors to enhance their detergents properties and lessen the corrosive effects.

Surfactants have advanced the technology of detergents immeasurably. Common soap (the saponification product of fatty acids, animal fat and alkali) was probably the first surfactant. Soaps perform all the desirable functions of a detergent with some serious shortcomings. They are limited to the alkaline pH range. They are precipitated by hard water producing the familiar "bath tub ring".

These inherent shortcomings can be overcome only by using an amount in excess to precipitate all the calcium and magnesium, thus softening the water, and by restricting their use to alkaline cleaning. Surfactants have largely supplemented soaps in the detergent industry. While their chemistry and classification is very complex, they can be classified quite broadly according to their electrical nature as:

1. Anionic
2. Nonionic
3. Cationic

SYNERGISM OF DETERGENT INGREDIENTS

To this point, we have limited the discussion to the various properties when several agents are combined in a commercial cleaning compound. Blending the inorganic builders and surfactants would appear to dilute the major contribution of each component from the pure form; and this does occur. However, one final concept of detergency comes to light. Synergism is defined as the combined or cooperative action of separate agencies which together have greater total effect than the sum of their individual effects. Caustic soda is an excellent saponifying agent but in hard water an adherent film of calcium carbonate. In addition, it is not capable of saponifying all fats and provides poor rinsing properties. The addition of polyphosphates or EDTA will soften the water eliminating the calcium film. The addition of a surfactant with good wetting properties will carry the detergent solution into the soil, lifting and dispersing the soil for more complete reaction with the alkali. Unreacted soil is emulsified or dispersed and held in solution until final rinsing, which has been greatly enhanced, due to the presence of phosphates and surfactants. One final point with regard to the
action of a built detergent, the detersive mechanism performs over a period of time at a specific temperature and concentration of detergent. Unless these three factors are carefully controlled, all the time, effort expended in development of detergent has been wasted.

FUNDAMENTALS OF SANITIZERS

The most important principle concerning sanitizing/disinfection is that a dirty surface can not be sanitized. The sanitizer won't be able to reach microorganisms trapped in and under soil. There are many types of sanitizers and each type will be discussed separately.

**Active chlorine:** For many years, active chlorine has been used because of its broad bactericidal spectrum and economic advantages. The active chlorine carrier has taken several forms with the liquid types based on inorganic chlorine compounds, such as sodium hypochlorites and the powder form based on organic chlorine compounds, such as the dichloroisocyanurate group. Most often the bactericidal effect of active chlorine is best in a neutral or weak-acidic condition (pH 5 to pH 7), but the chlorinated alkaline cleaners also have an excellent bactericidal effect against all groups of microbes. Tests have shown that chlorine renders a very fast kill on viruses, bacteria, yeasts, and molds. The activity against spore-forming bacteria is slightly slower. The question of the corrosiveness of hypochlorite solutions on metals such as stainless steel and aluminum is still a matter of intense debate, resulting in certain reservations regarding chlorine-based products.

Chlorine sanitizers have these additional advantages:

1. unaffected by hard water scales
2. non-filming
3. can be utilized at cool water temperatures without affecting activity

Disadvantages include:

1. precipitation when used in iron-laden water
2. short residual effect after sanitizing

As a general rule of thumb, if chlorine is used as a sanitizer prior to production, the equipment should be used within one hour after the sanitizing procedure.

**Iodophor Sanitizers:** Acidic iodine-based sanitizers have a universal killing effect on all types of microbes. The amount of active ingredients to achieve the same killing power is lower in iodophors than in active chlorine-based products. Usually by increasing the temperature of the sanitizing solution, the
killing time is reduced, and this is true for the iodine-type products as well. Iodine will gas off at temperatures of 102°F-120°F and the loss of the iodine is high. This and the possibility of corrosion make it standard practice to use iodophors at room temperature.

Advantages include:

1. Stable, long shelf-life
2. They are active against all microorganisms except bactericidal pores and phages
3. They are unaffected by hard water salts, with the exception of water which contains large amounts of chlorides

Disadvantages include:

1. They are not as effective against spores and phages as chlorine
2. They are expensive
3. Iodophors stain porous metal surfaces and plastics
4. Iodophors are severely affected by alkaline conditions above pH 7

**Hypochlorites:**

Advantages:

1. Powerful germicides controlling a wide range of microorganisms
2. Deodorizer
3. Non-poisonous to man at use concentrations
4. Free of poisonous residuals
5. Colorless and non-staining
6. Easy to handle
7. Most economical to use

Disadvantages:

1. Short shelf-life
2. Adverse effect on skin
3. Corrosive on some metals

Use Concentrations:
50-100 ppm available chlorine should be employed for sanitizing large equipment and utensils and 200 ppm for spraying applications of large equipment. The contact time for effective sanitation should be long enough to produce complete kill of bacteria, usually 10 seconds or longer.
Acid Anionic:

Advantages:

1. Non-staining, stable, long shelf-life
2. No objectionable odor
3. Removes and prevents milkstone and waterstone formation
4. Effective against a wide spectrum of organisms
5. Stable in concentrated form or use dilutions, action enhanced by high temperatures
6. Non-corrosive to stainless steel
7. Provides short duration residual bacteriostatic effect on stainless steel equipment

Disadvantages:

1. Effectiveness at acid pH only
2. Generation of foam
3. Low activity against spore forming organisms
4. Corrosive to metals other than stainless steel

Use Concentration: 100 ppm anionic surfactant.

Quaternary Ammonium Compound (QUAT)

Advantages:

1. Stable, long shelf-life
2. Active against many microorganisms
3. Forms a bacteriostatic film
4. Non-corrosive and non-irritating to skin
5. Stable in the presence of organic matter
6. Stable to temperature changes
7. Good penetration qualities
8. Combined with non-ionic wetting agents, it makes a good detergent sanitizer

Disadvantages:

1. Expensive
2. Incompatible with common anionic detergent components
3. Slow to dissipate (residual problem)
4. Germicidal efficiency varied and selective
5. Foam problem in mechanical application

