

**Thermal Processing of Foods**  
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**Lecture 1**  
**Food Microbiology: Concerns in Various Foods**

Good morning everyone, today we are going to see about food micro biology, microbial growth and concerns in various food.

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## Bacteria

- Bacteria are the most important microorganisms to the food processor. Most are harmless, many are highly beneficial (*Lactobacillus Acidophilus*, *Streptococcus Thermophilus*), some indicate the probable presence of filth (transmission of *salmonellosis*), disease organisms, spoilage (*Clostridium Perfringens* and *Bacillus Cereus*) and a few cause disease (*Clostridium Botulinum*).
- Thousands of species, single-celled (spherical, straight rods, and spiral rods)
- Reproduce by dividing into two cells. Under ideal conditions, doubling may occur every 15 mins.

## Bacteria

- Some rod-shaped bacteria exists in two forms: Dormant spores and active vegetative cells.
- Vegetative cells form spores under adverse conditions as a means of survival and spore forms preserve the bacteria from starvation, drying, freezing, chemicals, and heat. When conditions become favorable, the spores germinate, with each spore again becoming a vegetative cell with the ability to reproduce.
- Most sporulating bacteria that grow in the presence of air belong to the Genus Bacillus, and most that grow only in the absence of air belong to the Genus Clostridium.

So before going into that lecture, the thermal processing of food, though it talks about what are all the thermal processing applicable in for the food processing. Actually the thermal processing is employed to increase the shelf life of food products, for commercial production and also it paves the way to how processed food to store it for long time. So, it does not only depends upon the thermal processing of the food, it also involves thermo bacteriology and what are all the thermal processing methods available and how to design such process designs and how to validate them in the commercial sterilisation or pasteurization method. So everything includes in the thermal processing. So without thermal bacteriology I cannot design any thermal process to increase the shelf life of the food.

So, the thermal bacteriology or food microbiologists very much important before we go into detail about various thermal processing methods available for the food processing. So, in that concern in this class we are going to review about various microorganisms available and their concern on various food and how the food spoilage occurs and everything in big detail. Okay so the microorganisms, the moment we say then it includes micro bacteria, yeasts and molds as well as the viruses. So, the most important microorganism of food concern is bacteria. So most of the bacterias are harmless and many are highly beneficial for human, human beings. So lactobacillus; one such as Lactobacillus and Streptococcus Thermophilus. So, it mostly available in the dairy products.

Some indicate the probable presence of filth which is nothing but transmission of salmonellosis which is a disease, which mainly spreads through flies or cockroaches. And some are the reason for disease causing our spoilage which is nothing but example is Clostridium Perfringens and Bacillus Cereus. And few cause disease as well as, those were Clostridium Botulinum, okay. So, in (bac) bacteria there are thousands of species available and they are single celled micro organisms and they available in the spherical, straight, rods or spiral rods. And they reproduce by dividing into two cells, under ideal conditions the doubling time is about 15 minutes. So, each 15 minutes it goes doubling.

The rod shaped bacteria exist in two forms, one is the dormant spores and another one is active vegetative cells, okay. So the vegetative cells forms spores under adverse conditions as a means of survival and spore forms preserve the bacteria from starvation, drying, freezing, chemicals and heat. So these are adverse conditions for the bacterial growth. So, what is vegetative cells do is,

they form a spores and keep it as a reserve, when the favourable conditions comes they start forming, becoming a vegetative cell then it starts its reproduction. So when conditions favourable the spores germinate and each spore again become a vegetative cell with the ability to reproduce.

So, this conversion of spores to vegetative cell and vegetative cells to spores is nothing but a conversion of its form, it is not a pure reproduction so after forming the vegetative cell only it can reproduce. So most of the sporulating bacteria that grow in the presence of air belongs to the Genus Bacillus, most of the Bacillus bacterias were sporulating bacteria, they grow in the presence of air and most that grow only in the presence of air is the Clostridium Genus. So they are anaerobic bacterias.

And one more thing I would like to emphasis here is, we are going to see very basics of what are all the microorganisms available and there concern in various food but I would suggest you to read more about all these organisms and their concerns in various foods with proper food micro biology book, okay. So here we are only going to review certain important points which we need to know before designing any thermal processing.

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## Yeasts and Molds

- Yeasts (*Saccharomyces Cerevisiae* and *Aspergillus Niger*) are oval-shaped and slightly larger than bacteria.
- In budding each cell can produce several buds, or swellings, which break away to form new, fully formed daughter cells.
- Molds as found on bread, fruit, damp paper, or other surfaces are actually composed of millions of microscopic cells joined together to form chains, which have numerous branches, called hyphae.

## Yeasts and Molds

- Molds can thrive in conditions too adverse for bacteria or yeasts. They reproduce by spores that are frequently present as green or black masses on the protruding hyphae.
- Yeasts and molds grow on most foods, where there are small amounts of nutrient and moisture (dry foods, salted fish, bread, pickles, fruits, jams, jellies, and similar commodities). They predominate high salt or sugar environments
- Bacteria find conditions of low pH, moisture, or temperature and high salt or sugar unfavorable.

So, next we will see about the yeasts and molds. The yeasts the example is, Saccharomyces, Cerevisiae and Aspergillus Niger. So this is most common, so you might have known already, are oval shaped or slightly larger than bacterial cells. They reproduce by budding. So budding each cell can produce several buds or swellings which break away to form new, fully formed daughter cells. So the reproduction happens here by budding. Molds as found on bread, fruit, damp paper or other surfaces are actually composed of millions of microscopic cells which are joined together, form as a chain and which have numerous branches called hyphae, right.

So they form if you see, if you might have seen in the bread surface, the black masses or green masses, so which are nothing but a cells joined together to form a chain and which also have a branches like hyphae. Molds can thrive in conditions too adverse for bacteria or yeasts, okay. So they produce by spores again, the molds that are frequently present as a green or black masses on the protruding hyphae. Hyphae is nothing but branches. So, yeasts and molds grow on most foods where there are small amounts of nutrient and moisture.

It is opposite to bacteria. So the yeasts and molds grow on dry fruits, salted fish, bread, pickles, fruits, jams, jellies, and similar commodities. So they predominant high salt and sugar environments. Who? Yeasts and molds but for bacteria, they are adverse conditions. Bacteria find conditions of low pH, low moisture, or low temperature and high salt and sugar are unfavourable. But yeasts and molds grow in high salt and high sugar conditions.

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## Viruses

- Viruses are the smallest and simplest microorganisms.
- They are parasitic and they must first invade the cells of another living organism called the host, before multiply.
- Viruses which infect bacteria, called bacteriophages, cannot infect human beings or other animals. On the other hand, several animal viruses, known as zoonosis, can infect human beings.

