1. FOOD SAFETY

1.1 QUALITY CONTROL:

a) MICROBIOLOGICAL ANALYSIS OF FOOD:

Daily food samples shall be collected at random, at different stages (viz. raw, semi-processed, and processed) for microbiological analysis. This test results shall be communicated to the Executive Chef and the General Manager on daily basis. If any food sample is found to be of poor microbiological quality, sectional head and concerned staff shall be briefed about the remedial measures to be taken. Weekly all the test reports shall be once again reviewed and corrective action report highlighting remedial measures to improve microbiological standards shall be given to the Executive Chef for implementation.

Microbiological quality of perishable and non-perishable stock items shall be checked upon receiving and further at regular intervals, test results shall be communicated to the Purchase Manager / Store-in-Charge.

b) ANALYSIS OF WATER AND ICE CUBES:

The water sanitation is well controlled by agencies concerned with the public health but several water borne outbreaks have been reported due to water contamination by sewage, irrigation water. Microorganisms including pathogens do not multiply but may sustain life for a while. Hence the portability of water shall be checked regularly. The chlorine level in the water shall be maintained between 0.2 to 0.5 ppm. Hardness of water supplied to the dishwashing machine shall be regularly monitored (shall be maintained below 7ppm).

Incase of discrepancies regarding microbiological quality, chlorine content or hardness, Maintenance Manager / Chief Engineer shall be informed to take up corrective measures.

c) **MONITORING PRODUCTION HYGIENE AND PERSONAL HYGIENE BY SWAB TEST**:

Efficiency of manual and mechanical cleaning / washing / disinfection process shall be evaluated regularly by swab testing using RODAC plate or any other suitable method. Results shall be communicated to concerned departments. Concerned departments shall be briefed whenever corrective action is required.

Hand hygiene shall be monitored regularly by swab testing using RODAC plates. Defaulters shall be warned verbally on 1st instance. On 2nd instance warning letter shall be issued and on 3rd instance strict disciplinary action shall be taken against her / him.

1.2 Lab procedures:

a) MONITORING QUALITY OF CLEANING AGENTS:

Different types of detergents, disinfectant and other cleaning agents shall be checked for specification compliance, upon receipt. Test results shall be communicated to the Purchase Departments.

b) TEMPERATURE CONTROL:

Cold storages and deep freezer temperature shall be monitored every four hours. Discrepancies regarding temperature shall be immediately communicated to Maintenance department.

Food temperature at different stages of production shall be closely monitored in order to prevent bacterial multiplication in foods. Temperatures shall be recorded on daily basis.

Food poisoning is illness caused by eating noxious food. Food poisoning is broadly categorized into Biological and Chemical food poisoning.

c) CHEMICAL FOOD POISONING:

Chemical food poisoning can be caused by contamination of food by pesticides, detergents, disinfectants, excess quantities of additives or preservatives and heavy metals. Chipped enamel vessel can cause antimony poisoning. Use of galvanized equipment with acid food may result in zinc poisoning.

d) BIOLOGICAL FOOD POISONING:

Biological food poisoning can be caused by:

13 Parasites
14 Protozoa
15 Poisonous plants
16 Poisonous animals
17 Viruses
18 Yeasts
19 Molds
20 Bacteria

e) PARASITES:

Parasites such as round worm, tape worms can cause disease in human beings after eating contaminated food.

A disease called Trichinosis is caused by consuming pork infested with a round worm called Trichinella spiralis. The onset of symptoms is usually about two days offer consuming the pork contaminated with the larvae. Symptoms experienced are abdominal pain, nausea, vomiting, and diarrhea. Heating pork to 66°C kills the larvae.

The tape worm may cause disease in human beings when larvae infested meat is ingested. Though infection of human being with the tape worm is rare, it can be a serious affliction because Taenia solium may develop in the human being. Heating meat to 66°C renders the meat safe.

f) PROTOZOA:

Entamoeba histolytica causes disease in human beings called amoebas or amoebic dysentery. Food becomes contaminated from soil, rats, flies or unclean hands of infested persons. The symptoms vary from mild to violent diarrhea, alternating with constipation and abdominal pain.

g) POISONOUS PLANTS:

Certain types of mushrooms are extremely poisonous.

h) POISONOUS ANIMALS

Paralytic shell fish poisoning may result from eating mussels and clams which have become poisonous by feeding poisonous plankton. Also numbers of tropical fish are known to be poisonous to man e.g. parrot fish, porcupine fish, and goat fish.

i) VIRUSES:

Viruses are the smallest and perhaps simplest form of life known. Viruses may range from 1/100 to 1/3 the size of bacterium. Because of their small size they cannot see by standard optical microscope. Viruses can not multiply in food. The food is vehicle to transport the virus which may lodge in human host and reproduce abundantly causing disease. Viral hepatitis is most commonly known viral disease. Virus's diseases are often found in water contaminated with sewage or chemically untreated water. Outbreaks of viral disease are often attributed to poor personal hygiene and contaminated water supply.

j) YEASTS:

Yeast is slightly larger than the average bacterium. Yeasts are very useful group of organisms and very rarely involved in food poisoning. But presence of large number of yeasts is food leads to food spoilage. Usually food with high sugar and acids content are more prone to spoilage due to yeast growth.

k) MOLDS:

Molds size ranges from microscopic to macroscopic. Some species of molds are useful which are used for manufacturing antibiotics, ripening of cheese. But molds are responsible for food spoilage to great extent. Also, some types of molds can produce very potent chemical which has toxic and carcinogenic effect. Toxins produced by molds, penetrates the foods relatively quickly hence removal of visible mold deposits does not render the food safe. Some types of molds impart musty odor and flavor to the food or change their appearance. Such foods are not always detrimental to health but are unfit for human consumption.

l) BACTERIA:

Bacteria are of greatest concern to food service, since it is more commonly involved in cases of food borne illnesses than other biological forms mentioned above. Bacteria are extremely small living organisms which cannot be seen by naked eye (about one thousandth of a mm). Approximately one million bacteria lumped together would cover a pinhead. Bacteria vary in shapes and sizes. They may be spherical, rod shaped or comma shaped.