Use of Concentrations: For sanitizing equipment, a 200 ppm solution is sufficient to reduce bacterial counts with a one minute exposure.
<table>
<thead>
<tr>
<th>Specific Area or Condition</th>
<th>Recommended Sanitizer</th>
<th>Concentration (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aluminum equipment</td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>Bacteriostatic film</td>
<td>Quat</td>
<td>200</td>
</tr>
<tr>
<td>CIP cleaning</td>
<td>Acid sanitizer</td>
<td>130</td>
</tr>
<tr>
<td></td>
<td>Active chlorine</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Iodophor</td>
<td></td>
</tr>
<tr>
<td>Drain cleaning</td>
<td>quat</td>
<td>200</td>
</tr>
<tr>
<td>Film formation, prevention of</td>
<td>Acid sanitizer</td>
<td>130</td>
</tr>
<tr>
<td></td>
<td>Iodophor</td>
<td></td>
</tr>
<tr>
<td>Fogging atmosphere</td>
<td>Active chlorine</td>
<td>800-1000</td>
</tr>
<tr>
<td>Hand dip-production</td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>Hand sanitizer washroom</td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>Phenolic</td>
<td>2-3%</td>
</tr>
<tr>
<td>Hard water</td>
<td>Acid sanitizer</td>
<td>130</td>
</tr>
<tr>
<td></td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>High iron water</td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>Long shelf-life</td>
<td>Iodophor</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Quat</td>
<td></td>
</tr>
<tr>
<td>Low cost</td>
<td>Hypochlorite</td>
<td>-</td>
</tr>
<tr>
<td>Noncorrosive</td>
<td>Iodophor</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td>Quat</td>
<td></td>
</tr>
<tr>
<td>Organic matter, stable</td>
<td>Quat</td>
<td>200</td>
</tr>
<tr>
<td>in presence of</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Plastic crates</td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>Porous surface</td>
<td>Active chlorine</td>
<td>200</td>
</tr>
<tr>
<td></td>
<td>Quat</td>
<td>-</td>
</tr>
<tr>
<td>Processing equipment-stainless steel</td>
<td>Acid sanitizer</td>
<td>130</td>
</tr>
<tr>
<td></td>
<td>Active chlorine</td>
<td>200</td>
</tr>
<tr>
<td></td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>Rubber belts</td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>Tile walls</td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>Visual control</td>
<td>Iodophor</td>
<td>25</td>
</tr>
<tr>
<td>Walls</td>
<td>Active chlorine</td>
<td>200</td>
</tr>
<tr>
<td></td>
<td>Quat</td>
<td>200</td>
</tr>
<tr>
<td>Water treatment</td>
<td>Active chlorine</td>
<td>20</td>
</tr>
<tr>
<td>Wood crates</td>
<td>Active chlorine</td>
<td>1000</td>
</tr>
</tbody>
</table>

General recommendations

1. Make the cleaning and sanitizing of walls, ceilings, floors, and drains part of the daily cleanup program. Use a foaming chlorinated cleaner.

2. Eliminate all direct openings from outside into the processing and packaging rooms even though they may contain screens, curtains and louvers.

3. Use bacterial filtering systems on air handling units and create a positive pressure in the processing and packaging areas. Keep air from blowing onto product or food contact surfaces. This may seem extreme, but it is a positive approach to environmental hygiene.

4. Make sure dehumidifiers or air conditioning units drain away from the processing or packaging room and prevent condensate from re-entering the atmosphere. Coils and pans must be cleaned and sanitized routinely. Use a chlorinated foam product.

5. Repair all cracks in walls and floors. This is especially important for control of Listeria in areas around floor drains.

6. Sanitize pallets (wet) before placing them in the process areas.

7. Keep areas under floor conveyors and equipment clean and sanitized, especially in and around coolers. This is a critical point! There are numerous instances where Listeria has been detected at these points.

8. Allow only employees associated with processing and packaging into these areas. In traffic areas set up properly maintained foot sanitizing baths.

9. Require the use of proper clothing and footwear which must not be worn outside the plant, even at lunch time.

10. Establish a specific environmental cleaning and sanitizing program for all areas of the plant. Consider using a special crew for this purpose and include the floor drains.
Specific recommendations

Post Processing Sanitation Control Program: This program has been designed to cover sanitation in all areas following heat processing. (Ready to eat seafood products)

I. Floor Drains

   a) Must be working properly - maintenance.

   b) Don't use department hoses to unplug drains. Use a dedicated hose or other appropriate means supplied by mechanical department.

   c) Daily cleaning

       1) Remove covers and clean with detergent and brush.

       2) Heavily foam and brush down with at least a 3' long brush.

       3) Thoroughly spray all sides of drain cover and drain itself with sanitizer solution.

       4) Whenever a stopped-up drain has been fixed, the floor surrounding, drain cover and drain must be cleaned and sanitized as noted above.

II. Standing Water - puddles, under equipment, areas of disrepair or anywhere water collects

   a) A floor maintenance program must be put into effect that will: repair broken/depressed areas, seal cracks/crevices, make sure water will drain from under equipment, make sure water runs freely to and into drains.

   b) After final clean-up and sanitizing of floors, all free or standing water must be removed by vacuum or squeegee to and into drains.

   c) During production, any standing water on floors must be vacuumed or squeegeed into drains at each break, lunch and shift change. Note: This includes inside and under any equipment.

   d) Vacuum equipment, e.g., hose, nozzle and air filter coming out of tank must be cleaned and sanitized daily or the vacuum will become a reservoir for bacteria.

III. Floor Cleaning

   a) During production, no high pressure water that produces aerosols can be used to clean floors. Use squeegees and scoops for dry clean only and/or use very low pressure water that will not produce aerosols.
b) Night Clean-Up - After dry cleaning and foaming of equipment, the total floor must be foamed. After rinsing of equipment and floor, all equipment and total floor must be sanitized. **Note:** Pay particular attention that the total floor (under equipment, racks, in corners, around posts and beams, etc.) is thoroughly cleaned and sanitized.

IV. Ceilings, Cooling Units, Drip Pans, Overhead Pipes, Doors, Walls

a) Fix leaks and remove drip pans when possible.

b) Clean and sanitize weekly by:
   1) Thorough foam and rinse.
   2) After rinse apply a sanitizing solution of quaternary ammonia (10% strength) mixed 1 oz. per gallon of water.
   3) After cleaning and sanitizing overheads, all equipment must be cleaned and sanitized.
   4) Be sure all product, packaging materials, etc. are removed from rooms before cleaning and sanitizing.

V. Cleaning Aids (mops, brooms, squeegees), Floor Mats, Condensate Wipers

a) Cleaning aids
   1) After each usage, cleaning aids must be thoroughly cleaned (including handles) with detergent solution and then stored in an appropriate container filled with a solution of 10% quaternary ammonia mixed 1 oz. to 1 gallon of water. (All cleaning aids' handles must be nonporous material to facilitate cleaning.) The "head" or usage part must be submerged in the sanitizing solution. The sanitizing container must have a drain and must be re-charged each day. Squeegees used on product contact areas must be stored in a separate sanitizer container.

b) Floor Mats (rubber, soft plastic or porous material)
   1) Must be thoroughly cleaned with detergent, rinsed and stored until used in a container with sanitizing solution (same as in V.a) 1).

c) Condensate Wipers
   1) Handles must be non-porous material.
   2) When not in use, store absorbent head in sanitizer same as in V.a) 1).
   3) Absorbent heads must be replaced with fresh heads a minimum of once a weekly.

