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DISINFECTION IN CASES OF SALMONELLOSIS

by

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1. Introduction

Universal interest in the problems of salmonellosis is understandable. At the present time this is one of the most widely distributed zoonotic and foodborne diseases affecting millions of people and producing severe economic losses. Various specific measures for prevention and control of infectious diseases (such as vaccination, therapeutic treatment, etc) are less effective in the case of salmonellosis. The introduction of the pathogenic agent into soil, manure, reservoirs and air, and its transmission by vectors such as insects, birds, rodents, etc, as well as through foods of animal origin, are important factors in the spread of salmonellosis and the conversion of sporadic cases into epidemics. In this connexion, disinfection, which is a method of destroying aetiological agents in the environment and of eliminating the risk of human and animal infection from the surroundings, is of great importance.

Since the World Health Organization in the Seventh General Programme of Work Covering the Period 1984-1989 (VPH/MTP/84.1) pointed out that "... foodborne diseases related to animals and animal products such as salmonellosis... are receiving high priority", the purpose of this paper is to provide practical recommendations on disinfection in animal husbandry, incubator and poultry stations and in the environment in cases of this zoonotic and foodborne disease

2. Stability of salmonellae in the environment

In broth, salmonellae were inactivated by heating at 60°C and 75°C for twenty-five and five minutes, respectively. In a physiological solution, S. typhimurium and S. cholera-suis were inactivated by heating at 70°C for five minutes. Thus, Salmonella stability to heating may vary depending on the species sensitivity and conditions of microflora. Salmonellae exposed to sun rays for 10 days were inactivated.

Salmonellae were found to be stable to drying. It was established that they retained viability in room dust, coal ash, dry manure, dry calves faeces for 80, 136, 90 and 185 days respectively, as well as in dry cattle faeces, mouse faeces and human faeces for four years, one year and 39 days, respectively.

Salmonellae survived on cloth in summer for 48 days (in daylight) and 62 days (in the dark); they survived on nylon stockings, box calf, metal and glass objects during 40, 38, 55 and 43 days, respectively.

Salmonellae were found to remain viable on wooden floors, wooden walls and feed boxes for 87, 78 and 108 days, respectively; S. pullorum survived on these surfaces for 62 days.

Salmonellae were shown to have a high stability to the action of sodium chloride, especially in a medium with protein content. In brine containing 20% of salt, salmonellae survived up to 6 months. Salmonellae exposed to 8% acetic acid solution were inactivated within 18 hours, whereas treatment with a 6% solution took 24 hours. S. dublin was destroyed by 1% lactic acid solution in 12 hours.

The stability to heating of salmonellae in food products, e.g. meat foods was found to be very high. It was shown that salmonellae in meat can be regarded as decontaminated by a cooking process which provided a temperature of 80°C in the centre of the piece of meat.

Salmonellae not only remained viable, but could also grow, in meat held in cold conditions (up to 4°C). Processes of salting and smoking meat had a low effect on salmonellae. Thus, in 30% salt brine salmonellae survived for 8 months at 8°C.

Salmonellae were shown to survive in smoked dry sausage (prepared by cold smoking - 16 days), in egg powder (at 70°C), in butter (at room temperature), in curds, sweets, chocolate bars, wafers, rye-bread, wheat bread, rolls and buns, especially in biscuits, during 48 days, 8 hours, 52, 16, 196, 174, 88, 3, 7 days and about two months respectively.

In milk, salmonellae survived for 6 days at 15°C and 37°C. They survived for 2.5 hours and 18 minutes at 50°C and 65°C respectively.

Recently, in connexion with intensification of animal husbandry, many specialists have considered manure and sewage from the large animal farms as important factors in salmonellosis epizootology.

Thus, S. enteritidis survived in cattle faeces for 180 days; S. abortus bovis was viable for 90 days, S. pullorum and S. cholera suis survived in dry litter and liquid pig manure for 240 and 80 days (in winter) and up to 27 days (in summer), respectively.

Salmonellae remained viable for about 180 and 45 days in sewage residue and decomposed residue respectively. In sedimentation tanks at the activated sludge stations, some Salmonella types remained viable for about 11 months.

Salmonellae were shown to survive up to 11 days in sea water; 84 days in ponds and lakes; 118 days in rain water; about 3 months in well water; and about 29 days in tap water.