## Viruses

- Viruses require a live host cell and cannot multiply in food.
- Bacteriophage infections of starter cultures can interfere seriously with the manufacture of cheese, buttermilk, sauerkraut, pickles, wine, beer, and other desirable fermentative products.
- Although viruses require a live host cell and cannot multiply in foods, they can remain viable and infectious for long periods of time, even under highly adverse conditions, such as drying, freezing, and pasteurization.

Then comes the viruses, so viruses are smallest and simplest microorganism. Actually they are parasitic and they must need another living organisms to first invade and reproduce. They are called the living organism, with which they invade first is called a host cell, then after that only they can multiply. So viruses which infect bacteria are called bacteriophages. so they cannot infect human beings or other animals.

On the other hand, several animal viruses known zoonosis, so can infect human beings. So viruses they cannot themselves reproduce, they before multiplication they need a host cell, so if those host cells are bacterias they called as, they called bacteriophages. So they would not infect

the human beings but certain animal viruses are there, so they can infect human beings. Actually in the food also, it requires a live host cell without which they cannot multiply in the food. So the infections of two categories, one is the bacterial phage infection, so they are of starter cultures which can interfere seriously with the manufacture of dairy products which of cheese, buttermilk, sauerkraut, pickles, wine, beer and other desirable fermentative products, they may create an infection.

Although viruses require a live host cell, they cannot multiply in food. So they can remain viable and infectious for a long period of time and even under highly adverse conditions such as drying, freezing and pasteurization. So in these conditions they can remain viable and infectious for a long time. But they occur definitely a host so they cannot do on independently, okay. So that is all about the basic microorganisms, bacteria, yeasts and molds and viruses. So, what are all their shapes and which kind of foods they find it favourable conditions for the growth.

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### **Factors Affecting Growth of Microorganisms**

The food processor reduces potential problems from microorganisms in several ways:

- ✓ Removing or destroying them by trimming, washing, heating, pickling, by adding chemicals, or by encouraging competition by acid- or alcohol-forming organisms.
- ✓ Minimizing contamination from equipment, people, the environment, and from unprocessed food.
- ✓ Minimizing microbial growth on equipment, by cleaning and sanitizing, and in the product itself by adjusting storage temperature, pH, and other environmental factors.

### Factors Affecting Growth of Microorganisms

- ✓ Psychrophiles grow only at refrigeration temperatures.
- ✓ Psychrotrophs grow well at refrigeration temperatures (Grow best from 58 - 68 °F), but better at room temperature.
- ✓ Mesophiles grow best at or near human body temperature (Grow best from 86 - 98 °F), but grow well at room temperature.
- ✓ Thermophiles (optimum: 122 - 150 °F; spores can survive 250 °F for 1+ hr) grow only at temperatures about as hot as the human hand can endure, and usually not at all at or below body temperature.

So then we are going to see what are all the factors affecting growth of microorganisms in the food, right. There are three ways, one is removing or destroying them by trimming, washing, heating, pickling, by adding chemicals or by encouraging competition between acid and alcohol forming organisms. Actually the first one is before going for any thermal processing the pre-treatment would be refer for example canning operations, so if you want to can any food, so the previous operations were these, I mean trimming, washing, the heating is nothing but they call it as a blanching. So, it is done for many reasons, one is to inactivate enzymes as well as to remove the microorganisms.

So, these to be done prior to food storage, or food processing, any food processing. So that way we can reduce the growth of microorganisms and sometimes it is done by using addition of chemicals or sometimes we play with the pH so that competition between the acid and alcohol forming organisms because we are going to see what are all the favourable pHs, temperature and water activity condition for the growth of microorganisms. So we can control in such a way that, so the pH and temperature or water activity is not favourable for the growth of the microorganisms. And some of these pre-treatment methods also we can control the growth of microorganisms.

And another thing is minimizing contamination from equipment, people and the environment and from unprocessed food, right. So, though the main two thermal processing equipments were pasteurization and the sterilization, even after the sterilization, the storage and the handling, for

example we are going for high treatment process, so there may be if the equipment is not properly washed or properly sterilized before employing the food for further thermal processing, so that may also contaminate the food.

And the handling, the personnel should be aware of the good sanitation practises and the environment everything also affect the growth of microorganism. And minimising microbial growth on equipment by cleaning and sanitizing and in the product itself by adjusting the storage temperature, pH and other environmental factors. For example, we do pasteurization process to kill the pathogenic microorganism but it is the pasteurization temperature is not enough for the, it only kill the microorganism but if it goes to normal temperature environment then there may contamination of microorganisms again.

So those pasteurized products should be refrigerated for further to increase the shelf life of the food. So these factors also have to be taken care because though we process the food by required thermal processing, then further storage temperature and pH and environmental factors also affect the growth of microorganisms and sometimes they serve us a favourable conditions for the microorganisms to grow, though it was processed thermally before storage. So, based on that the bacterias were divided into four categories, one Psychrophies, they can grow only at refrigeration temperatures. The second one is Psychrotrophs; they grow well at refrigeration temperatures but better at room temperature.

And the third category is Mesophiles which grow best or or near human body temperature, so most of the organisms are most of the organisms which found in the food is Mesophiles but grow well at room temperature. The fourth category is Thermophiles. So which has a optimum temperature of 120 to 150 degree Fahrenheit. Theirs spores can survive around 250 degree Fahrenheit for more than one hour, they grow only at the temperatures about as hot as the human hand can endure and usually not at all or below the body temperature. So the Thermophiles can survive at hot conditions and it cannot survive below the body temperature which is nothing but a 98 degree Fahrenheit, okay.

So there are four categories, Psychrophies, which grows at refrigeration, Psychrotrophs which which also grow well at refrigeration but better at room temperature, Mesophiles which grow well at room temperature and it can up to the human body temperature which is nothing but 98



degree Fahrenheit, Thermophiles survive at hot conditions but they cannot survive at below body temperature.