VI. Hoses (air, water, sanitizer)

a) Air Hoses and Air
1) Air must be clean and dry through the use of dryers and water traps.
2) Air hoses should be of the coiled (like a telephone cord) type suspended so that when in use they cannot contact the floor.
3) Must be cleaned and sanitized each day during normal clean-up.
4) Air filters and traps must be sanitized at least weekly and drained daily.

b) Water Hoses
1) Must not be allowed to lay on the floor other than during clean-up.
2) After each usage, the hose must be hung on wall holder so that it does not contact the floor and must be sprayed with sanitizer as it is hung up.
3) Persons that handle ready-to-eat product that is not packaged must not be allowed to use hoses.

c) Sanitizer Hoses
1) Must be of a type (coiled or on a retractable drum) that will not allow them to contact the floor when in use.
2) Must be sprayed with sanitizer after each use paying particular attention to the outlet end that is being handled.

VII. Pipes

a) Any overhead sewer or drain pipes must be checked weekly for any leakage or seepage.

b) Any insulated pipes must be fully covered with impervious insulation covering. Any exposed insulation is to be replaced and properly covered.

VIII. Air Handling Systems

a) Air make-up units
1) All ductwork must be kept clean and dry on a continuing basis.
2) All filters are to be in place and changed regularly.
3) Check to make sure there are no bird nests or birds roosting on or near air intakes.

IX. Employee Practices/Facilities

a) Employees working on the "raw" side of the plant are prohibited from entering coolers or packaging areas used for ready-to-eat products. Conversely, employees from ready-to-eat side are prohibited from entering the raw side. A different color helmet or apron could be used to
distinguish each area.

b) All ready-to-eat product handlers must:
   1) Wash and sanitize their hands before going on the line after touching their hair or face, after picking up floor scraps, after adjusting equipment or handling non-product contact items.
   2) Wear hairnets that cover hair and ears. Snooods must be worn over beards.
   3) Put on disposable gloves, aprons and arm guards after hand washing and sanitizing. After putting gloves are put on, they must be dipped in sanitizing solution.
   4) Keep clipboards, etc. off product contact areas.
   5) Not put tape, labels, stickers, etc. on equipment as they can harbor moisture and be a breeding ground for microbes.
   6) Wear frocks and shirts.
   7) Spray and wipe down with clean paper towels, all control knobs with sanitizer before start, at breaks and lunch.
   8) Not use wooden handled knives. Stations when knives are not in use. Recharge sanitizer at lunch and shift changes.
   9) Place knives in a pan of sanitizer at individual workstations.
   10) Wash, sanitize and cover hands with fresh disposable gloves when opening reject packages.
   11) Leave frocks in department when going to break or lunch.
   12) Use company furnished knives and at end of shift leave them in the department for daily cleaning and sanitizing.

c) Mechanics
   1) Before working on ready-to-eat lines, the mechanic must wash and sanitize his hands and tools.
   2) After working on the line, the area or areas touched by the mechanic, his clothes or his tools must be sprayed with sanitizer and wiped down with clean paper towels.

d) Lunch Rooms, Locker Rooms, Restrooms
   1) Lunch Rooms - must be kept free of litter and table tops and chairs must be wiped down with sanitizer. Floor must be cleaned and sanitized daily.
   2) Locker Rooms - Floor should be washed and sanitized daily. Lockers should be wiped down with detergent and sanitized on a periodic basis.
   3) Restrooms - Floors, commodes, (including seats), urinals, sinks, etc., must be washed and sanitized daily. Commodes and urinals must be maintained in working order. If there is any overflow from a clogged
commode or urinal, the restroom is to be closed until washed and sanitized.

X. Fork Lifts, Trash Dumpsters

a) Fork Lifts - must not be taken from raw area to ready-to-eat or from ready-to-eat to raw area.

b) Trash Dumpsters - must not be taken from raw area to ready-to-eat or ready-to-eat to raw area.

c) Air lock door pull cords, electrical on/off buttons, must be sanitized daily (use caution).
APPENDIX 1

SUMMARY - TERMS

To summarize the basic compounds and cleaning terminology, the following quick reference will help:

<table>
<thead>
<tr>
<th>Class of Compounds</th>
<th>Major Functions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Basic alkalis</td>
<td>Soil displacement/emulsifying, saponifying and peptizing.</td>
</tr>
<tr>
<td>Complex phosphates and water</td>
<td>Soil displacement by emulsifying, peptizing, dispersion of soil, softening and prevention of soil depositions.</td>
</tr>
<tr>
<td>Surfactants</td>
<td>Wetting and penetrating soils, dispersing of soils and prevention of soil redepositions.</td>
</tr>
<tr>
<td>Cheating</td>
<td>Water softening, mineral control, soil displacement by peptizing and prevention of redeposition.</td>
</tr>
<tr>
<td>Acids</td>
<td>Mineral deposit control and water softening.</td>
</tr>
<tr>
<td>Sequestering/Cheating</td>
<td>The ability to prevent deposition of undesirable mineral salts on surfaces being cleaned.</td>
</tr>
<tr>
<td>Wetting</td>
<td>The ability to lower the surface tension of the water medium so as to increase its ability to penetrate soil.</td>
</tr>
<tr>
<td>Saponification</td>
<td>The chemical process of breaking down an animal (insoluble) or vegetable fat by alkali into soap (soluble) and glycerol.</td>
</tr>
<tr>
<td>Emulsification</td>
<td>This is a mechanical process. Fats are broken into tiny globules and are suspended in the cleaning solution. The alkalis and the complex phosphates are used for emulsification.</td>
</tr>
<tr>
<td>Term</td>
<td>Definition</td>
</tr>
<tr>
<td>-----------------------------</td>
<td>---------------------------------------------------------------------------</td>
</tr>
<tr>
<td>Synergism</td>
<td>When a chemical is used as a builder with a soap or detergent, the detergency which results from this combination is greater than the total detergency of the chemical and the soap is used independently.</td>
</tr>
<tr>
<td>Surfactant</td>
<td>A wetting agent or a compound reducing surface tensions.</td>
</tr>
<tr>
<td>Peptizing</td>
<td>The breaking up of solid particles into extremely small particles.</td>
</tr>
<tr>
<td>Dispersion or Deflocculation</td>
<td>The action of breaking up solid aggregates of soil into small particles.</td>
</tr>
</tbody>
</table>
APPENDIX 2

Seafood Microbiology

GENERAL

Every food has unique microbiological characteristics. Therefore knowledge of potential microbiological problems in one food does not guarantee that another type of food will have similar problems. It is important to be aware of the microbiological considerations for reasons of safety and food quality.