3. Disinfection in animal husbandry

Experience in the prevention and control of salmonellosis has shown that the desired results from disinfection are unattainable without employing effective decontaminating agents.

Disinfection of animal and poultry houses was shown to be an important factor for maintaining stock free from infectious diseases. Before carrying out disinfection, animals should be moved out from the houses, then cleaning is conducted by means of mechanical methods, thereby removing manure and other contamination. Disinfection of houses should be conducted simultaneously with decontamination of the surroundings including land, all articles, implements, etc., which were in contact with the animals or were contaminated by their faeces. Disinfection may be considered to be preventive, current and terminal.

Preventive disinfection is conducted to avoid the introduction of infectious agents and, thus, not to allow their accumulation in animal houses and other objects, as well as to prevent a possibility of microflora change in animals and contamination of sheds, clothes and footwear of personnel, implements, etc.

Current disinfection is conducted systematically in farms from the time of first appearance of an infection case in animals, as well as each time a newly detected case is isolated, and when the usual examination of a herd with Salmonella infection is carried out.

All houses, standings, food troughs, implements, bedding, manure, footwear and clothes of farm personnel are subjected to the current disinfection process. Simultaneously, disinfection of territory is conducted, as well as of other places where the infected animals were kept.

Disinfectant barriers should be arranged at the entrance to an infected house (baths, mats, boxes with moisture-absorbant material), to decontaminate footwear of staff. These barriers should be filled daily with a suitable disinfectant (2% formaldehyde solution; chloramine solution, containing 2% active chlorine, 5% creoline water emulsion). In winter (at temperatures below 0°C), 10-15% sodium chloride is added to these solutions to prevent them freezing.

Terminal disinfection is conducted before the lifting of quarantine after eradication of salmonellosis in the farms. This type of disinfection is the final stage in the eradication of a pathogenic microorganism. In this process, the house is sprayed initially with a weak disinfecting solution or water; it is then carefully cleaned of manure andother contamination and proper disinfection is conducted.

In the process of preventive disinfection of animal houses, 5% hot calcinated soda solution may be used, or alternatively 3% creoline emulsion, 3% hot caustic soda-potassium carbonate mixture, 1% formaldehyde solution, 2% caustic soda solution, calcium hypochlorite solution (with 2% active chlorine). An effective preventive disinfection is also obtained with formaline aerosols (40% formaldehyde solution) at a rate of 10 ml of disinfectant solution per m^3 of a house with an exposure period of not less than 6 hours. In this case, for successful disinfection the environmental temperature should be within the limits of 12-15°C.

To conduct preventive disinfection of poultry houses, 5% calcinated soda solution, 2% caustic soda solution, 1% formaldehyde solution or other disinfectants could be used.

Since pests and rodents could transmit salmonellae, it is necessary on the farm to control flies (a treatment of 1% chlorofos solution*), as well as cockroaches (a treatment of 1% chlorofos solution*, applied every 6-7 days) and rodents (mechanical and chemical control methods employ zoocumarin*, zinc phosphide, etc).

Vehicles used for transporting cattle should be treated with 2% formaldehyde solution, 2% sodium hydroxide solution or calcium hypochlorite (2-3% active chlorine).

Clothing of personnel can be treated with 0.2% calcium hypochlorite solution or lime milk.

Where sick animals are detected, the current and terminal disinfections are carried out.

Current disinfection is conducted every 5-10 days before lifting quarantine or limiting measures. All implements should be subjected to daily disinfection. It is necessary to check that the disinfecting barriers are completely filled with 2% caustic soda solution or other disinfectants with addition of 10% sodium chloride in winter.

For current and terminal disinfection, 4% caustic soda solution is used to disinfect animal houses, as well as 2% formaldehyde solution, calcium hypochlorite solution (3% active chlorine content) etc.

In the case of salmonellosis in poultry (S. pullorum) 2% caustic soda solution or 1% formaldehyde solution are used.

Clay floors may be disinfected by means of 5% caustic soda solution or calcium hypochlorite solution (5% active chlorine).

Aerosol disinfection of animal houses may be achieved by using formalin at a rate of 15 ml per m^3.