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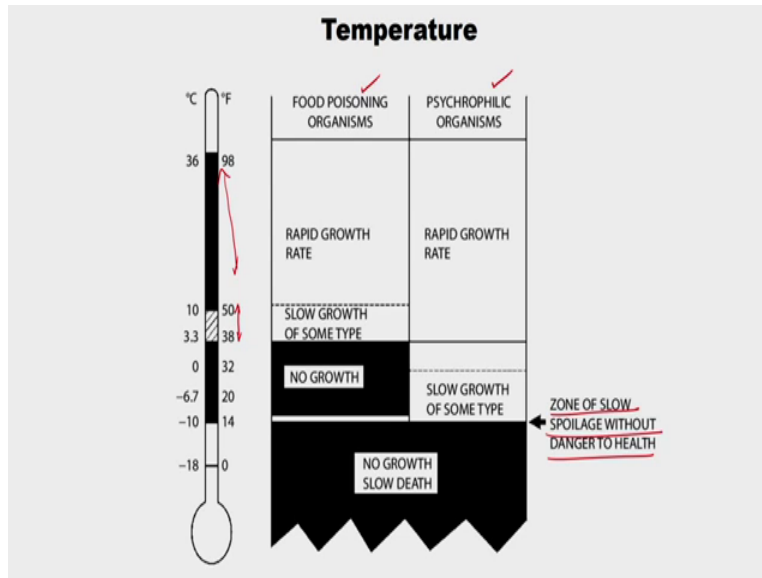
### Temperature

- Some psychrotrophic microorganisms grow very slowly in foods below freezing, but usually not below 19°F. Standard storage temperature for frozen foods, 0°F, does not permit microbial growth.
- Most psychrotrophs have difficulty growing above 90°F.
- Most foodborne disease organisms are mesophiles. A good rule of thumb is to store perishable foods below 40°F or above 140°F.
- In the temperature range where both mesophilic and psychrotrophic organisms grow (about 41°F to about 90°F), the psychrotrophs grow more rapidly, causing spoilage and at the same time frequently interfering with the growth of foodborne disease organisms
- The rate of growth increases rapidly as the temperature is raised.

Though we can classify all the bacterias in any of the four categories but there are some exceptions. Some psychrotropic microorganisms grow very well very slowly in foods below freezing but usually not below 19 degree Fahrenheit. Standard storage temperature for frozen foods are 0 degree Fahrenheit which does not permit microbial growth. And most psychrotrophs have difficulty growing above 90 degree Fahrenheit. Most of the foodborne disease causing microorganisms are Mesophiles. I have already mentioned you. The general rule of thumb is to store perishable foods below 40 or above 140 degree Fahrenheit to suppress the growth of microorganisms.

In the temperature range where both mesophilic and psychrotrophic organisms grow, the psychrotrophs grow more rapidly causing spoilage and the same time frequently interfering with the growth of food borne microorganisms. Actually if both mesophilic and psychrotrophic organisms grow, the temperature window, which is nothing but 41 to 90 degree, that particular temperature condition is favourable for both mesophilic as well as psychrotrophic but the psychrotrophics grow more rapidly compared to mesophilic and it causes the spoilage at the same time frequently interfering with the other foodborne disease organisms. Their rate of growth always increases rapidly as the temperature is raised.

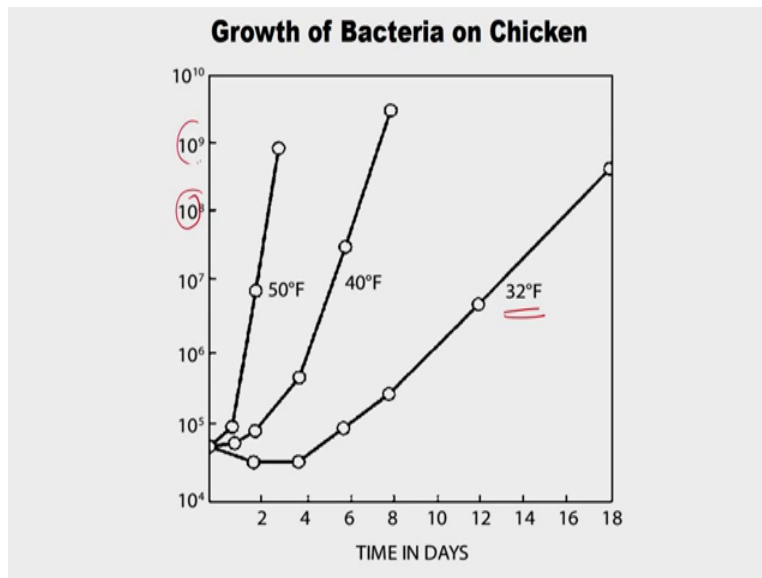
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So this is just a comparison where food poisoning microorganisms versus psychrophilic microorganisms. So in the temperature range of 50 to 98 degree Fahrenheit, so where food poisoning microorganisms will have rapid growth rate, the same with psychrophilic microorganisms but the temperature range between 38 and 50, food poisoning microorganisms show slow growth of some type and still you will find rapid growth for the psychrophilic. So, psychrophilic the temperature range is 38 to 98 degree almost.

So your food poisoning microorganisms found no growth around 14 to 38 degree or 35 degree Fahrenheit which founds no growth and if we see psychrophilic microorganisms, they found slow growth rate. But below 14 degree Fahrenheit no growth of both, the slow death happens. So this is the zone of slow spoilage without any danger to health. It is just a comparison how the temperature window of its various microorganisms.

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So this is another example, growth of bacteria on the chicken. So if you see the 32 degree Fahrenheit, it finds almost 18 days, so the growth is around 10 to the power of 8, okay. So, but if you increase the temperature then the growth stops within 8 days and if you further increase to almost 50 degree Fahrenheit, it is around 2 to 3 days, the maximum is 10 to the power of 9.

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### Water Activity

- Water activity ( $a_w$ ) is a term describing the availability of water to microorganisms. It is only roughly related to percent moisture.
- The GMP regulations for low-acid canned foods defined water activity as the vapor pressure of the food product divided by the vapor pressure of pure water under identical conditions of pressure and temperature.
- The regulations define low-acid foods as foods, other than beverages, with a finished equilibrium pH value greater than 4.6 and a water activity greater than 0.85  $a_w$  for bacteria > yeasts > molds.  $pH = 7$   
< 7  
> 7
- Certain molds and bacteria can grow on fish immersed in saturated salt solution where the  $a_w$  is about 0.75
- Some molds can grow in foods with  $a_w$  0.62 - 0.65
- At these lower limits, growth is very slow (crackers or sugar is about 0.10)
- The stability of intermediate moisture foods ( $a_w = 0.75 - 0.90$ ) such as dried fruits, jams, and soft moist pet foods, depends on combinations of factors, such as low  $a_w$ , low pH, pasteurization, chemical additives, and impervious packaging.

So the next important factor is water activity. So water activity is nothing but a availability of water to the microorganism. It is only the roughly related to the presence of moisture. So the

GMP regulations, GMP regulation is nothing but a good manufacturing practices even I told about the previous slides, the equipment; how the contamination happens in the equipment.