In general the flesh of seafood is more perishable than the meat of terrestrial animals. This is due to many seafoods having a moist surface, a relatively high pH (6.2) (bivalve mollusks have a high pH when fresh but it decreases during spoilage) compared to red meats and a normal microflora which is often psychrotrophic. In addition marine fish muscle has high levels of nutrients including nonprotein nitrogen compounds such as trimethylamine oxide (TMAO). As might be expected, microbial spoilage of seafood is mostly a proteolytic process (bivalve mollusks are an exception).

The flesh of freshly caught fish is normally free of bacteria. However bacteria may be found in high numbers on skin (log 2 - log 7/sq cm), gills (log 3 - log 9/g) and in the intestines (log 3 - log 9/g)(24). The types of bacteria are influenced by the harvesting environment. Fish from clean cold waters generally have fewer organisms than fish taken from polluted or tropical waters. Bacteria on the skin and gills are predominantly aerobic, whereas bacteria from the intestinal tract are principally anaerobic or facultative aerobic.

The organisms which comprise the microbial population are largely determined by their harvesting location. Bacterial populations of fish from temperate waters are mostly psychrotrophic, reflecting the cool temperatures of the sea bed (<10 C). Gram negative, rod shaped bacteria predominate this psychrotrophic population. Pseudomonas species, Moraxella - Acinetobacter species, Alteromonas, Cytophaga, Flavobacterium species and Vibrio species are usually the main organisms isolated. On the other hand, bacteria on fish from tropical areas are mostly mesophilic. A large percentage of this population may be Gram-positive and many researchers have reported that Micrococcus, coryneforms and Bacillus species may account for a large portion of the population.(IEMFS-1980)

It is often believed that the bacterial population of marine fish is halophilic. In actuality the organisms can usually grow in salt concentrations from <0.5 - 10%. The practice of icing
fish dilutes out many of the obligate halophils. However, many organisms on the fish may show improved growth when salt is added to growth media(1). Many of the spoilage bacteria present in fish can grow at higher salt concentrations than those normally encountered in red meat products. Of course cooking will destroy most spoilage bacteria.

A variety of bacteria are introduced during harvesting and processing. Organisms such as coliforms, Staphylococci, etc. may be introduced by contamination from decks, boxes, human hands and clothing. Processing further introduces terrestrial bacteria including coliforms and non-halophilic spoilage organisms.

In summary, fish tend to spoil more quickly than red meat products. The quality of fish is influenced by their environment and by handling practices. Spoilage is a proteolytic process. Most of the spoilage bacteria are heat sensitive and are killed by cooking. With proper training processors of red meat products should have no problem handling fish products.

SAFETY

Seafood-borne diseases are frequently divided into three categories based on the major source of the responsible agent; 1. agents naturally present in the environment; 2. agents derived from pollution of the aquatic environment; and 3. agents from workers, and processing and food service environments. Of the agents naturally present in the environment, members of the family Vibronacaeae and Clostridium botulinum type E are considered to be the most important bacterial pathogens. In addition, parasites and natural toxins are of great importance.

Vibronacae

Vibrios associated with food-borne illness

Historically, Vibrio cholerae 01 has been the Vibrio of greatest public health concern. All other species and serotypes were referred to as non-agglutinable or non-cholera Vibrios (Blake 1980, Desmarchelier 1984). However, Vibrio is a large genus containing 28 species plus numerous biotypes and chemovars (Oliver 1985). At least 11 species of Vibrio are recognized as pathogenic or potentially pathogenic to humans (Table A-1).

Vibrios may cause a variety of diseases including gastroenteritis, wound infection, ear infection, and primary and secondary septicemia (Table 1) (Morris and Black 1985). Of the eleven species pathogenic for humans only six are associated with foodborne disease; V. cholerae, V. hollisae, V. mimicus, V. parahaemolyticus, V. vulnificus, and V. furnisii. These
foodborne species will be the focus of this section.

**Vibrio cholerae:** is usually divided into serotype 01 and non-01 *V. cholerae*, which can be further subdivided as toxigenic and non-toxigenic. Toxigenic strains are capable of producing cholera toxin or a very similar toxin. *V. cholerae* 01 is the causative agent of endemic or asiatic cholera. The 01 serotype contains two biotypes; classical and El Tor, both of which may contain toxigenic and non-toxigenic strains. The classical biotype predominated world-wide until the 1960s, however, the El tor biotype is currently dominant and is the biotype associated with recent cases in the U.S. (Blake et.al., 1980, Levine 1981, Morris and Black 1985, CDC, 1986).

Symptoms of *V. cholerae* 01 infection can range from asymptomatic or mild diarrhea to severe cases (cholera gravis), which can cause profuse watery diarrhea, dehydration and death if not promptly treated. The incubation period varies from 6 hours to five days. Fortunately, cholera gravis is relatively uncommon. Cholera gravis results in 1-4% of infections from the El tor biotype and in 10-20% of the infections by the classical biotype. The infective dose for *V. cholerae* is estimated to be approximately one billion cells; however, consumption of antacids or medication to lower gastric acidity markedly lowers the infective dose (Blake 1987). Only toxigenic strains can cause cholera. Non-toxigenic strains of *V. cholerae* have been implicated in wound infections and can cause diarrhea but not cholera.

Cholera is relatively rare in the United States. The U.S. had no identified cholera outbreak from 1911 until 1973, then a single unexplained case occurred in Texas. A second cholera outbreak occurred during August, September and October of 1978 when 11 people were infected with *V. cholerae* 01 El Tor from recontaminated cooked crabs (Blake et.al., 1980). In 1981 there were two cases of cholera involving residents of the Texas Gulf Coast and 17 additional cases on an oil rig in the Gulf (Morris and Blake, 1985). Thirteen cases of domestically acquired cholera occurred in 1986; 12 in Louisiana and one in Florida (CDC 1986). Inadequate cooking or improper handling of crustaceans seems to have been the vehicle in this outbreak. Ten patients had severe diarrhea, with 7 requiring hospitalization. The *V. cholerae* 01 was of the El Tor biotype.

Cholera was once thought to be mostly a water-borne disease but in recent years its transmission by food has been well established (Kolvin and Roberts 1982). Outbreaks of cholera have occurred in several countries where food was the vehicle of transmission. Foods involved have included raw or partially cooked mollusks, other raw seafood, recontaminated cooked crabs and even raw vegetables. *V. cholerae* 01 has a relatively short
generation time and can grow quickly if foods are temperature abused.

V. cholerae 01 is widely distributed and is probably part of the indigenous bacterial flora in estuarine waters (APHA 1985, Clowell 1984). There is evidence of seasonal variation and most cases of domestically acquired cholera have occurred during the late summer and fall.