Bacteria multiply by simple division into two, under suitable conditions of environment i.e. moisture, food, and temperature of 37°C. This occurs every 20 or 30 minutes. Thus one bacterium under optimum condition multiplies into two in 20 minutes and more than a billion in 10 hours. Most bacteria reproduce best at temperature between 5°C and 63°C (danger zone). Higher (above 60°C) or lower (below 0°C) temperatures retards growth. Above 63°C and below 5°C growth is almost standstill. Most of the food poisoning bacteria are killed at 74°C.Bacteria are present almost everywhere, in the environment, in air, water, soil, including ourselves. There are thousands of different types of bacteria. Many of them beneficial e.g. production of curds, cheese whereas few types are found to be harmful. Some bacteria spoil the food bye their growth, some produce acids making the food sour, other breakdown proteins, causing putrid odor, some may contribute to sliminess, discoloration. These undesirable changes indicate the presence of high number of bacteria. Some bacteria do not contribute any changes but still render the food unsafe for human consumption, they either grow in food or produce toxins or they enter the human body through food and multiply in the body, discharging toxic wastes.

m) COMMON FOOD POISONING BACTERIA:

Of the thousand types of bacteria, which exist following are the types which are well recognized as casual agents of food poisoning and food borne infection.

n) STAPHYLOCOCCI:

Staphylococcal food intoxication is one of the most common types of food borne illness. Staphylococcal bacteria are commonly found in human nasal passages, throat, on the hands, and skin and especially in infected cuts and wounds, burns, boils, pimples. Hence, it can be easily transferred into foods while processing and preparation if proper hand hygiene practices are not followed. These bacteria multiply in food and produce heat resistant toxin. Within 1 to 7 hours after consumption of food with toxin, usually symptoms of vomiting and diarrhea are experienced by a person. Contamination can be prevented if basic principles of hygiene are observed by food handler, further chilling the food products below 10°C after preparation to prevent undesirable growth is necessary.

o) SALMONELLA:

After Staphylococci, Salmonella are the most frequent cause of disease from contaminated food. Salmonella is commonly found in animal fodder, birds, and firm animals particularly poultry. Also human being infected by Salmonella can become a source of contamination, if hands are not washed after visit to the w.c. Improper thawing, inadequate cooking, improper handling of suspect foods like meat, egg, poultry leads to contamination of food by salmonella. After consuming food contaminated with salmonella. Symptoms are experienced usually after 12 – 36 hrs. Illness is marked by diarrhea, abdominal pain, vomiting and fever.

p) CLOSTRIDIUM PERFRINGES:

Clostridium per fringes is a common organism frequently found in soil, human and animal excreta. Careless handling of raw poultry, meat soiled vegetables may contaminate the food. It can survive heat by forming dormant forms called spores. Unlike staphylococci, Clostridium per fringes produces toxins only when it is inside the body. Onset of symptoms like nausea, diarrhea, abdominal pain is experienced after 8 - 22 hrs.

q) CLOSTRIDIUM BOTULINUM:

Food poisoning by Clostridium botulinum is very rare. Clostridium botulinum is found in soil, water, human and animal intestine. Food implicated in botulinum outbreaks are improperly processed usually home canned, low acid goods e.g. mushrooms, corns, beef, tuna and smoked vacuum packed fish. The toxin is produced by the bacteria when they are growing in food under strictly anaerobic conditions. Toxin is highly poisonous. (People have died after eating only a mouthful of infected food). Symptoms usually appear after 12 – 36 hrs, after ingestion of contaminated food. Symptoms expressed are giddiness, double vision, headache, nausea, vomiting. Botulism attacks central nervous system, resulting in paralytic attack. If anti-toxin is not administered, botulism may prove fatal. To prevent botulism reject any swollen / blown cans, never use home canned foods for commercial food service establishment.

r) BACILLUS CEREUS:

Bacillus cereus is found in soil and dust. It can form spores when conditions are unfavorable. Spores are often found in cereals, dry mix powder. Some spores survive cooking and germinate into bacilli which under warm storage conditions in cooked food grow and produce toxin. Onset if symptoms may be sudden with acute vomiting and diarrhea, usually after 1 – 16 hrs. of ingestion of infected food. Cooked food should be cooled and refrigerated rapidly and reheated to prevent the bacillus cereus poisoning.

s) VIBRO PARAHAEMOLYTICUS:

Vibro parahaemolyticus is naturally occurring organism in marine environment. Hence, it can be insolated from seafood, gross mishandling practices, such as improper refrigeration, insufficient cooking, cross contamination and recontamination. The average incubation period is about 15 hrs and there is rapid onset of illness with profuse diarrhea often leading to dehydration, vomiting and fever.

t) ESCHERICHIA COLI:

E.coli is normal inhabitant of the intestinal tract of man and animal. E.coli is transferred to the food either through the raw products or while processing the food by means of hands which are not washed properly after visiting the toilet. Many E.coli strains are enteropathogenic and give rise to acute diarrhoea in infants. Some serotype causes diarrhoea in adults. Incubation period is 12 hours to 3 days. Symptoms may be diarrhoea or dysentery.

u) LISTERIA MONOCYTOGENES:

Listeriosis **is a serious infection** caused by eating food contaminated with the bacterium called *Listeria monocytogenes*. *Listeria* is found in soil and water. Vegetables can become contaminated from the soil or from manure used as fertilizer. Animals can carry the bacterium without appearing ill, contaminating their food products such a meat and dairy.