So these regulations to be followed, even how to clean the equipment and where to keep the equipment, and the food processes to be kept in which area and environment conditions, everything comes from this GMP regulations that is good manufacturing practices regulations. For low-acid canned foods defined water activity as the vapour pressure of the food product divided by vapour pressure of the pure water under identical conditions of pressure and temperature. So the regulation defines low-acid foods such as foods other than beverages with a finished equilibrium pH value greater than 4 point 6, right.

Low-acid food means normally pH, anyway we are going to see pH is also one of the factors affecting. So pH 7 means, so that is neutral, so less than 7 that is acid, greater than 7 that is alkali condition. So in the less than 7 acid conditions also there are two categories, one is low acid and high acid. So the high acid is below 4 point 6 and low acid food is above 4 point 6 and water activity greater than point 85. So the regulations defines the low-acid foods, so this is the favourable conditions for the *Clostridium botulinum* to grow. So that produces the toxins and also the diseases, botulinum, so we are going to see anyway, what are all the symptoms and related food spoilage.

So the conditions for low-acid food is pH should be greater than 4 point 6, at the same time water activity should be greater than point 85. So if you compare the water activity, bacteria it is high and then there comes the yeast and the last one is molds. Molds we have already seen. It requires minimum water as well as the nutrient condition; even with that they can grow. Certain molds and bacterias can grow on fish immersed in saturated salt solution. So we told that already the yeasts and molds can grow at high salt, even high sugar environment where the (act) aw is about point 75 and some molds can grow with the water activity of point 62 to point 65. At these lower limits, the growth is very slow; if it has to grow in the presence of sugar then the water activity of about point 1 is itself enough.

The stability of intermediate moisture foods, which is nothing but point 75 to point 9 which also includes point 85, such as dried fruits, jams and soft moist pet foods depends upon the combination of factors. Though we say that water activity of point 85 is favourable for the

bacteria to grow, it also depends upon other factors which is low pH, the pasteurization, chemical additives, how much, what all are the chemical additives we add and the impervious package. So it is not only, we can say that if you maintain this particular water activity where spoilage can occur but it also depends upon other factors.

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<b>Water Activity</b>	
The water activity ( $a_w$ ) limits for growth of principal foodborne disease organisms	
Microorganism	Minimal $a_w$ for growth
<i>Salmonella</i>	0.945 ✓
<i>Clostridium botulinum</i>	0.95 ✓
<i>Clostridium perfringens</i>	0.93 ✓
<i>Staphylococcus aureus</i> ✓	0.86 ✓
<i>Vibrio parahaemolyticus</i>	0.94 ✓

So the water activity limits for growth of principal foodborne disease organisms. So the minimal  $a_w$  is given; for *Salmonella* species it is point 945, for *Clostridium botulinum* it is point 95, the *Clostridium perfringens* it is point 93, *Staphylococcus aureus* point 86 and *Vibrio* species it is point 94. So this is we told that minimal  $a_w$  for the growth of microorganisms. So to toxin to provide, for example it may also produce the toxins which causes the further diseases, so that activity may vary little bit from the growth conditions. For example, *Staphylococcus*, if it can grow at the water activity of point 86 but it starts producing its toxins at for say point 92 water activity. So these both are, both may be different.

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<b>Water Activity</b>	
The water activity ( $a_w$ ) limits for growth of principal foodborne disease organisms	
Microorganism	Minimal $a_w$ for growth
<i>Salmonella</i>	0.945 ✓
<i>Clostridium botulinum</i>	0.95 ✓
<i>Clostridium perfringens</i>	0.93 ✓
<i>Staphylococcus aureus</i> ✓	0.86 ✓
<i>Vibrio parahaemolyticus</i>	0.94 ✓

So and is a pH and initial population count; what is a effect of pH and initial population count on the food products; growth of microorganisms on the food products. So this we have already discussed; what is less than 7, equal to 7 and greater than 7. So as we already told, many bacteria grow best at about neutral pH, they grow poorly or not at all grow below pH 4, alright. The favourable is 7 but still it can grow in the low-acid food but below 4 it the probability is very much low. Yeasts and molds therefore predominant in low pH foods where bacteria cannot compete.

This we have seen from the first slide onwards because the bacteria, yeasts and molds require opposite conditions to grow. The lactic acid bacteria can grow in high acid foods and actually produce acid to give a sour milk. So the lactic acid bacteria, so this we might have seen if we keep the milk at high temperature for particular period, it become a sour milk. So the pickles, fermented meats and similar products also suffer with lactic acid bacteria. Some strains called *Leuconostoc* species which contributes off-flavours to orange juice, orange juice is highly acidic but the flavour will go if the, if these species are found in that orange juice.

And one more things is, actually the GMP suggest that, that 12 log reduction, we will talk about that when we talk about the kinetic parameters, we talk about this also that thermal death time. So they call it as a, for particular temperature to reduce the microorganism count for one log, right. How long it requires; so that is the time which is nothing but the thermal death time.

So normally, the practice what the food microbiologists follow is the 12 log reduction, so how long, what is at particular temperature, if 12 log reduction happens, to happen 12 log reduction how long it requires.

So that is the practice they follow but still if the initial count is high then there may be a possibility for the microorganisms to be there in the food, right. That is what is initial population count. So this is the major importance to the processor of refrigerated food the shelf life of which is enhanced by good sanitation. So a high level of spores also increases the possibility that a few will survive to spoil the heat process to products.

That is what I told, so even though the best practices say 12 log reduction at the particular temperature, but if we have high level of spores so it is nothing but a probability, right. So we cannot kill all the spores, it is 90 percentage or 99 point 9 percentage only, even that point 1 or point 001 percentage can cause the spoilage. So high level of spores increases the possibility that few of them will survive even after it is thermally processed.

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**pH**

Mean pH Values of Selected Foods	
pH Value	Selected Foods
2.3	Lemon juice (2.3), Cranberry sauce (2.3)
3.0	Berries (3.0 – 3.9), Peaches (3.7), Orange juice (3.7), Apricots (3.8)
4.0	Cabbage red (4.2), Pears (4.2), Tomatoes (4.3)
4.6	<b>Meats, Most vegetables, Most starch based foods, and Most protein-heavy foods</b>
5.0	Spaghetti in tomato sauce (4.9), Figs (5.0), Onions (5.2), Green Beans (5.3), Potatoes (5.5)
6.0	Tuna (5.9), Codfish (6.0), Beef (6.0), Pork (6.1), Evaporated milk (6.1), Chicken (6.2), Corn (6.3)
7.0	Milk (6.8), Ripe olives (6.9)

So these all are pH values of selected foods, so the pH values about 2 point 3 range, so lemon juice and cranberry sauce; and 3 about berries, peaches, orange juice, apricots, and 4 around cabbage red, pears, tomatoes, and 4 point 6 this is what we told, low-acid food, so this is the favourable conditions for Clostridium botulinum. So this can happen in meats, most of the

vegetables, starch based foods and protein heavy foods. So 5 is nothing but spaghetti in tomato sauce, figs, onions, green beans and potatoes.