Non 01 V. cholerae: At least 70 other groups of V. cholerae are known to exist. They are referred collectively as non-01 V. cholerae or non-agglutinable (NAG) V. cholerae. The majority of the strains isolated from seafood and patients are non-toxigenic strains; less than 5% of the non-01 strains from human sources in the United States produce cholera toxin. The non-toxigenic strains are principally associated with gastrointestinal illness; but in the U.S. about one-third of the human isolates are from extraintestinal sources, including wound infection, ear infection and primary and secondary septicemia (Morris and Black 1985). Associated symptoms of gastroenteritis have included diarrhea (100% of cases, abdominal cramps (93%) and fever (71%). Nausea and vomiting occurs in 21% of the victims. The diarrhea may occasionally be severe; with as many as 20-30 watery stools per day (Morris and Black 1985).

Almost all of the cases of non-01 V. cholerae infections in the U.S. have been associated with eating raw oysters; but egg and asparagus salad and potatoes also have been vehicles for the bacteria. Considering the relative frequency of isolates from seafood, the incidence of illness is very low. There is evidence that victims often have an underlying liver disease, which might be a host factor for the disease. Also, in many cases the disease may not be severe enough to warrant medical attention, so the incidence goes unreported.

Non-01 V. cholerae strains are widely distributed in the United States, Asia and Europe. They occur most frequently in bays and estuaries with salinity in the area of 0.4-1.7% (Colwell and Kaper 1978) but also have been found in rivers and brackish inland lakes with salinity levels as low as 0.01%. Their presence in oysters and water samples show a seasonal variation with the highest numbers being isolated June-August (Madden et al. 1982). Non-01 V. cholerae are free living organisms and are part of the autochtonous flora.

V. parahaemolyticus was first associated with food poisoning in 1950 in Osaka Japan (Fujino et al. 1974). Since its discovery, it has been implicated in more than 1,000 outbreaks per year in Japan and accounts for 45-70% of that country's bacterial food poisonings. Food poisoning in Japan is usually related to the consumption of raw seafood during the warm months.
Typical symptoms include diarrhea (sometimes bloody), abdominal cramps, nausea, vomiting, headaches, fever and chills (Fujino et al. 1974). The infective dose for humans is between $10^5$ and $10^7$ viable cells; however, a decrease in stomach acidity may decrease infective dose. The time for onset of symptoms is usually 9-25 hours and the duration of the illness is usually 2.5-3 days. No deaths have been reported in the United States, but a death rate of 0.04% is reported for Japan. In Japan, raw seafood is the usual vehicle for the organism, but in the U. S. most of the foods implicated in <i>V. parahaemolyticus</i> outbreaks are cooked seafoods that have been recontaminated; although raw oysters and raw crabs have been implicated in some outbreaks (Barker 1974, Blake 1980, Spite et al. 1978).

<i>V. parahaemolyticus</i> is widely distributed in nature and has been isolated from coastal waters worldwide. Its presence has been documented in virtually all the marine coastal environs of the United States (Fujino 1974, Blake 1980). It is not considered a microorganism of the open sea because of cool temperatures and high hydrostatic pressure (Kaneko and Colwell 1978, Colwell 1984, Schwarz and Colwell 1974). Its presence in estuarine environments and in the seafood harvested from these environments usually shows a seasonal variation, being present in the highest numbers during the summer (Kenako and Colwell 1978, Hackney et al. 1980).

While <i>V. parahaemolyticus</i> is a common contaminant of seafood, often present in high numbers, almost none of the isolates from seafood are capable of causing gastroenteritis in man (Fujino et al. 1974, Blake 1980, Hackney 1981). The test most widely used to differentiate between virulent and avirulent strains is the Kanagawa reaction. Isolates from the marine environment and seafood are predominantly (> 99%) Kanagawa negative whereas food poisoning victims usually only excrete Kanagawa positive isolates. Studies have demonstrated that isolates do not change in the intestines and that Kanagawa positive types are probably part of marine <i>V. parahaemolyticus</i> populations, but present in low numbers.

<i>V. vulnificus</i>: Called the new "terror of the deep," <i>V. vulnificus</i> is one of the most invasive species ever described (Oliver 1985). It has been identified as a halophilic "lactose-positive" marine vibrio. Foodborne infection may result from eating contaminated, raw or undercooked seafood, particularly oysters and clams, with illness usually starting 16-48 hours after ingestion. The organism penetrates the intestinal tract and produces a primary septicemia. The illness usually begins with malaise, followed by chills, fever, and prostration. Vomiting and diarrhea are uncommon, but sometimes occur shortly after chills and fever. Hypotension (systolic blood pressure $\geq 80$ mmHg) is present in approximately a third of the cases (Blake et al. 1979). The infection progresses rapidly and may cause death
in 40-60% of patients (Oliver, 1985). Infection by *V. vulnificus* is usually associated with certain risk factors including: liver disease, gastric disease, malignancy, hemochromatosis and chronic renal insufficiency. (Oliver 1985, Blake et al, 1979). **INFECTION DOES NOT OCCUR IN NORMAL HEALTHY PEOPLE.** The most common vehicle for the organism is raw oysters, and most of the cases have occurred along the Gulf coastal states.

*V. vulnificus* is widespread in the environment and has been isolated from estuarine waters of most coastal states. Infection via the intestinal tract is most often associated eating raw oysters. Its presence in water and shellfish is seasonal being most prevalent when the water temperature is high (>20°C). Low salinity (0.5-1.6%) also favors the presence of *V. vulnificus* in seawater (Kelly 1982). Some strains of *V. vulnificus* show bioluminescence and these strains may also be pathogenic (Oliver 1986). Environmental isolates are phenotypically indistinguishable from clinical isolates and produce virulence factors identical to clinical isolates (Tison and Kelly 1986).

*V. mimicus* is biochemically similar to *V. cholerae*, with the exception that the strains are sucrose negative. In the past, they were listed as *V. cholerae* of the Hieberg group 5; however DNA homology studies demonstrated that many of the sucrose negative strains were a separate species and in 1981 the name *mimicus* was proposed because of their similarity to *V. cholerae* (Shandera et al. 1983, Colwell 1984). Both toxigenic and non-toxigenic strains have been isolated, however, the food poisoning cases have been mostly from the non-toxigenic strains. Symptoms include diarrhea in most cases, and approximately 67% of the cases had nausea, vomiting and abdominal cramps. Diarrhea may be bloody and will last 1 to 6 days.

Raw oysters and boiled crawfish (crayfish) have been implicated as vehicles for the organism. *V. mimicus* is widely distributed in nature and can be found in fresh and brackish waters. It does show seasonal variation, with the highest numbers in the warmer months (Bockemuhl et al. 1986, Colwell 1984).