Manure is decontaminated by the biothermal method for one month in summer and two months in winter.

4. Disinfection in incubator and poultry stations

4.1 Disinfection of incubators

Preventive disinfection of an incubator and its implements is conducted twice a year, at the beginning and end of the incubation season.

Preventive disinfection of incubators is conducted after a careful mechanical cleaning. During preventive disinfection, the ceilings and walls of the incubator house are whitewashed twice with 2% fresh slaked lime solution. Calcium hypochlorite solution or sodium hypochlorite solution (containing 2% active chlorine) can be used. The disinfection of incubator houses and implements contained therein could be conducted by aerosols of 40% formaldehyde solution. This type of disinfection is also used for decontamination of air at a rate of 20 ml of the above formaldehyde solution per m^3, for 3 hours.

Hatching incubators and implements are always disinfected at the end of the technological cycle, i.e. after every hatching of young poultry.

Among the recommended disinfectants are 10% calcinated soda solution or the chlorine-containing products. The most effective means is disinfection with formaldehyde vapours. For this purpose, 45 ml of water and 30 mg of potassium permanganate are used per m^3 of the internal incubator. The disinfection is conducted at the same temperature (37°C) and

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* These are the commercial names of the veterinary drugs used in the USSR.

humidity (68-70%) as those in the incubator. The incubator including its vents are immediately closed and remain so for not less than one hour. Incubator ventilation should be run for 5 to 10 minutes to spread vapours of disinfectant throughout the room, and then it should be switched off for the required exposure time. After disinfection, formaldehyde is neutralized by spraying the incubator floor with 25% ammonia solution. The quantity of this solution should be half of the quantity of the disinfectant used.

All implements are rinsed and disinfected in the washer room, where hot water and steam should be provided. To disinfect implements, 10% hot calcinated soda solution is used as well as other disinfectants.

In a suspected salmonellosis case in rearing young poultry, the flock should be taken away, and the treatment of the incubator by formaldehyde vapours should be conducted for one hour, to be followed by mechanical cleaning and repeated disinfection.

Before removing chickens from the incubator floor and trays, it is necessary to conduct cleaning (e.g. with a vacuum cleaner). After removal of chickens, incubation residues should be burnt and the incubators should be subjected to disinfection twice.

4.2 Veterinary sanitary treatment of eggs

Veterinary sanitary treatment of eggs could be conducted two to three hours after egg laying and before placing them in the incubator, as well as during incubation. However, disinfection of eggs before they are placed in the incubator should be considered as a compulsory measure.

Formaldehyde vapour and ozone have been shown to be the most acceptable means for mass egg disinfection. Before incubation, small egg lots can be disinfected by a wet method, i.e. by iodine chloride solutions.

Pedigree eggs are subjected to disinfection with formaldehyde vapours or ultraviolet irradiation. Disinfection with formaldehyde vapours could be performed in an incubator or special room at 37°C and 68-70% relative humidity, at a rate of 30 ml formalin (36-40% formaldehyde solution), 20 g of potassium permanganate and 15 ml of water per m^3 of the internal incubator (chamber) volume.

Chambers for egg disinfection should be spacious, with slotted shelves for ensuring good air circulation. This chamber should be equipped with two ventilators (one for circulation of the fumigant and the other for removal of it from tne chamber). A timer should be placed in this chamber with a bell to notify completion of the process.

Entering air to be disinfected should be filtered and heated to ensure that there is no supercooling of eggs.

After exposure time, 25% ammonia solution could be sprayed (at a rate of half the amount of formalin used) to neutralize the disinfectant.

Ultraviolet (UV) radiation was shown to be a quite effective means of disinfection as well as of promoting an increase in young poultry hatchability (by 4-8%) and an improvement of their vitality. Mercury discharge - quartz lamps are used for irradiation purposes.

The treatment of eggs by UV rays is conducted on two sides (from above and from below); eggs laid on the hatching trays are placed on a table at a distance of 40 cm from the above lamps. The lamp below is placed at a distance of 70-80 cm from the eggs. Chicken, turkey and duck eggs are irradiated for two minutes and goose eggs for three minutes.

In a house, where the hatching eggs are irradiated, forced and exhaust ventilation should be envisaged with 8-9 fold air change per hour.