6 around tuna, cold fish, beef, pork, evaporated milk, chicken, corn; 7 is around milk and ripe olive. So if you know whatever the examples I have taken is for chicken, for the bacterial growth and milk, most of them, like the bacterias, so affected. So their favourable conditions are around neutral pH and the 4 point 6 is favourable for Clostridium species.

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**pH**

**The minimal pH minimal for growth of principal foodborne disease organisms**

Microorganism	Growth reported at but not below pH
<i>Staphylococcus aureus</i>	4.5
<i>Salmonella</i>	4.0 ✓
<i>Clostridium botulinum</i> : Types A and B	4.8 ✓
<i>Clostridium botulinum</i> : Type E	5.0 ✓
<i>Clostridium perfringens</i>	5.0 ✓
<i>Vibrio parahaemolyticus</i>	4.8 ✓
<i>Bacillus cereus</i>	4.9 ✓

The minimal pH minimal for principal foodborne disease organisms is *Staphylococcus aureus*. So if you see this is growth reported it but not below this pH. So below 4 no 5 this *Staphylococcus aureus* cannot growth, the *Salmonella* it is 4, *Clostridium botulinum* there are A, B, E, F types; A, B types 4 point 8 and *Clostridium botulinum* type E it is 5. So see point 2, so the control of pH is very much important, within point 2 we will get different types. And *Clostridium perfringens* it is again 5 and *Vibrio* species it is 4 point 8 and *Bacillus* species it is 4 point 9. So if you see it is mostly between 4 point 5 to 5. So these are all principal foodborne disease causing microorganisms.



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### Oxygen

- Oxygen is essential for growth of some microorganisms: Aerobes, anaerobes and microaerophilic.
- Strict aerobes grow only on food surfaces and cannot grow in foods stored in cans or in other evacuated, hermetically sealed containers.
- Anaerobes grow only beneath the surface of foods or inside containers.

So the next one is, the next factor affecting is oxygen. So this is essential for growth of some organisms so based on that we divide into three categories: one is aerobes, anaerobes and microaerophilic. Microaerophilic needs little oxygen, they can grow in little oxygen conditions, so that means aerobes as well as anaerobes. So strictly aerobes grow on the food surfaces and cannot grow in the food stored cans or in other evacuated, hermetically sealed containers. So normally hermetically sealed containers, when we see canning operations so we would be seeing so how to remove the air which is contained in the cans.

So because aerobes need oxygen for their growth, they may not grow in the cans, foods stored in the cans or in other evacuated or hermetically sealed containers. Anaerobes grow only beneath the surface of the food or inside the containers but when microorganisms are involved, for example, they need little oxygen mostly are anaerobes. But certain organisms can grow in both the conditions but in such case if the air is found, their growth will be higher compared to anaerobic conditions. That means they will be seen in the food surfaces.

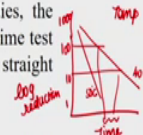
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## Lethal Effects of Temperature

- Most practical and effective means to destroy microorganisms.
- Cell reduction occurs just above maximal growth temperatures and the rate of death increases markedly as the temperature is raised.
- Pasteurization, the destruction of vegetative cells of disease-causing microorganisms, consists of a temperature of 140°F for 30 minutes, or about 161°F for 16 seconds. Yeasts, molds, and the vegetative cells of spoilage bacteria also die at pasteurization temperatures.
- Commercial sterility is the destruction and/or inhibition of the organisms of public health significance as well as organisms of non-health significance which could spoil the product.
- Low-acid foods requires temperatures above 212°F. Canners process certain canned foods at 240°F or 250°F (121°C) for a considerable length of time (usually 15 or 20 minutes), sometimes an hour or more

## Lethal Effects of Temperature

- In thermal destruction studies, also called <sup>D<sub>T</sub></sup> thermal death time studies, the logarithm of the numbers of survivors is plotted against the length of time test cultures are subjected to a given temperature. The result is usually a straight line.
- The slope of this line becomes steeper as the temperature is increased, indicating that less time is required to kill a population at higher temperatures. It also takes longer to kill a high population of organisms than it does to kill a low population.
- The rate of thermal destruction is greater in foods with high a<sub>w</sub>, than in those with low a<sub>w</sub>.
- The recommended pasteurization process to destroy Salmonella in liquid egg albumen prior to freezing is 140°F (60°C) for 3.5 minutes, whereas that for dried egg albumen is 140 (60°C) to 158°F (70°C) for several days.
- Presence or absence of organic matter, oil or fat, pH, strain of organisms, quality of available nutrients, and age of the culture



### Lethal Effects of Temperature

- Chilling to temperatures below the growth range, but above freezing, stops reproduction but kills few cells except for extremely sensitive organisms, such as vegetative cells of *Clostridium perfringens*
- Freeze kills part of a microbial population within a few hours and storage continues to be lethal at a much slower rate, which depends on nature of the food. Ex: Rapid drop in aerobic plate count (“total count”) occurred in orange juice
- Bacterial spores die very slowly, if at all, during freezing and frozen storage. For example, the vegetative cells of *Clostridium perfringens* generally all die, but the spores survive.
- *Staphylococcus aureus* and related organisms survive well
- Freezing is not a dependable means to destroy microorganisms since some cells of the original population almost always survive

So then lethal effects of temperature. The most practical and effective means to destroy microorganism is nothing but a thermal processing. So heat it to higher temperatures, so when you increase the temperature then the growth of the microorganisms will be decreasing. So the cell reduction occurs just above the maximal growth temperatures, right, growth temperatures in the sense, for example it can grow at say, 42 degree Fahrenheit, so if you increase the temperature above than that then the cell reduction starts occurring and the rate of death increases markedly as the temperature is raised.

So, when you increase the temperature, the rate of death increases. There are two main thermal processing is involved, one is pasteurization, another one is a sterilization. The pasteurization is nothing but a destruction of vegetative cells of microorganisms which are disease-causing which consist of a temperature of 140 degree Fahrenheit for about 30 minutes or 161 degree Fahrenheit for 16 seconds. So this time-temperature combination is also designed by the kinetic processing. So we will see in the subsequent lectures. So various processing needs, various time temperature combinations and we say, low temperature less time and high temperature less time and high temperature high time.