*V. hollisae* (formerly EF 13) has been implicated in relatively few cases of food poisoning. Symptoms have included diarrhea and in approximately half the cases vomiting and fever. Seafood in including raw oysters, clams and shrimp (Morris et al. 1982) is implicated as the vehicle for *V. hollisae*. Its ecology is not well understood because it grows poorly or fails to grow in TCBS, the medium most used to isolate members of the *Vibrio* genus.

*V. furnissi*, previously classified as biovar II of *V. fluvialis*, has been implicated in food borne illness (Brenner et
al. 1983). It produces gas from glucose, which is an unusual characteristic among Vibrio species. Symptoms include diarrhea, abdominal cramps, and sometimes nausea and vomiting.

**Pleisomonas shigelloides**

P. shigelloides (formerly Aeromonas shigelloides) has been implicated in human gastroenteritis for 40 years (Miller and Koberger 1985). Symptoms of the infection include diarrhea (94%), abdominal pain (74%), nausea (74%), chills (49%), fever (37%), headache (34%), and vomiting (33%) (Miller and Koberger 1985). Symptoms usually appear 24-50 hours after ingestion of the food. Foods implicated as vehicles for P. shigelloides include cuttle fish salad, salt mackerel, raw oysters and undercooked oysters. In the U.S. raw oysters are probably the most implicated food.

P. shigelloides is widespread in nature, being mostly associated with fresh surface water, but may also be found in seawater. It is more often isolated during the warmer months (Miller and Koberger 1985). Most strains of P. shigelloides have a minimum growth temperature of 8°C, but at least one strain has been reported to grow at 0°C. The organism is sensitive to pH of <4 and salt concentrate of >5% (Miller and Koberger 1986)

**Aeromonas hydrophila:**

A. hydrophila is a marine bacterium which has been implicated in human disease. It will grow at refrigeration temperatures and may increase during storage of shell stock shellfish and other seafood. Symptoms of illness include diarrhea and abdominal pain often accompanied by nausea and vomiting. The organism is very wide spread in nature and is found in many foods besides seafood. Oysters are the most often vehicle implicated, but other seafoods are possible sources.

**Effect of Processing on Survival of Vibrionaceae**

Members of the family Vibrionaceae are sensitive to many of the methods used to process food. All are relatively sensitive to heat, acid, radiation and subfreezing temperatures. All are killed rather quickly by temperatures of 70°C or greater radiation doses of between 50,000 - 100,000rads, and most are sensit-subfreezing temperatures. Aeromonas hydrophila and V. cholerae are exceptions and survive freezing well.

**Clostridium botulinum**

Human botulism is relatively rare; however, the control and
prevention of botulism is one of the most important considerations in food processing. History has shown repeatedly that an outbreak of botulism can cause severe, often ruinous economic problems for processors. When problems arise, a whole section of the food industry is often affected, in addition to the processor involved (Eklund 1982, Eyles 1986).

_C. botulinum_, the etiological agent of botulism, is divided into eight types, based on serological differentiation of the neurotoxin, A, B, C₁, C₂, D, E, F and G (Sakaguchi 1979). The types have been divided into 4 groups by Smith (1977) according to proteolytic activity. Group I and II are the most important with respect to human botulism. Group I includes type A and proteolytic strains of type B and F. This group is strongly proteolytic and produces putrid, unpleasant odors. This group produces highly heat resistant spores and has a minimum growth temperature of about 10°C. Group II includes all type E and non-proteolytic strains of B and F. Group II is neither proteolytic nor gelatinolytic and cultures do not produce putrid odors in food. This group can grow at temperatures as low as 3.3°C and the spores of members of this group are heat liable.

_C. botulinum_ is widely distributed in soils and all types may be isolated from the aquatic environment (Dolman 1964). Type E is the most frequently isolated from aquatic environments and is most often implicated in botulism associated with seafood products. The spores of type E are most often isolated from fresh water and marine sediments in temperate zones (Dolman 1964).

Most outbreaks of botulism associated with fishery products have implicated semi-preserved products, i.e., smoked, salted or fermented products that are eaten without further cooking (Eklund 1982, Lynt et al. 1982). Type E is inhibited by water activity of <0.975 (5% NaCl) and pH value of less than 5.3. (Emodi and Lechowich, 1969). The spores are sensitive to heat. Decimal reduction times at 82.2°C (180°F) range from 0.49 minutes to 6.6 minutes, depending upon the heating medium and the strain (Lynt et al 1982, Simonovic et al. 1985). The spores are most resistant in tuna packed in oil. For foods not packed in oil a D value of 4.3 minutes at 82.2°C is usually considered the maximum. Z values range from 4.8°C to 9.6°C (Simonovic et al 1985). For comparison, other members of Group II produce slightly more heat resistant spores with D-values for nonproteolytic type B ranging from 1.49-32.3 minutes at 82.2°C. The D-values for nonproteolytic F are similar to nonproteolytic B. Most of the heat processes applied to seafood during processing will destroy _C. botulinum_ type E.

Other forms of preservation processes may not provide safety. Radurization will kill spoilage bacteria but not destroy spores or toxin of type E (Eklund 1982; Skulberg 1965). Modified
atmosphere storage, coupled with temperature abuse may create unsafe conditions (Garcia et al. 1987). Post et al (1985) used inoculated and uninoculated fillet and demonstrated the formation of toxin population before or simultaneous with sensory rejection. Therefore storage at a temperature of <3.3°C is critical.
Table 1. Diseases Associated With *Vibrio* Species

<table>
<thead>
<tr>
<th>Organism</th>
<th>Gastro-enteritis</th>
<th>Wound infection</th>
<th>Ear infection</th>
<th>Septecemia 1°</th>
<th>Septecemia 2°</th>
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<td><em>Vibrio alginolyticus</em></td>
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<td><em>Vibrio cincinnatiensis</em></td>
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<td><em>Vibrio damsela</em></td>
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<td><em>Vibrio fluvialis</em></td>
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<td><em>Vibrio furnissii</em></td>
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<td><em>Vibrio hollisae</em></td>
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<td><em>Vibrio metschnikovii</em></td>
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<td><em>Vibrio mimicus</em></td>
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<td><em>Vibrio parahaemolyticus</em></td>
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<td><em>Vibrio vulnificus</em></td>
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* most common clinical presentation
• other clinical presentations
(•) very rare presentation
Listeria monocytogenes

The most common manifestation of monocytosis in man is meningitis in infants. Also, abortions, especially in the first trimester, and septicemia in pregnant women is common. In adults the illness is often described as flu-like. (Smith et al. 1987, Bryan, 1979) L. monocytogenes, the causative agent, is a small coccoid Gram-positive motile rod which may resemble Corynebacterium or Streptococcus species. The temperatures for growth of L. monocytogenes range from 1-45°C with the optimum being between 30 and 37°C. At 4-5°C it's growth rate is 50 times slower than at 35-37°C, but it can still grow. The optimum pH for growth is near natural to slightly alkaline. The pH range for growth is 5.0 - 9.6. L. monocytogenes is quite tolerant to salt and low water activity. It can grow in the presence of 10% NaCl and survive at 25% NaCl. (Smith et. al. 1987, Medallion, 1987, Bryan 1979)

The number of cells that are needed for infection is fairly low, but the exact infective dose has not been determined. An intracellular parasite within the lymphocytes, Listeria exerts its influence on the brain and central nervous system, the gaud uterus, placenta and fetus. Victims of Listeria are generally either comprised (i.e., had a malignancy, receiving immuno suppressive drugs, severe alcoholics) or pregnant (Smith et al.).