4.3 Disinfection of packages for eggs

All circulating egg packages should be disinfected preferably in formalin vapour chambers of 100-500 m^3 volume (depending on the amount of packages loaded). The chamber should be equipped with: a ventilator for removing disinfectant after completion of the treatment; an air stove to heat air to 25-30°C; slotted shelves along the walls; a reservoir to sublimate formaldehyde or aerosol generator, and two hermetic doors on the opposite sides of the chamber. The internal chamber walls should be covered with heat insulating material. The premises in which a chamber is placed should consist of two isolated section, one for package recieiving and cleaning, and the other for unloading of decontaminated packages.

Cardboard, plastic or wooden boxes with interlayers inserted vertically at a distance of 0.5 cm are placed on the chamber shelves to enable a space between boxes to be 0.5-1 cm. Rolled cardboard boxes are placed on shelves vertically providing a space between boxes and interlayers of not less than 0.5 cm. After loading, 40% formaldehyde solution is sprayed into the chamber at the rate of 40 ml per m^3 for a one hour exposure time.

Another method of egg package disinfection is by mixing formaldehyde solution and calcium hypochlorite in a special vessel at the rate of 50 ml and 50 g respectively per m^3 of chamber and for 30 minutes exposure time. After disinfection is completed, formaldehyde is neutralized by ammonia solution or by intensive ventilation in a storehouse for one to two days.

4.4 Disinfection of cages and other containers for chicks

The above-mentioned procedure may be used for disinfection of chick cages and other implements. The most effective means of achieving proper decontamination is to have a suitable disinfection chamber available, in which the appropriate temperature and humidity can be maintained.

Clean cages for chicks should be loaded so that circulation of a disinfectant is uniform throughout. Sometimes, it is difficult to fumigate the standard egg trays, if they are made of very porous material. It is necessary to conduct permanent rejection of heavily contaminated and imperfect trays.

Metal and plastic boxes intended for poultry meat packaging should be cleaned before disinfection, washed with hot water and loaded into a chamber. Formaldehyde solution could be sprayed into the chamber at a rate of 30 ml per m^3 with a 30 minute exposure time.

Metal, wooden and plastic packages can also be disinfected by dipping into vessels containing, for example, calcium hypochlorite solution (0.5% active chlorine) or 2% sodium hypochlorite solution. Disinfection of these objects could also be done by spraying with the above disinfectants.

4.5 Disinfection of poultry down and feathers

Poultry down and feathers can be contaminated with salmonellae resulting in the spread of infection. In this case, to decontaminate infective feathers and down steam can be effectively used (at 105-110°C for 10 minutes), as well as formaldehyde (3% solution, 45-50°C).

Disinfection by means of any of the above methods should be arranged so that loading of infective material and unloading of decontaminated material should be conducted in separate rooms.

5. Disinfection of soil, runs and roads

In poultry farms, the surroundings, runs, solaria, ground floors, and roads should be subjected to preventive and forced disinfection.

To conduct preventive disinfection, 2% hot caustic soda solution or chlorine-containing disinfectant solutions (2% active chlorine) are recommended for use.

To conduct forced disinfection and decontamination of the soil surface, 20% fresh-hydrated lime suspension, calcium hypochlorite suspension (2% active chlorine) or 2% caustic soda solution may be used.

In ground floor houses, it is necessary to remove the upper layer of soil (about 10 cm) and decontaminate the floor with calcium hypochlorite solution (5% active chlorine), at a rate of 3 litres of disinfectant per m^3 of floor surface. Then, the disinfected floor could be covered by a fresh soil layer.

The territory of runs should be carefully cleaned of down, feathers and litter; these residues should be burnt. The upper layer of soil is then removed and the run surface is sprayed with calcium hypochlorite solution (2% active chlorine), at the rate of 2 litres per m^2 of surface and a fresh soil layer should be spread on the disinfected surface. Poultry should not be allowed on these runs for two to three weeks after disinfection.

Hard covered roads should be disinfected by spraying 2% caustic soda solution or calcium hypochlorite solution (2-3% active chlorine).

6. Disinfection of vehicles used for transportation of animals and poultry

After manure and solid residues have been removed, the vehicles should be carefully cleaned by washing with a pressure water jet. Manure and solid residues originating from vehicles, spur tracks and stalls should be removed to authorized places where they are subjected to biological decontamination. Liquid waste (e.g. rinsing water, urine) should be treated at sewage treatment plants.