2. Microbial inspection procedure

inspection procedure	Responsibility	
	Performed by	Monitored by
a) The scope of this SOP defines the microbial testing for finished products, in-process, raw materials, etc. 1		
b) Food samples are taken at random from different areas in the kitchen & checked for micro-organisms. (ref sampling plans).	Microbiologist	Mgr QC
c) A daily report of the food samples tested is given to the executive chef for his reference and a copy is retained in the lab.	Microbiologist	Mgr QC
d) Corrective actions for the unacceptable samples are carried out by the chef & the samples are re-tested for compliance. Results are recorded in the microbiological report.	Microbiologist	Mgr QC
e) Equipment swabs & hand swabs are also taken at random (ref sampling plans). In case of hand swab unacceptable samples are shown to the concerned staff and then re-tested. Retest is done for failed equipment swabs.	Microbiologist	Mgr QC
f) Portability of water and ice cubes are analyzed from different areas at random. (ref sampling plans). Reports of the unacceptable samples of water and ice are given to the engineering dept for corrective action to be taken.	Microbiologist	Mgr QC
g) All samples are tested in accordance with the approved test methods. (See lab manual).	Microbiologist	Mgr QC
h) Results are checked against the standards, if results are not within limits correction and/or corrective actions are taken.	Microbiologist	Mgr QC
i) All reports are noted in a rough register and then written down in detail in respective formats. Each sample is identified by sample number corresponding to the name mentioned in the register.	Microbiologist	Mgr QC
j) The following computer generated (or manual) records are kept in the laboratory a) Microbiological food reports b) Equipment swab reports c) Hand swab reports d) Water / ice reports e) Chlorine concentration	Microbiologist	Mgr QC

k) All the above records are approved by a signature	Mgr QC	G. M.
k) A monthly report is drafted taking into account the number of samples tested, no of samples acceptable or unacceptable and copies are given to the Corporate chef.	Mgr QC	Mgr QC
l) Temperatures are monitored on a daily basis. Temperatures are recorded on temperature chart. Specified temperatures are displayed and defined in the HACCP plan. Fluctuations outside target actual temperatures are communicated verbally to the engineering department.	Engineering	Mgr QC

2.1 Sample Description	Frequency	Type of Tests
a) KITCHEN	05 samples per kitchen per month	TPC, Coliforms,E.coli,S.aureus
	(Semi processed)	Salmonella,
Buffet (Hot)	2 samples per day	TPC, Coliforms,E.coli,S.aureus
		Salmonella,
Buffet (Cold)	3 samples per day	TPC, Coliforms,E.coli,S.aureus
		Salmonella,
Banquets	10 samples per week	TPC, Coliforms,E.coli,S.aureus
		Salmonella
b) BUTCHERY		
1. Raw meat and poultry	once in a week	TPC, Coliforms,E.coli,S.aureus
		Salmonella
2. Sea food	once in a week	TPC, Coliforms,E.coli,S.aureus
		Salmonella,
c) PANTRY		
1. Fresh Juices	5 samples per month	TPC, Coliforms,E.coli,S.aureus
		Salmonella,
2. Ice Creams	2 samples per month	TPC, Coliforms,E.coli,S.aureus
		Salmonella, Yeast & Molds
Sample Description	Frequency	Type of Tests
d) STORES		
1. Spices & pickles	1 samples per month	TPC, Coliforms,E.coli,S.aureus
		Salmonella,, Yeast & Molds
2. Canned & bootled items	1 samples per month	TPC, Coliforms,E.coli,S.aureus
		Salmonella, Yeast & Molds
3. Cheese	2 samples per month	TPC, Coliforms,E.coli,S.aureus
		Salmonella, Yeast & Molds

e)RECEIVING		
1. Dairy products	once in a week	TPC, Coliforms,E.coli,S.aureus
		Salmonella, Yeast & Molds
2. Raw Vegetables & fruits	once in a week	TPC, Coliforms,E.coli,S.aureus
		Salmonella,
3. Wheat flour & maida	once in 6 months	Gluten content, Moisture (external Laboratory
Toxin Analysis	once in 6 months	Out side Laboratory
Heavy metal test in seafood	once in 6 months	Out side Laboratory
f) Swimming Pool Water	Monthly Twice	TPC, Chemical Analysis & Coliforms, Ecoli

2.2 SWAB TESTS

Sample Description	Frequency	Type of Tests
Chopping boards	Randomly	TPC
Table top & work surfaces	Randomly	TPC
Kitchen Equipments	Randomly	TPC
Knives	Randomly	TPC
Hand Swabs	Randomly	TPC/Ecoli
Linen Swabs	Randomly	TPC
AERIAL COUNTS	Each kitchen twice a month	TPC, Molds
WATER (Microbiological Tests)	Twice a month	TPC, Coliforms, E.coli
ICE CUBES	Twice a month	TPC, Coliforms, E.coli
Food Safety Training Classes	As per schedule given by training dept	

GRADE	REMARK
A	Excellent
B	Good
C	Satisfactory
D	Not acceptable
<	Less than
□	Less than or equal to
>	Greater than
*	Test not considered as routine
All bacterial counts are expressed as no of Bacteria per gram	

3. Microbiological Interpretation Guide lines

TYPE I

a) Heat Treated Food Not Manipulated Other than Portioning

CLASSIFICATION	A	B	C	D
Total viable count	$\leq 10^3$	$\leq 10^5$	$\leq 10^6$	$>10^6$
Coliform count	$\leq 10^2$	$\leq 10^3$	$\leq 3x10^3$	$> 3x10^3$
1. coli	ABSENT	ABSENT	ABSENT	PRESENT
S.aureus	$\leq 10^2$	$\leq 10^2$	$\leq 10^3$	$> 10^3$
Salmonella	ABSENT	ABSENT	ABSENT	ABSENT
Yeast*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$> 10^3$
Molds*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$> 10^3$
• cereus*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$> 10^3$
C. perfringes*	$\leq 10^2$	$\leq 10^2$	$\leq 10^3$	$> 10^3$
Campylobacter*	ABSENT	ABSENT	ABSENT	PRESENT

TYPE II

b) Heat Treated Food Manipulated After Heat (Cream, Desserts or Deboned Poultry)