L. monocytogenes has been isolated frequently from meat, seafood and poultry products. Recently the Center for Disease Control has determined that epidemiological evidence indicates that meat products may indeed be vehicles for listeriosis and that there is reason for concern when it is isolated from ready to eat products including ready to eat seafoods.

Listeria monocytogenes is widespread in nature. It has been isolated from pond reared trout and from crustaceans. A high part of the sea gull population carries Listeria and their feces often contaminates the environs of seafood processing plants. Listeria also is widespread in the environment and can be found in soil, vegetation, crop debris and, of course, fecal material. It appears to be around part of the saprophytic flora of grass and other plants.

Staphylococcus aureus

S. aureus is the second or third leading cause of food poisoning in the United States. The illness is an intoxicification resulting from consuming food in which the bacteria has grown and produced toxin. Symptoms include nausea, vomiting, abdominal cramps and diarrhea. The onset time for symptoms averages 2-4 hours, but may range from 0.5 to 2 hours. The illness usually
only lasts 1-2 days. (Berdgoll 1979, Jay 1987)

The toxin responsible for illness is released during growth and has been well characterized. There are six enterotoxin types, which are single polypeptide chains with molecular weights of between 27,000 and 34,000. The enterotoxins are heat resistant and can withstand temperatures above boiling for long periods of time. (Bergoll 1979)

Conditions affecting S. aureus growth are often quite different than those for toxin production. In general, the relative ranges for toxin production are narrower than those for growth. For example, the temperature range for growth of S. aureus is between 10-45°C (some strains can grow at 7°C) with an optimum at 35-37°C. Toxin production is maximum at the optimum, but very little toxin is produced at lower temperatures (<20°C). The pH range for growth is 4.5 - 9.3, but enterotoxin production is limited to between 5.1 to 9. The optimum pH for both growth and toxin production is near natural. Also the water activity minimum for growth and toxin production is different; 0.85 and .93, respectively.

The amount of enterotoxin produced will vary with the type of enterotoxin and the strain; the amount of toxin needed for illness varies, but values of between .015 - .357 µg/kg body weight are most often reported. As little as 1 µg of enterotoxin A has been reported to cause illness. In food, the S. aureus concentration needed to produce this much toxin is between 100,000 and 1,000,000 cells per gram.

S. aureus is a common bacterium found in all humans, but the carrier rate for enterotoxin producing strains is 6-60% with an average of 25-30% of the population being positive. The main reservoir is the nose, but it is also found on skin, hands, wounds, boils, etc. It also can be isolated from the air, dust, floors and other environmental surfaces. It survives well in the environment. It is often isolated from ready to eat seafood products and has been responsible for seafood-related outbreaks of illness. The organism comes from the workers and lack of temperature control allows it to grow to dangerous levels.

**Salmonella**

Salmonella is either the first or second leading cause of food poisoning in the United States (Jay 1987). Gangarosa (1978) estimates that there are more than 2.5 million cases of salmonellosis per year, resulting in hospitalization of 500,000 persons and over 9,000 deaths. Gangarosa estimated that in 1978 Americans spent over $1.2 billion on medical treatments for salmonellosis. Assuming the number of cases remained constant, the inflation adjusted figure for medical treatment would now
exceed $2 billion. However, according to the Center for Disease Control, the incidence of Salmonella food poisoning among the general population has been increasing; therefore, the costs are probably far greater.

The food poisoning syndrome develops after ingestion of a food that contains a sufficient number of Salmonella cells to cause infection, usually between 100,000 and 100,000,000 cells. The symptoms usually develop 12-14 hours after ingestion of the food, although incubation times of greater than 24 hours are not uncommon. Symptoms consist mainly of diarrhea along with nausea, vomiting, abdominal pain, headaches and chills. The symptoms are often accompanied by prostration, muscle weakness, moderate fever and drowsiness. Symptoms usually last only 2-3 days. The death rate is less than 0.2% (Jay 1987).

Raw foods, particularly those of animal origin, are the major vehicles for salmonellosis (Cox and Bailey 1987, Allred et al. 1967). The five most common food vehicles for Salmonella in the United States are beef, turkey, homemade ice cream (containing eggs), pork and chicken (Jay, 1987, Cox and Bailey 1987). Turkey is the most common vehicle in Canada. However, many other foods have been involved in salmonellosis. For example, in 1985, the largest outbreak ever reported (18,000 cases) was traced to pasteurized milk.

Salmonella is often isolated from seafood products; however the numbers are usually low. Salmonella is of concern in many pond reared aquacultured products. The closed environment and the presence of birds often leads to salmonellae contamination. The numbers are usually very low, but care must be taken to avoid temperature abuse. Cooking will kill the organisms.

**Yersinia enterocolitica**

_Yersinia enterocolitica_ is a Gram-negative nonspore-forming rod belonging to the family Enterobacteriaceae. The bacterium is interesting in that incubation temperatures affect the characteristics of the organism. For example, it is motile when grown at 28°C but not when grown at 37°C. Also, its nutritional requirements vary with incubation temperature. The temperature range for growth is from 0°C to 41°C. It is psychrotrophic and grows well at refrigeration temperatures. In foods such as milk, beef and pork, it can grow to infectious levels in 4-14 days at refrigeration temperatures. (Stern, 1982, Jay 1987)

The clinical manifestation of _Y. enterocolitica_ varies somewhat with the age of the victim. _Yersinia_ gastroenteritis is characterized by acute abdominal pain, fever, diarrhea, vomiting and often pseudo-appendicitis. Milk has been the vehicle for many of the outbreaks. (Stern 1982)
Y. enterocolitica is wide spread in nature. It has been isolated from swine (which appears to be a major reservoir - the highest isolation rate is from the tonsils and tongue), humans, dogs, rodents, cows, sheep, horses, deer, and other animals. It has also been isolated from oysters in low levels (personal communication, Harold Kator, Virginia Institute of Marine Science). The investigators do not know of any cases where seafood has been implicated in Y. enterocolitica food poisoning.