So there are many possible ways to come up with time-temperature combination based on the food quality, nutrient quality and bacterial deactivation, so that we will see in the next few lectures. But here I would like to mention two examples, if we are maintaining 140 degree Fahrenheit it is about the processing should be done about 30 minutes but if it is 161 degree

Fahrenheit it is almost 16 seconds are enough. And yeasts, molds and the vegetative cells of spoilage bacteria also die at pasteurisation temperature. So remember vegetative cells, if there is, as we have seen already, they may form a spores and they can survive pasteurisation temperature.

When the favourable conditions occurs, they may start converting into vegetative cells but one thing we should remember is after pasteurisation the storage is important; it should be stores in the refrigerated conditions. The commercial sterility is the destruction and/or inhibition of the organisms of public health significance as well as the organisms of non-health significance which could spoil the product. So, here mostly we take care of the yeast, molds or vegetative cells but sterilization is the higher order thermal treatment. So here we destruct as well inhibit the organisms of public health significance which both disease causing otherwise the spoilage organisms of the food.

So the sterilised product which does not require the further storage conditions, it can survive in the room temperature about particular time period, maybe 6 months to 1 year. The low-acid foods require temperature above 212 degree Fahrenheit. Canners process certain canned foods at 240 to 250 degree Fahrenheit, it is about 121 degree centigrade for a considerable length of time usually 15-20 minutes, sometimes an hour or more. This an hour and more so if you reduce the temperature, the time-temperature combination, there are many.

So normally, the low-acid foods requires 121-degree centigrade sterilization temperature for about 15-20 minutes. As I mentioned you already, there is DT that is nothing but a thermal death time. So which is nothing but a logarithmic numbers of survivors plotted against the length of the time so this is time versus log reduction of your microorganisms. So for example, 1, 10, 100, 1000 kind of thing. So one log reduction, so how much time it requires at constant temperature. So temperature is constant here.

Subjected to here given temperature, the result is usually a straight line, we normally get, so this to this, so this is one log reduction from 100 to 10 to reduce it how long how much time it requires. Actually we say that it is a straight line but there are exceptions where we would not get this as a straight line. The slope of this line becomes steeper as the temperature is increased, for example if it is, this is say around 40 degree centigrade. So if I increase 50 then my line becomes

this because you require very less time. This I already mentioned so if you increase the temperature you would require less time to kill the same population.

Required to kill population at higher temperature, it also takes longer to kill a high population of organisms. For example, here we put 100, so if I go for 1000 to 10, right. So it requires longer time, because my initial starts with 100 then I may require 15 minutes, initially itself if I have 10 to the power of 3 number of spores then it requires more time. The rate of thermal destruction is greater in foods with high water activity than in those with low water activity; the rate of thermal destruction. So one example is recommended pasteurisation process to destroy Salmonella species in liquid egg albumen prior to freezing, before freezing they do pasteurization. So that is if we maintain 60 degree for 3 point 5 minutes.

So if it liquid egg, whereas for dried egg albumen it is the say 140 degree or 60 degree centigrade to 158 or 70 degree for several days. So that means if we have high water activity, the rate of thermal destruction is higher compared to low water activity. So it can be, if it is a liquid egg it is 3.5 minutes but if it is dried egg albumen, it takes several days so the temperature also have to be bit higher. The presence of, presence or absence of organic matter, oil or fat, pH, strain of organisms, quality of available nutrients, age of the culture; so these all also affect the time temperature combination.

For example, so not only I need to see how much log reduction I will be able to get in a particular time temperature combination so if I heat that particular temperature, that long hours so what happens to my nutrients in the food and what is the pH alteration during the time temperature combination, what is the strain of microorganisms I have what is the age of culture. So everything else is also matter when we fix the time temperature combination. And one more thing is another thing what we think is like we always talk about the higher temperature.

So what happens if we store it in a chilling because I already told the pasteurisation, after pasteurisation we need to preserve it in the refrigeration temperature. So what happens during chilling? Chilling to temperatures below the growth range but above freezing stops the reproduction, but kills few cells except for extremely sensitive organisms such as vegetative cells of Clostridium perfringens. So it stops reproduction but kills few cells only but there are some certain sensitive organisms which still survives even at its chilling temperatures. I already told

though we give example but one thing is the probability of the spores present in the food and also there are many factors which affects and sometimes the closely related species also will have different effect in the particular temperature.

The freeze kills part of microbial population within a few hours and storage continues to be lethal at much slower rate which depends upon the nature of the food. For example, rapid drop in aerobic plate count occurred in orange juice, so if I keep orange juice in the chilling conditions then aerobic plate count we are going to see; this is nothing but a qualitative count of the microorganisms. So there is a drop in aerobic plate count in the orange juice. So actually bacterial spores die very slowly in the chilling temperature, if at all, during freezing and frozen storage, for example, the vegetative cells of *Clostridium perfringens* generally all die but the spores survive.

I already told vegetative cells may die here but spores may survive even at the chilling temperature. And *Staphylococcus aureus* and related organisms still survive well in the chilling temperature range. Actually freezing is not a dependable means to destroy microorganisms since some cells of the original population almost always survive. So the freezing comes like, before freezing normally we do either of the thermal processing; pasteurisation or sterilization, mostly pasteurisation. So as I already told, even after pasteurisation there are many factors which affects the survival of the microorganisms after the thermal processing as well. So freezing is not a favourable condition to destroy microorganisms. It may be used for storage, further storage after the particular thermal processing is employed.

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### Aerobic Plate Count

- Aerobic Plate Count (APC) measures only that fraction of the bacterial flora that is able to grow to visible colonies under the arbitrary test conditions provided in the time period allowed.
- High APC reveals:
  - ✓ Failure of sorting, trimming, washing, and destroying operations to remove or destroy bacteria from raw ingredients adequately.
  - ✓ Inadequate heat processing.
  - ✓ Insanitary equipment, particularly near the end of the process.
  - ✓ The food has reached or is approaching the end of its refrigerated shelf-life.
  - ✓ The food has been stored at or above room temperature for too long.
  - ✓ The food is at least partly decomposed.  
Ex: *Coliform Bacteria*

So what is this aerobic plate count, so this is aerobic plate count means it is a fraction of the bacterial flora that is able to grow as a visible colonies under the arbitrary test conditions provided in the time period allowed. So particular time period, how much visible colonies I am able to see at particular test conditions. So if the test conditions are changed then there may be you will get the other number of counts. So the high APC, high aerobic plate count reveals; so it is the one of the measuring technique.