CLASSIFICATION	A	B	C	D
Total viable count	$\leq 10^5$	$\leq 10^6$	$\leq 10^7$	$>10^7$
Coli form count	$\leq 10^2$	$\leq 10^3$	$\leq 10^4$	$> 10^4$
14 Coli	ABSENT	ABSENT	ABSENT	PRESENT
27 aureus	$\leq 10^2$	$\leq 10^2$	$\leq 10^3$	$> 10^3$
Salmonella	ABSENT	ABSENT	ABSENT	PRESENT
Yeast*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$> 10^3$
Molds*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$> 10^3$
17 cereus*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$> 10^3$
1. per fringes*	$\leq 10^2$	$\leq 10^2$	$\leq 10^3$	$> 10^3$
Campylobacter*	ABSENT	ABSENT	ABSENT	PRESENT

TYPE III

c) Raw fruits, frozen raw vegetables, or items containing such

CLASSIFICATION	A	B	C	D
Total viable count	$\leq 5x10^5$	$\leq 5x\ 10^6$	$\leq 10^7$	$>10^7$
Coliform count	$\leq 2.5x10^3$	$\leq 5x10^3$	$\leq 10^5$	$> 10^5$
25 Coli	ABSENT	ABSENT	ABSENT	PRESENT
8 Aureus	$\leq 10^2$	$\leq 10^2$	$\leq 10^3$	$> 10^3$
Salmonella	ABSENT	ABSENT	ABSENT	PRESENT
Yeast*	$\leq 5x10^3$	$\leq 5x10^4$	$\leq 10^5$	$> 10^5$
Molds*	$\leq 5x10^3$	$\leq 10^4$	$\leq 10^5$	$> 10^5$
12 cereus*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$>10^3$
11 per fringes*	_____	_____	_____	
Campylobacter*	ABSENT	ABSENT	ABSENT	PRESENT

TYPE IV

m) Vacuum packed items: Smoked meat& fish salami, smoked salmon etc.

CLASSIFICATION	A	B	C	D
Total viable count	$\leq 10^6$	$\leq 5x\ 10^6$	$\leq 10^7$	$>10^7$
Coliform count	$\leq 10^2$	$\leq 10^3$	$\leq 10^4$	$>10^4$
25 Coli	ABSENT	ABSENT	ABSENT	PRESENT
25 Aureus	$\leq 10^2$	$\leq 10^2$	$\leq 10^3$	$> 10^3$
Salmonella	ABSENT	ABSENT	ABSENT	PRESENT
Yeast*	$\leq 10^2$	$\leq 5x\ 10^2$	$\leq 10^3$	$>10^3$
Molds*	$\leq 10^2$	$\leq 5x\ 10^2$	$\leq 10^3$	$>10^3$
10.0 cereus*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$>10^3$
20 perfringes*	_____	_____	_____	_____
Campylobacter*	ABSENT	ABSENT	ABSENT	PRESENT

TYPE V

n) Fermented items like cheese, yoghurt, sour cream products

CLASSIFICATION	A	B	C	D
Total viable count	_____	_____	_____	_____
Coliform count	$\leq 10^2$	$\leq 10^3$	$\leq 5x10^3$	$>5x10^3$
25 Coli	ABSENT	ABSENT	ABSENT	PRESENT
22 Aureus	$\leq 10^2$	$\leq 10^2$	$\leq 10^3$	$> 10^3$
Salmonella	ABSENT	ABSENT	ABSENT	PRESENT
Yeast*	$\leq 10^2$	$\leq 5x\ 10^2$	$\leq 10^3$	$>10^3$
Molds*	$\leq 10^2$	$\leq 5x\ 10^2$	$\leq 10^3$	$>10^3$
24 cereus*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$>10^3$
10 perfringes*	_____	_____	_____	_____
Campylobacter*	ABSENT	ABSENT	ABSENT	PRESENT

TYPE VI

o) Raw meat / Raw fish

CLASSIFICATION	A	B	C	D
Total viable count	$\leq 10^5$	$\leq 10^6$	$\leq 10^7$	$>10^7$
Coliform count	$\leq 10^3$	$\leq 5x10^3$	$\leq 10^4$	$>10^4$
16 Coli	≤ 10	$\leq 10^2$	$\leq 10^3$	$> 10^3$
24 aureus	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$> 10^3$
Salmonella	ABSENT	ABSENT	ABSENT	PRESENT
Yeast*	$\leq 10^2$	$\leq 5x\ 10^2$	$\leq 10^3$	$>10^3$
Molds*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$>10^3$
7 cereus*	$\leq 10^2$	$\leq 5x10^2$	$\leq 10^3$	$>10^3$
15 perfringes*	_____	_____	_____	_____
Campylobacter*	ABSENT	ABSENT	ABSENT	PRESENT

4. MICROBIOLOGICAL WATER & ICE CUBES STANDARDS

a)Drinking Water		
Sr no.	**Microbiological Criteria**	**Desirable limits**
1	Total count/ ml at 37°C	<100 cfu
2	Total count/ ml at Room Temperature.	<100 cfu
3	Total no. of Coliforms/ 100 ml	absent
4	Total no. of E.coli/ 100 ml	absent
5	Total no. of Salmonella/ 100ml	absent
Sr no.	**Others**	**Desirable limits**
1	Appearance	Clear
2	Taste	Agreeable
3	Odour	Agreeable
4	Available chlorine	0.2 - 0.5 ppm
5	Total hardness	< 50 ppm
6	TDS	< 100 ppm
7	pH	6.5 - 8.5

b) Packaged Drinking Water		
Sr no.	**Microbiological Criteria**	**Desirable limits**
1	Total count/ ml at 37°C	< 20 cfu
2	Total count/ ml at Room Temperature.	<100 cfu
3	Total no. of Coliforms/ 250 ml	absent
4	Total no. of E.coli/ 250 ml	absent
5	Total no. of Salmonella/ 250ml	absent
Sr no.	**Others**	**Desirable limits**
1	Appearance	Clear
2	Taste	Agreeable
3	Odour	Agreeable
4	Available chlorine	Max 0.2 ppm
5	Total hardness	< 50 ppm
6	TDS	< 100 ppm
7	pH	6.5 - 8.5

c)Ice cubes		
Sr no.	**Microbiological Criteria**	**Desirable limits**
1	Total count/ ml at 37°C	<100 cfu
2	Total count/ ml at Room Temperature.	<100 cfu
3	Total no. of Coliforms/ 100 ml	Absent
4	Total no. of E.coli/ 100 ml	Absent
5	Total no. of Salmonella/ 100ml	Absent
Sr no.	**Others**	**Desirable limits**
1	Appearance	Clear
2	Suspended matter	Absent