Disinfection of motor transport is carried out at specially arranged hard covered ground. In cold seasons, treatment is performed in special disinfecting houses.

To conduct wet disinfection of transport, 1.5-2% formaldehyde, or mixture of 3% caustic soda with hypochlorite (2% active chlorine) solutions could be used. Calcium hypochlorite solution (2-3% active chlorine), 2% formaldehyde solution or 2-4% hot caustic soda solution may be used for disinfection of carriages.

In the case of aerosol disinfection of trucks, 38-40% formaldehyde solution (formalin) is recommended for use (30-40 ml/m^3 for an exposure time of 60 minutes). Aerosol disinfection is conducted in a hermetic chamber of a special disinfecting house.

Disinfection of ships could be carried out with calcium hypochlorite solution or hypochlorine solution (2% active chlorine) as well as with caustic soda solution (4% concentration) heated to 60-70°C or 2% formaldehyde solution, at the rate of 0.5 litres per m^2 of surface.

7. Disinfection of equipment in dairy farms and dairy plants

Milk is a favourable medium for the growth of Salmonella and could be a source of human infection. Therefore, sanitary treatment of equipment in dairy farms has a direct public health significance.

In the case of portable milking apparatus, the following procedure is recommended after milking:

- all milk residues should be rinsed off with warm water;
- after rinsing with water, hot detergent or disinfectant solutions should be poured into a bucket and, under vacuum, they should be circulated through the milking apparatus;
- the final operation is rinsing the apparatus with hot water from all disinfectant residues.

Milking buckets, milk cans, milk gauge rods and other vessels should be subjected to sanitary treatment. It is best to treat all vessels with milk can steamers. For disinfection of milking apparatus, 0.1-1% calcinated soda solution or 0.5-1% detergent powder, etc., may be used.

The sanitary treatment of milkers could be conducted by means of an automatic washer in the following sequences:

- washing with warm water to remove milk residues;
- washing with hot detergent (e.g. 0.5% calcinated soda solution);
- rinsing with hot water.

The milker should be periodically disinfected. For this purpose, sodium hypochlorite could be used (200-250 mg of active chlorine per litre at 40-45°C).

Milk cisterns, tanks, separators, bottling centrifuge cleaners, bottles, buttermaking and packing machines, and pasteurizers at dairy plants, should be subjected to careful washing and disinfection.

Synthetic detergent powders have been shown to give excellent cleaning and disinfecting effects, as well as sodium hypochlorite.

Superheating steam could also be effectively used (for 10 minutes) for disinfection purposes. Other chemical disinfectants include iodofors and quaternary ammonium compounds. Iodofors are always mixed with a detergent in acid medium, therefore, they are especially suitable for conducting disinfection where an acid cleaner is needed. These substances possess a rapid effect and a broad antimicrobial activity. To disinfect clean surfaces, a solution of about 25-50 mg of active iodine per litre is needed. Iodofors have a corrosive effect on metals, therefore the careful rinsing of surfaces is necessary after the appropriate contact period. These substances are easily inactivated by organic material. Iodofor effectiveness is observed visually with their colours disappearing when the residual iodine is reduced to a noneffective level. When used in the correct concentration, they are non-toxic and possess weak taste and smell. However, they cause spoilage of food product substances. Therefore, the equipment should be carefully rinsed after use of these disinfectants.

Quaternary ammonium compounds have good characteristics as disinfectants and detergents. They are colourless, have some corrosive effect on metals and a very low toxicity but have a rather bitter taste. They are less effective against gram-negative bacteria than chlorine-containing disinfectants and iodofors. Solutions of these compounds tend to adsorb to surfaces, therefore the careful rinsing of surfaces is necessary. These substances should be used at a concentration of about 200 mg/litre. They are easily inactivated by magnesium, soaps and anionic detergents.

8. Disinfection in slaughterhouses

Preventive disinfection in a stockyard could be carried out with 2% caustic soda or caustic potassium solutions or by calcium hypochlorite solution (2% active chlorine), 2% formaldehyde solution, etc.