Failure of sorting, trimming, washing, destroying operations to remove or destroy bacteria from raw ingredients adequately. As I told, for example if we take canning operations so before that they do some of the pre-treatment; one of the pre-treatment is blanching, right. So although it was done to inactivate the enzymes, sometimes it is also kills certain microorganisms. So if we still find after those operations, if we still find APC count high then that means that was not done properly though if it is meant to reduce the microorganisms count then it should not give high APC. And inadequate heat processing; sometimes what happens, though we, the time temperature combination is not adequate enough to reduce the microorganisms to the particular log level.

And insanitary equipment, particularly near at the end of the process, near at the end of the process. I already told though after processing is done, even GMP regulations are just that near the walls also you need to maintain the sterilization temperature or pasteurisation temperature.

So there is a possibility, the temperature is lower than the particular processing temperature then there may be a possibility of the contamination. And the food has reached or is approaching the end of its refrigerated shelf life and sometimes what happens is, they say that even it should be used at the refrigerated storage but the condition is 3 days or 4 days, right.

So at that time also we need to probably check in the refrigerated conditions also what is the time period or what is the shelf life of the refrigerated product. The food has been stored at or above room temperature for too long. For example, if we see, if something says that, if we go for shop and buy some refrigerated food products, if they say that it should be kept at refrigerated temperature. We come home and we keep it some 6 hours in the normal temperature then we remember and keep it in the refrigerator so that storage in the room temperature at 6 hours also counts, also effects on some of the microorganisms which grow on the food.

The food is at least partly decomposed; partly decomposed in the sense even though it is already spoiled but we store it in refrigeration that will not improve anything. The refrigeration is nothing but we can restrict the growth of microorganisms, if already there it cannot do anything about it. So one is Coliform Bacteria; so this available in all the dairy products so if the conditions are not maintained properly then this spoilage occurs due to this bacteria.

One thing I would like to mention is here is the high, whatever the points we discuss, it also lies with the manufacturers, for example if it has to be stored under refrigerated condition so it should be legibly written in the carton, I mean the label, and also used by date is very much important because one thing what we discussed is an end of its refrigerated shelf life. Even in the refrigeration condition, it cannot be stored for most one year or two years, right, there is a use by date for that, even for refrigerated products.



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## Food Poisoning

- Human illnesses caused by foodborne microorganisms are popularly referred to as food poisoning.
- Apart from illness due to food allergy or food sensitivity, foodborne illness may be divided into two major classes, food infection and food intoxication.
- ✓ Food infection results when foods contaminated with pathogenic, invasive, food poisoning bacteria are eaten. These bacteria then proliferate in the human body and eventually cause illness.
- ✓ Food intoxication follows the ingestion of preformed toxic substances which accumulate during the growth of certain bacterial types in foods.
- The period of time between the consumption of contaminated foods and the appearance of illness is called the incubation period. It can range from less than one hour to more than three days, depending on the causative organisms or the toxic product.

The next one is food poisoning, human illness caused by foodborne microorganisms are popularly referred as a food poisoning. Apart from illness due to food allergy or food sensitivity, we have this right for example I am allergic towards a high fat food or I am allergic to high oily food, so apart from these two they may be divided into two major classes, one is food infection, another one is food intoxication. The food infection results when foods are contaminated with pathogenic, invasive, and food poisoning bacteria are eaten, right the bacteria itself we had it and these bacteria then proliferate in the human body, mostly intestine and eventually cause illness. So I have eaten the bacteria itself which causes the disease.

The second one is food intoxication which happens, for example the microorganisms grows in the food and we have discussed, right certain pH there is a toxins are produced by the microorganisms. So the toxins were produced by the microorganisms so I have eaten those toxic substances, right. So the second one is the ingestion of preformed toxic substances which accumulate during the growth of certain bacterial types in food, so that is food intoxication, the earlier one is food infection. The period of time between the consumption of contaminated foods and the first appearance of my illness is nothing but a incubation period, for example, morning I have eaten, after two hours I started vomiting. So the in between time is nothing but a incubation period. It ranges from less than one hour to more than three days, depending upon the causative organisms or the toxic product.

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Characteristics of the important bacterial food intoxications and foodborne infections			
Disease	Etiologic Agent	Incubation Period	Symptoms
Botulism ✓	<i>Clostridium botulinum</i> A,B,E,F toxin ////	Usually 1 to 2 days; range 12 hours to more than 1 week	Difficulty in swallowing, ✓ double vision, difficulty in speech. Occasionally nausea, vomiting, and diarrhea in early stages. Constipation and subnormal temperature. Respiration becomes difficult, often followed by death from paralysis of muscles of <u>respiration.</u>
Staphylococcal food poisoning	Staphylococcal enterotoxin	1 to 6 hours; average 3 hours	<u>Nausea, vomiting, abdominal cramps,</u> diarrhea, and acute prostration. Temperature subnormal during acute attack, may be elevated later. Rapid recovery-usually within 1 day.

Characteristics of the important bacterial food intoxications and foodborne infections			
Disease	Etiologic Agent	Incubation Period	Symptoms
salmonellosis	Specific infection by <i>Salmonella</i> spp.	Average about 18 hours; range 7 to 72 hours	Abdominal pains, ✓ diarrhea, chills, ✓ fever, frequent vomiting, prostration. Duration of illness: 1 day to 1 week. ✓
Shigellosis (bacillary dysentery)	<i>Shigella sonnei</i> , s. <i>flexneri</i> , s. <i>dysenteriae</i> , s. <i>boydii</i>	Usually <u>24 to 48</u> hours; range 7 to 48 hours	Abdominal cramps, fever, chills, ✓ diarrhea, watery stool (frequently containing blood, mucus, or pus), spasm, headache, nausea, dehydration, prostration. Duration: a few days.

Characteristics of the important bacterial food intoxications and foodborne infections			
Disease	Etiologic Agent	Incubation Period	Symptoms
Enteropathogenic <i>Escherichia coli</i> infection	<i>Escherichia coli</i> serotypes associated with infant and adult infections	Usually 10 to 12 hours; range 5 to 48 hours	Headache, malaise, fever, chills, diarrhea, vomiting, abdominal pain. Duration: a few days.
<i>Clostridium perfringens</i> food poisoning	<i>Clostridium perfringens</i>	Usually 10 to 12 hours; range 8 to 22 hours	Abdominal cramps and diarrhea, nausea, and malaise, vomiting very rare. Meat and poultry products usually involved. Rapid Recovery. ✓
<i>Vibrio Parahaemolyticus</i> food poisoning	<i>Vibrio Parahaemolyticus</i>	Usually 12 to 14 hours; range 2 to 48 hours	Abdominal pain, ✓ severe watery diarrhea, usually nausea and ✓ vomiting, mild fever, chills and headache. Duration: 2 to 5 days.