5. MICROBIOLOGICAL STANDARDS FOR SWAB TEST		
a) Hand Swabs		
Criteria	Satisfactory	Unsatisfactory
Coliform Count	< 5cfu/ plate*	> 5cfu/ plate*
E. Coli	absent	Present
b)Service Equipment Swabs		
Criteria	Satisfactory	Unsatisfactory
Total Count	< 50cfu/ plate*	> 50cfu/ plate*
c) Kitchen Equipment Swabs		
Criteria	Satisfactory	Unsatisfactory
Total Count	< 100cfu/ plate*	> 100cfu/ plate*

5.1 MICROBIOLOGICAL STANDARDS FOR AERIAL COUNT

Criteria	After 30mins exposure
Total Count	< 50 cfu / plate *
Yeast Mould	< 50 cfu / plate *
* Inner diameter of Plate: 9.0 cm	

6. MICROBIOLOGICAL GUIDELINES

6.1 NON-MANIPULATED ITEMS

This category refers to items that are sampled directly from boilers or ovens in the Hot Kitchen area before any slicing or handling has taken place.

a) ESSENTIAL MICROBIOLOGICAL CRITERIA

- Escherichia Coli	not detected in 1 gram
- Salmonella species	not detected in 25 gram
- Staphylococcus aureus	less than 100/g
- Clostridium perfringens	less than 100/g
- Bacillus species (as applicable)	less than 1000/g
- Listeria monocytogenes	not detected in 25 gram
- Campylobacter (as applicable)	not detected in 25 gram

b) NON-ESSENTIAL MICROBIOLOGICAL CRITERIA

Colony Plate Count less than 10 000/g

6.2 MANIPULATED ITEMS AFTER COOLING

This applies to food items such as cooked and stripped chicken, fish, plain rice and pasta, meats for hot meals and hot desserts. It includes the portioning of hot meals.

a) ESSENTIAL MICROBIOLOGICAL CRITERIA

-	Escherichia Coli	not detected in 1 gram
-	Salmonella species	not detected per 25 gram
-	Staphylococcus aureus	less than 100/g
-	Clostridium perfringens	less than 100/g
-	Bacillus species (as applicable)	less than 1000/g
-	Bacillus species (as applicable)	not detected in 25 gram

b) NON-ESSENTIAL MICROBIOLOGICAL CRITERIA

-	Colony Plate Count	less than 100,000/g
-	Total Coli form Count	less than 1,000/g
-	*Campylobacter (as applicable)	not detected in 25 g

* This test should be carried out on poultry and poultry products.

6.3 MANIPULATED COLD MEAL ITEMS

This indicates food items such as prawns, cold sliced meats (cooked 'in house' products and 'bought-in' products), pate and high risk items that will receive no further heat treatment. It DOES NOT include smoked or fermented products or raw fruit and vegetables.

a) ESSENTIAL MICROBIOLOGICAL CRITERIA

-	Escherichia coli	not detected in 1 gram
-	Salmonella species	not detected per 25 gram
-	Staphylococcus aureus	less than 100/g
-	Clostridium perfringens	less than 100/g
-	Bacillus species	less than 1000/g
-	Campylobacter (as applicable)	not detected in 25 gram
	*Listeria monocytogenes	less than 100/g
-	Colony Plate Count	less than 1000 000/g
-	Total Coliform Count	less than 10 000/g

b) NON-ESSENTIAL MICROBIOLOGICAL CRITERIA

-	Yeast and mould Counts	less than 10 000/g
-	+ Vibrio species (as applicable)	not detected in 25 g

*ideally this should be detected in 25 grams but experience dictates that low levels are sometimes detected.

+this includes Vibro parahaemolyticus and Vibro cholera 01 and Non 01. It is recommended that all seafood and fish are tested for the presence of Vibrio species.

6.4 COLD SMOKED' OR FERMENTED MEAL ITEMS

This applies to cold items such as smoked salmon, smoked mackerel, salami, cheese, tofu and yoghurts, etc

a).ESSENTIAL MICROBIOLOGICAL CRITERIA

-	Escherichia Coli	not detected in 1 gram
-	Salmonella species	not detected in 25 gram
-	Staphylococcus aurous	less than 100/g
-	Clostridium perfrigens	less than 100/g
-	Bacillus species (as applicable)	less than 100/g
-	Campylobacter (as applicable)	not detected in 25 gram

b)NON-ESSENTIAL MICROBIOLOGICAL CRITERIA

-	Yeast and Mould Counts	less than 10 00/g

7. ITEMS REQUIRING FURTHER COOKING

This category refers to blanched vegetables, sealed meats for example steaks, rare beef and certain hot breakfast items.

a) ESSENTIAL MICROBIOLOGICAL CRITERIA

-	Escherichia Coli	less than 10/g
-	Salmonella species	not detected in 25 gram
-	Staphylococcus aurous	less than 100/g
-	Clostridium perfrigens	less than 1000/g
-	Bacillus species (as applicable)	less than 1000/g
-	Campylobacter (as applicable)	not detected in 25 gram

b) NON-ESSENTIAL MICROBIOLOGICAL CRITERIA

-	*Listeria monocytogenes	less than 100/g

*although this should not be detected in 25g, Listeria monocytogenes is ubiquitous and these food items would not have received sufficient heat treatment to have killed all organisms present. The further cooking on the aircraft should kill the remaining organisms.