**Escherichia coli**

*Escherichia coli* is often thought of more as an indicator of fecal pollution than as a pathogen, but it can be both. It is most often associated with the intestinal tract of warm blooded animals and man. Its use as an indicator for food and water is well established and many standards are based on *E. coli* counts. For example, fresh crabmeat must contain less than 3.6 *E. coli* /g as determined by the standard most probable number procedure.

However, some strains of *E. coli* are pathogenic. These fall into four categories; 1) enterotoxigenic, 2) enteropathogenic, 3) hemorrhagic and 4) enteroinvasive. (Medallion, 1987, Mehlman, 1982)

Enterotoxigenic strains can produce both heat stable and/or heat labile toxins (Lt). The Lt toxin is similar to cholera toxin. The enteropathogenic strains have been implicated in infant diarrhea; mostly in non-food outbreaks. The enteroinvasive strains produce dysentery similar to shigellosis. The hemorrhagic strain (0157:H7) causes a hemorrhagic colitis-bloody diarrhea. Hemorrhagic strains are not invasive.

*E. coli* is a mesophile with a growth range of between 10-47°C. It is very wide spread in nature, being present in the feces of warm-blooded animals. It often finds its way into food through unsanitary conditions. It can grow quickly if the conditions are suitable.

**Campylobacter jejuni**

*Campylobacter jejuni* is a Gram-negative-microaerophilic motile rod. It may be the first or second leading cause of food poisoning in Western countries including the United States. Only recently has its importance been realized because methodology to detect the organism in food and feces was not available. (Doyle 1981)

*C. jejuni* causes a gastroenteritis with diarrhea being the symptom. In severe cases the diarrhea may be bloody, accompanied
by high fever and severe cramping. The incubation period for onset of the symptoms is normally seven days and the illness normally lasts 2-7 days. The infective dose may be as little as 500 cells.

*C. jejuni* is a somewhat fragile microorganism, easily destroyed by acids, sanitizers, and drying. It does not grow at refrigeration temperatures. *C. jejuni* temperature range for growth is 32-45°C.

The main reservoir for *C. jejuni* is the intestinal tract of animals and man. Turkeys and chickens are almost 100% positive for *C. jejuni*. Other meat animals also frequently harbor the bacterium. (Doyle, 1981, Jay 1987)
References


Center for Disease Control. 1986. Cholera in Louisiana - Update. MMWR. Dept. Hlth. and Human Ser. Atlanta, GA.

Center for Disease Control. 1986. Toxigenic *Vibrio cholerae O1 Infection - Louisiana and Florida. MMWR. Dept. Hlth. and Human Ser. Atlanta, GA.


Gangarosa, E.J. 1978. What have we learned from 15 years of Salmonella surveillance? In "National Salmonellosis Seminar". Washington, DC.

Garcia, G., Genigeorgis, C. and Lindroth, S. 1987. Risk of growth and toxin production by Clostridium botulinum non-proteolytic types B, E, and F in salmon fillets stored under
modified atmospheres at low and abused temperatures. J. Food Protect. 50:330.


Thompson, C., and Vanderzant, C.  1976. Serological and hemolytic characteristics of Vibrio parahaemolyticus from marine sources. J. Food Sci. 41:204


SANITATION

Test Questions:

All questions are answered True or False.

1. The two best ways to control bacteria in seafoods is to control their initial numbers and their rate of growth.

2. Bacterial reproduce by dividing in two.

3. HACCP is the abbreviation for Hazard Analysis Critical Control Point.

4. Cleaning is a process which will remove soil and prevent accumulation of food residues, which may decompose to support the growth of disease or spoilage organisms.

5. Psychrotrophs are organisms which will grow at 0°C, but their optimum temperature is higher than 15°C, usually between 20-30°C.

6. Fluctuations in temperature during processing can greatly affect the shelf-life of a product. For example, if a filet is left at room temperature for 3-4 hours, the shelf-life will be reduced by days.

7. Food safety and quality problems are most often caused by contaminated raw food or ingredients; improper holding temperatures (time-temperature abuse); improper cooling of foods (failure to cool the seafood to refrigeration temperatures within 2-4 hours); improper handling of the seafood after processing; cross contamination (between products or between raw and processed product); infrequent breakdown and cleaning of equipment; inadequate plant design to separate the raw product from the cooked product; and/or poor employee hygiene and poor sanitation practices.

8. No food processing establishment has an ideal water supply, therefore, the cleaning compounds must be tailored to the individual water supply and the processing operation.

9. Water is the major component in the majority of cleaning solutions; it acts as both a solvent and as a carrier for the various ingredients in the cleaning solution.

10. The hardness or softness of water is related to the amount of calcium and magnesium present, the higher the concentration, the "harder" the water.
11. There are two types of hardness, temporary and permanent; hardness is temporary when it can be removed by boiling the water, whereas permanent hardness can only be removed by chemical treatment.

12. It is important that cleaning and sanitization be a two step process; the use of sanitizers, without proper cleaning is usually a waste of money and time.

13. Soil is defined as matter out of place.

14. The variables of time, temperature, concentration and mechanical action affect cleaning regardless of the object to be cleaned.

15. The correct amount of cleaning solution usually does an excellent job, but too much can greatly decrease effectiveness.

16. The term detergent is quite broad, referring to any agent that will remove one undesirable substance from another.

17. An effective detergent must have the following properties: rapid penetrating and wetting power; ability to control water hardness; high detergent power to remove soil; suspending power to keep the removed soil from redepositing on the surface being cleaned; easy rinsability; and non-corrosiveness to surfaces being cleaned and to cleaning equipment.

18. Saponification is the chemical conversion by alkali of water insoluble fatty acids soils into more soluble substances - soaps.

19. The most important principle concerning sanitizing/disinfection is that a dirty surface can not be sanitized.

20. Chlorine and chlorine-base sanitizers are the most commonly used sanitizers in the seafood industry.

21. In general the flesh of seafood is more perishable than the meat of terrestrial animals.

22. Seafood - borne diseases are frequently divided into three categories based on the major source of the responsible agent; 1. agents naturally present in the environment; 2. agents derived from pollution of the aquatic environment; and 3. agents from workers, and processing and food service environments.

23. Vibrios may cause a variety of diseases including gastroenteritis, wound infection, ear infection, and primary and secondary septicemia.
24. Infection by *Vibrio vulnificus* is usually associated with certain risk factors including; liver disease, gastric disease, malignancy, hemochromatosis and chronic renal insufficiency. Infection does not occur in normal healthy people.

25. Members of the family *Vibrionaceae* are sensitive to many of the methods used to process food including heat, acid and radiation.