So certain things we discuss here, as I told this is botulism, the disease, the etiologic agent which causes is the *Clostridium botulinum*. So there are A, B, E, F type toxin. So usually the incubation period is 1 or 2 days, range 12 hours to more than 1 week. So the symptoms are swelling, double vision, difficulty in speech, occasionally nausea, vomiting and diarrhoea. In early stages, constipation and subnormal temperature, respiration becomes difficult, often followed by death from paralysis of muscles of respiration. So this is a dangerous one and Staphylococcal food poisoning, so they caused by Staphylococcal enterotoxin, so the incubation period is 1 to 6 hours, again nausea, vomiting, cramps and temperature subnormal during acute attack, maybe elevated later, rapid recovery usually within a day.

Another is Salmonellosis, I already told it is spread by cockroaches, flies and specific infection by salmonella species, average about 18 hours, range from 7 to 17 hours. Again abdominal pain, diarrhoea, fever, frequent vomiting, then during of illness is 1 day to 1 week. Then shigellosis (bacillary dysentery), so this caused by *Shigella* species, usually 24 to 48 hours, again here abdominal cramps, fever, chills, diarrhoea and headache, nausea, dehydration, these are all symptoms. And *Escherichia coli*, I already told coli bacteria, this is *Escherichia coli* serotypes associated with infant and adult infections.

So usually 10 to 12 hours, headache, fever, and diarrhoea, vomiting, duration is few days, it will be rewind. *Clostridium perfringens* food poisoning, so the agent is *clostridium perfringens*, usually 10 to 12 hours, so abdominal cramps, vomiting is very rare, nausea is another symptom,

meat and poultry products usually involved this kind of bacterias, rapid recovery is possible. Then vibrio species, it is again virus, usually 12 to 14 hours; abdominal pain, and watery diarrhoea, usually, nausea and vomiting, mild fever also so the recovery period is around 2 to 5 days.

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### Food Spoilage

- Problem facing the food industry is simple spoilage by bacteria, yeasts, or molds that are not hazardous to health. Chilling slows spoilage; proper freezing, drying, canning, and pickling arrest it completely.
- Chilled foods must be transported to the consumer before spoilage microorganisms make them unfit for consumption.
- **Refrigerated Foods**
  - ✓ Products are convenient to use and have a “close to fresh” image ✓
  - ✓ The optimum storage temperature is 33°F. or as close to freezing as possible. However, most refrigerated cases are holding near 45 or even 45°F.
  - ✓ First of all, always assume pathogenic organisms are present in a food product.
  - ✓ Secondly, refrigeration temperatures may slow or prevent replication of most pathogenic microorganisms, but some will continue to multiply (psychrotrophs).
  - ✓ Thirdly, manufacturers should expect some temperature abuse of the foods during storage and distribution; this includes handling at the consumer level.

Then the food spoilage, actually the problem facing by the food industry is spoilage by the bacteria, though I have already told the thermal processing applied in two folds, one is spoilage bacteria to control growth of spoilage bacteria, another one is disease causing bacteria, alright. So the spoilage is of every food industry concern which is not hazardous to health. The chilling slows spoilage, proper freezing, drying, canning and pickling arrest it completely. Chilling I already told, so we need to employ a thermal processing and further the growth is controlled by chilling and proper freezing, drying, canning also do it completely to arrest the food spoilage microorganisms.

Chilled foods must be transported to the customers before spoilage microorganisms make them unfit for consumption. I already told if some food product should be kept at refrigerated condition, so it should be transferred immediately, so in between the transport and storage at maximum time without the chilling temperature, also makes the food to spoil. Refrigerated foods products are convenient to use have a “close to fresh” image. As we already told, the refrigerated

conditions are maintained for safety but it is most of the time to maintain the product quality, so it gives a “close to fresh” look.

The optimum storage temperature is 33 degree Fahrenheit or as close to freezing as possible, however most refrigerated cases are holding near 45 or even 45 degree Fahrenheit. There are 3 things we need to remember, even at refrigerated food, first of all we have to assume pathogenic organisms are always present in the food product. The second one is the refrigeration temperatures may slow or prevent replication of most of the pathogenic microorganisms, but still some will continue to multiply, example is psychrotrophs, we have already seen how even at refrigerated temperature.

Thirdly, manufactures should expect some temperature abuse of the foods during storage and distribution; this includes handling at the customer level as well. So manufactures always need to take care, for example, so from the shop to home or where ever from the place and there is a transportation and storage also there. So they should take care of that temperature level before it reaches to consumer.

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- **Canned Foods:**
  - ✓ The shelf-life of canned foods results from the destruction of microorganisms capable of growth within the container during normal handling and storage.
  - ✓ Follow the GMP regulations for low-acid foods. pH 4.4 a<sub>w</sub> 0.85
  - ✓ Reduce the spore level in the food. As a general rule, food with a high spore level requires more retort time and/or temperature in the same or similar operations. Assuming the same retort time and/or temperature, the incidence of spoilage will be higher in the canned food with a high initial spore level when all other factors are the same
  - ✓ Follow good sanitation and good container handling techniques during the container cooling and post-cooling period. It is also important to cool heat processed containers quickly to about 100°F (38° C) since thermophilic outgrowth may occur with low spore numbers if containers are stacked or cased while hot.
  - ✓ Maintain good seams on cans and tight lids on glass containers by regular control and testing.

Canned foods, the shelf life of canned foods results from the destruction of microorganisms capable of growth within the container during normal handling and storage. They follow GMP regulation for low-acid foods that is I already told, low acid food the condition is pH 4 point 6

above and water activity is point 85, most of this condition is most favourable for Clostridium botulinum. So they have to follow GMP regulations and they reduce the spore level in the food, as a general rule, food with a high spore level requires more retort time and temperature in the same similar operations.

We have already told, if we maintain the say, for example, there two cases, one with high spore and another with low spore, low spore require low time, low temperature compared to higher one. Assuming, same the incidence of spoilage will be higher for canned food with a high initial spore level. The next one is the follow good sanitation and good container handling techniques so everything should follow GMP regulations and cooling as well as post cooling period.

The important thing is, even though we employ pasteurization or sterilization at a high temperature immediately the product should be cooled to normal room temperature because it reduces the possibility of growth of the thermophiles organisms because the thermophiles survived higher temperature. It should be cool to normal room temperature to avoid the growth of thermophiles. And also maintain good seams on cans because the air tight container should maintain the perfect sealing to control the growth of the microorganisms, okay.

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So I have showed one particular table which is taken from the prevention of microbial and parasitic hazard associated with processed food, a guide for the food processor and pH value as well as the water activity value were taken from some of the references and the rest are the books and references which are used in the particular lecture. So that is all for today, so tomorrow we will see about next lecture. Thank you.