7.1 RAW VEGETABLES AND RAW FRUITS

This applies to salad items and some pre-prepared dressed salads, also uncooked fruit such as strawberries, mangoes etc.

a) ESSENTIAL MICROBIOLOGICAL CRITERIA

- Escherichia coli — less than 10/g
- Salmonella species — not detected in 25 gram

b) NON-ESSENTIAL MICROBIOLIGICAL CRITERIA

- Total Coli form Count — less than 10/g
- Yeast and Mould Count — less than 100 000/g

7.2 PRE-PREPARED SALAD ITEMS

Prepared salads with a mayonnaise, vinaigrette or similar dressing include a range of different products. At one extreme are 'higher risk' products such as those containing pasta, prawns, or other proteins foods? At the other, are vegetables mixes with only a light dressing? It is impracticable to categorize prepared salads into one group on the basis of their typical microbiology.

It is suggested that the criteria in other sections will apply as appropriate to the particular salad. For example, for the pasta or meat containing products, section 1.3 is the most appropriate. For vegetables in a minimum of dressing, then the raw vegetables criteria (1.6) are more applicable.

7.3 WATER AND WET ICE

Water samples should be taken as outlined in 'The Bacteriological Examination of Drinking Water Supplies 1982 (reports on Public Health and Medical Subjects No. 71).

Samples of wet ice should be taken aseptically and allowed to thaw gently.

a) ESSENTIAL MICROBIOLOGICAL CRITERIA

- Colony Plate Count — less than 100/ml
- Total Coli form Count — not detected per 100 ml
- Escherichia Coli — not detected per 100 ml

7.4 MICROBIOLOGICAL ANALYSIS OF FOOD - GUIDELINES

All the food samples which are collect at random shall be checked for total viable count, total coli form count, total S.aureus, presence/absence of E.coli and Salmonella. Tests for other food poisoning organisms viz Clostridium, Vibrio, Campylobacter, Listeria, Bacillus cereus, shall be carried out for specific foods and where ever necessary. Test for yeast and molds shall be performed for products like yoghurt, cheese, juices etc.

a) SAMPLE COLLECTION:

Collect food sample, aseptically in a sterile plastic bag or Aluminum foil pan. Keep the sample refrigerated until processed.

b) PREPARATION OF DILUENT:

Dissolve 1g peptone in 1000 ml distilled water. Fill 225 ml of peptone water in each 250 ml Erlenmeyer flask, plug it with nonabsorbent cotton and sterilize by autoclaving at 15 lbs /sq.inch (121xC) for 20 minutes.

c) MEDIA REQUIREMENT:

- Soya bean casein digest agar
- MacConkey's agar
- Baird Parker agar
- Phenol red lactose broth
- Brilliant green lactose bile broth (B.G.L.B)
- Phenol red lactose broth (P.R.L.B.)
- Selenite broth
- Bismuth sulphite agar
- Eosin methylene blue agar
- Phenol red egg yolk agar
- Rose Bengal agar
- Brain heart infusion broth
- Robertson's cooked meat medium
- Lactose broth
- Thiosulphate citrate bile salt sucrose agar
- Blasers agar (HI-FD006)
- Butzlers agar (HI-FD007)
- Brucella FBP BROTH
- Tryptose sulphite cycloserine agar
- XLD agar

d) SAMPLE PREPARATION

- Weigh 25 g of food sample, add 225 ml of diluents.

- Homogenize the food sample using sterile blender or stomacher (10^{-1} dilution).

- Prepare serial dilutions by transferring -

  10 ml of 10^{-1} dilution to 90ml of sterile diluents (10^{-2}).

  10 ml of 10^{-2} dilution to 90 ml of sterile diluents (10^{-3})

  10 ml of 10^{-3} dilution to 90 ml of sterile diluents (10^{-4})

e) DETECTION OF S.AUREUS :

- Inoculate 0.1 ml of 10^{-1} dilution in Baird Parker agar plate by spread plate technique.
- Incubate the plates at 37 °C for 48 hours and record the results.
- Typical colonies are grey to jet black convex (1 to 1.5 mm dial) surrounded by opaque zone and clear halo beyond opaque zone.

Confirm with biochemical tests.

f) DETECTION OF E.COLI:

- Inoculate 1 ml of 10^{-1} dilution to BGLB broth with inverted Durham's tube.
- Incubate at 44.5°C for 24 hours.
- Isolate loopful from the tubes showing gas production on EMB agar.
- Incubate the plates at 37°C for 24 hours and record the results.
- Typical colonies are dark centered with greenish metallic sheen.

g) Detection of Salmonella:

- Inoculate 5 ml of 10^{-1} dilution in 10 ml of double strength lactose broth.
- Incubate the tubes at 37°C for 24 hours.
- Transfer 1 ml of prenticed culture to 9 ml of tetrathionate / Selenite cysteine broth.
- Incubate at 37° for 24 hrs and isolate loopful on XLD agar/Bismuth sulphite agar
- Incubate the plates at 37°C for 24 hours and record the results.
- Typical colonies on XLD agar are red with black center.

21 Typical colonies on Bismuth sulphite agar are black with metallic sheen.

h) Total coli form count:

- Inoculate 1 ml of 10^{-2} (or appro.) dilution into MacConkey's agar by pour plate technique.
- Incubate at 37°C for 48 hours.
- Count typical pink / red colonies and record the results.

i) Total viable count:

- Inoculate 1 ml sample from of 10^{-3} (or appro) dilution in soybean casein digest agar by pour plate technique.
- Incubate the plates at 37°C for 48 hours.
- Count total number of colonies and record the results.

j) Detection of Yeast & Mold:

- Inoculate 0.1 ml of 10^{-1} (or appro) dilution in Rose Bengal agar by spread plate technique.

Incubate at room temperature for 48 to 72 hours and record the results.

k) Detection of Vibrio

- Weigh 25 g of sample; add 225 ml of alkaline peptone water. Homogenize using sterile blender or stomacher.
- Incubate the homogenized sample at 37°C for 6 - 8 hours.
- Isolate loopful on TCBS agar at 37°C for 24 hours and record the results.
- Typical colonies are large (2-3mm), smooth, yellow (occasionally slow sucrose fermentors are green), slightly flattened with opaque centre and translucent peripheries.
- Confirm by biochemical tests.

l) Detection of Campylobacter

- Food samples shall be processed rapidly to ensure optimum isolation of Campylobacter jejune since organisms are sensitive to many environmental conditions. Micro aerobic conditions while sample enrichment and incubation shall be maintained.
- Weigh 10 g of the sample. Homogenize the sample with 90 ml of Brucella FBP broth.
- Incubate at 42°C for 16 - 18 hours.
- Isolate 1 - 2 loopfuls on Butzlers agar/Blaster's agar.
- Incubate at 42°C for 24 - 48 hours.
- Typical colonies are medium (1-2 mm), but can range from pinpoint to 4.5 mm in diameter, smooth, convex, glistening with distinct edge or flat translucent shining and spreading with irregular edge and non-hemolytic.
- Confirm with biochemical tests.

m) Detection of Bacillus cereus:

- Spread 0.1 ml of sample from 10^{-1} (or appro.) dilution on Manito yolk polymyxin agar (MYP).
- Incubate at 37°C for 24 hours and record the results.
- Typical colonies are pink to violet surrounded by large precipitation zone.
- Confirm by biochemical tests.

n) Detection of Clostridium botulinum:

- Inoculate 2 g of food sample in 15 ml of cooked meat medium.
- Incubate at 37°C for 7 days.
- Examine tubes for gas, digestion of meat particles and check odor.
- Prepare the smear and examine microscopically.
- If Clostridium botulinum cells are observed, confirm by toxin tests.

o) Detection of Clostridium perfringens:

- Inoculate 0.1 ml of 10^{-1} (or appro.) dilution on TSC agar.
- Overlay the plates when dry, with 10 ml of TSC agar without egg yolk.
- Incubate at 37°C ± 1 for 18 - 24 hours and record the results.
- Typical colonies are small black surrounded by an opaque white halo.

Confirm by biochemical tests.

8. MICROBIOLOGICAL ANALYSIS OF WATER / ICE CUBES

a) SAMPLE COLLECTION:

- Collect water / ice cube sample aseptically in sterile container.
- Keep the samples refrigerated until processed.
- Ice cubes shall be thawed gradually prior to processing.

b) EQUIPMENT:

- Membrane filter assembly
- Bacteriological membrane filters of pore size 0.45 micron.

c) MEDIA REQUIREMENT:

- Soya bean casein digest
- MacConkey's agar
- EMB agar
- Lactose broth
- BGLB broth
- Tetrathionate broth
- XLD agar

d) Enumeration of total viable count

By pour plate technique:

- Inoculate 1 ml of water sample in two soya bean casein digest agar plates by pour plate technique.
- Incubate one plate at room temperature and second plate at 37°C for 48 to 72 hours.
- Record the results by counting all colonies present.

e) By membrane filter technique

- Filter 100 ml of water sample through membrane filter aseptically.
- Place the filter aseptically on previously dried soya bean casein digest agar plate.
- Repeat the above step using 2nd membrane filter.
- Incubate one plate at room temperature and 2nd plate at 37°C for 48 hours.
- Count all the colonies and record the results.

f) Enumeration of total coli forms

- Filter 100 ml of water sample through membrane filter aseptically.
- Place the filter paper aseptically on previously dried MacConkey's agar plate.
- Incubate the plate at 37°C for 48 hours.
- Count all typical pink / red colonies and record the results.

g) Detection of Salmonella

- Filter 1-2 liters sample of water through membrane filter aseptically.
- Place the membrane filter in 50 ml of tetrathionate broth.
- Incubate the flask at 37°C for 18-24 hours.
- Isolate on brilliant green agar and record the results.

9. EQUIPMENT & HAND SWAB TECHNIQUES

a) RODAC plate method:

- Carefully press the agar surface on the surface being sampled.
- Incubate the plates at 37°C for 24 hours and record the result.

b) Cotton Swab Method:

- Rub the sterile cotton swab (non-absorbent) over 50 cm^2 of surface, turning the swab head slowly.
- Replace the swab in 10 ml of buffer rinse solution.
- Inoculate 1 ml of rinse solution to soya bean casein agar by pore plate technique.
- Incubate the plates at 37°C for 48 hours and record the results.

10. ESTIMATION OF TOTAL HARDNESS OF WATER

a) REAGENTS:

- Eriochrome Black T - indicator tablet
- Ammonia Buffer solution
- 0.02 N. Ethylene Diamine Tetra Acetic Acid Solution.

b) PROCEDURE:

- Take 50 ml water sample in 250 ml conical flask.
- Add 2 ml of Ammonia Buffer solution and crushed indicator tablet.
- Titrate against 0.02N E. D.T.A. solution until red colour disappears.
- End point will be blue, blue grey depending on the water.

c) CALCULATION:

$$\text{Total hardness} = \frac{1000 \times \text{ml of 0.02N EDTA consumed}}{\text{Volume of water sample}} = BR \times 20$$

11. ESTIMATION OF PERMANENT HARDNESS OF WATER

a) CHEMICAL & REAGENTS :

- Eriochrome Black T – indicator tablet
- Ammonia Buffer solution
- 0.02 N Ethylene Diamine Tetra Acetic Acid Solution.

b) PROCEDURE :

- Take 100 ml of water sample into 250ml conical flask.
- Heat to boiling point, and then cool to room temperature.
- Add 2 ml of ammonia buffer and crushed indicator tablet.
- Titrate against 0.02 N EDTA solutions until red colour disappears.
- End point will be blue or blue grey depending on water.

c) CALCULATIONS :

$$\text{Permanent hardness} = \frac{1000 \times \text{ml of 0.02N EDTA used}}{\text{Volume of water sample}}$$

12. ESTIMATION OF AVAILABLE CHLORINE-I

a) CHEMICALS AND REAGENTS:

- 0.1N Sodium thiosulphate
- Glacial acetic acid
- 10% Potassium iodide
- 0.5% Starch indicator

0.1N SODIUM THIOSULPHATE:

- Dissolve 25g of Sodium thiosulphate ($Na_2S_2O_3$) in 1000 ml of D/W
- Standardize against 0.1N Potassium dichromate.

10% POTASSIUM IODIDE:

- Dissolve 10g of Potassium iodide in 100 ml of D/W.Store in dark place.

0.5% STARCH INDICATOR:

- Dissolve 1.0g in 100 ml of D/W. Heat with continuous stirring till it becomes colorless.

b) PROCEDURE:

- Weigh 0.5ml of sample in Erlenmeyer flask.
- Add 100 ml of D/W.
- Add 20 ml of 10% Potassium iodide and 15 ml of glacial acetic acid.
- Titrate immediately against 0.1N Sodium thiosulphate till solution turns straw yellow colour.
- Add 1 ml Starch indicator.
- Titrate further till blue colour disappears for at least 60 seconds.
- Determine a blank titration reading using same volume of D/W as that of the sample.

Before calculating subtract the blank titration reading from sample titration reading (if blank requires more than 0.05 ml of 0.1N Sodium thiosulphate examine reagents and equipment for source of contamination.

c) CALCULATIONS:

Available chlorine (p.p.m.)

$$= \frac{\text{ml of 0.1N } Na_2S_2O_3 \text{ X N of } Na_2S_2O_3 \text{ X } 35450}{\text{ml of sample used}}$$

= (0.709 X ml of $Na_2S_2O_3$ consumed)

13. ESTIMATION OF AVAILABLE CHLORINE

(FOR 500 PPM - 2000 PPM)

a) CHEMICALS AND REAGENT:

- Glacial acetic acid
- Potassium iodide crystals (USP)
- 0.1N Sodium thiosulphate
- 1% Starch indicator

1.0% STARCH INDICATOR:

- Dissolve 1.0 gm in 100 ml D/W
- Heat with continuous stirring till it becomes colorless

0.1N SODIUM THIOSULPHATE:

- Dissolve 25 g of Sodium thiosulphate (Na2S2O3) in 1000 ml of D/W.
- Standardize against 0.1N Potassium dichromate.

b) PROCEDURE:

- Place few crystals of potassium iodide in a wide mouth Erlenmeyer flask.
- Add appox. 10 ml of distilled water.
- Add about 2g glacial acetic acid.
- Add 10 - 30 ml sample by means of volumetric pipette. The tip of pipette should almost touch surface of water in order to avoid surface loss.
- Titrate with standard Sodium thiosulphate using starch as an indicator.

c) CALCULATIONS:

Available chlorine (p.p.m.)

$$= \frac{\text{ml of 0.1N } Na_2S_2O_3 \text{ used X } 3.545}{\text{ml of sample}} \text{ X } 1000$$

14. DETERMINATION OF MOISTURE CONTENT OF FLOUR

a) Procedure:

Weigh accurately about 5 g of the flour in previously dried and weighed Petri dish. Place the Petri dish in an electric oven maintained at 105 ± 1°C five to six hours. Cool the Petri dish in a desiccators. Repeat the process of heating, cooling, and weighing at half an hour intervals until the loss in weight between two successive weightings is less than 1 mg. Record the weight.

b) Calculation:

$$\text{Moisture, percent by weight} = \frac{100\ (W_1 - W_2)}{W_1 - W}$$

WHERE:

W1 = Weight in g of the dish with the material before drying,

W2 = Weight in g of the dish with the material after drying, and

W = Weight in g of the empty dish.

15. DETERMINATION OF ACTIVE MATTER OF ANIONIC DETERGENTS

CHEMICALS AND REAGENTS:

- Methylene blue indicator solution.
- Chloroform - AR Grade
- 0.15 % Cetrimide solution.

METHYLENE BLUE: (stock solution)

Dissolve 1.0 g Methylene blue powder by pasting in Sulphuric acid (H_2SO_4) solution (0.5g H_2SO_4 + 3 ml Distilled water) Dilute the above prepared paste to 500 ml with distilled water.

METHYLENE BLUE: (Indicator solution)

Dissolve 50 g of Sodium sulphate (Na_2S0_4) (anhydrous) in sufficient distilled water. Add 6 ml of concentrated H_2SO_4 to 15 ml of Methylene blue stock solution, and dilute to 1000 ml with distilled water.

STANDARD 0.15% CETRIMIDE SOLUTION:

Dissolve 1.5g of A.R.Grade cetrimide (100% purity) in little water and dilute it to 1000 ml with distilled water.

a) PROCEDURE:

- Weigh accurately about 3 g of the sample in a 100 ml beaker dissolve in distilled water.
- Dilute it to 500 ml by transferring completely into a 500 ml volumetric flask using distilled water.
- Pipette out 10 ml of the above solution in 100 ml stoppered measuring cylinder.
- Add 20 ml of chloroform and 20 ml of Methylene blue indicator solution to the cylinder.
- Shake well and titrate against standard cetrimide solution taken in a burette.
- Titrate with intermediate shaking of the cylinder very well, the colour of the 2 layers exactly match each other, when viewed thorough diffused light.
- Note down the burette Reading (BR).

b) CALCULATIONS:

$$\% \text{ Activity} = \frac{1.5 \text{ X } 50 \text{ X} 100 \text{ X Mol.wt.of Anionic X BR}}{1000 \text{ X Mol. wt. of standard cationic X W}} = 2.5 \text{ X BR}$$

Where:

B.R = ml of 0.15% cetrimide consumed.
W = Weight of sample taken in gms
Molecular weight of standard cationic cetrimide = 364
Molecular weight of Anionic = 359