In slaughterhouses, knives used for blood collection should be treated with 0.5-1% caustic soda solution; filters should be disinfected with 0.3% caustic soda solution. Blood containers are sprayed with 1% caustic soda solution or 2% active chlorine-containing solution, and then thoroughly washed.

Cisterns and blood basins should be rinsed with water and are then disinfected with 0.1% chloramine solution or are washed with hot water. Live steam can also be used for disinfection.

In sausage departments, the floor and equipment are cleaned with 1% calcinated or caustic soda solutions before disinfection. The working places are wiped daily with one of the following disinfectants:

- calcium hypochlorite solution (0.2% active chlorine);
- 0.2% chloramine solution;
- 0.5% hydrogen peroxide.

Small metal implements should be preferably sterilized by steam.

In departments producing biopreparations, 0.2% chloramine solutions could be used for disinfection purposes as well as calcium hypochlorite solution (0.2-0.3% active chlorine) or 0.5% hydrogen peroxide solution, etc.

Forced disinfection is always conducted at slaughterhouses where sick animals are found at a stockyard or in case of Salmonella detection on the conveyor line or during processing of animal products.

The stockyard premises should be disinfected with 2% caustic soda (caustic potassium) solution, calcium hypochlorite solution (2% active chlorine) or 2% formaldehyde solution.

In slaughter premises and other departments of meat enterprises, 2% caustic soda solutions are used for one hour, as well as other chemicals for disinfection of floors, walls, etc. To decontaminate steel, wooden and cement surfaces infected by salmonellae, calcium hypochlorite solution (3% active chlorine), hypochlorine solution (2% active chlorine) could be successfully used for a one hour exposure period.

In meat and bone producing shops, the equipment could be decontaminated by 4% caustic soda solution at the rate of 1 litre/m^2 for one hour. After disinfection, all surfaces should be washed with water to remove residues of disinfectants.

9. Disinfection of overalls and footwear

Overalls of personnel who look after the animals as well as of workers dealing with raw animal materials, can be an important route of salmonellae transmission. The bacteria survived on cambric, cotton fabric, sacking, capron and cotton stockings, tarpaulin, waterproof capetent, sheep skin, box calf skin, and felt boots for quite a long period of time (about two months).

Cotton fabrics and tarpaulin overalls could be disinfected in steam chambers, autoclavers, by boiling as well as by chemical disinfectants. In large slaughterhouses disinfection of overalls is conducted by fluctuating steam or formaldehyde in special chambers.

In country conditions, disinfection could also be done by formaldehyde vapour which can be produced by mixing formaldehyde solution and calcium hypochlorite. Boiling water is also a very effective means of disinfecting overalls in country conditions.

Decontamination of tarpaulin, cotton and felt articles could be conducted by soaking them in a disinfectant solution (e.g. 2% chloramine). The same method of disinfection could be used for cords, horse clothes, rubber boots and rubbers, brushes, scrapers, buckets, brooms, spades, etc.

Overalls, cloaks, tarpaulin covers (for lorries) of water proof canvas are disinfected with chloramine without adding wetting agents, or by steam, or by aerosols of formaldehyde.

10. Control of disinfection quality

When conducting disinfection it should be kept in mind that its final task is to eliminate a causative agent in the site of infection, and those objects which were contaminated. Much research has shown that E. coli can serve as a test microbe for grade estimation of disinfection effectivety. This bacteria is a ubiquitous environmental agent and similar to the intestinal infectious agents including salmonellae by its resistance to disinfectants. It could be isolated in 100% of cases in different objects of animal husbandry, meat, poultry, slaughter and processing plants, before disinfection.

For control of disinfection efficacy, microbiological samples should be taken from various sites, e.g. floor surfaces, walls, equipment. These samples should be taken with sterile cotton wool wads wet in sterile neutralizing solution.

It is important to neutralize the remaining activity of the disinfectant before microbiological analysis of the samples. For this purpose, in case of disinfection with caustic soda solution, acetic acid solution is used as a neutralizing agent; in case of formaline, calcium hypochlorite and alkaline, neutralizing solutions are ammonium hydroxide, hyposulphite solutions and a mixutre of acetic acid and ammonium hydroxide solutions respectively. Neutralizing solutions are prepared in concentrations of one tenth that of the disinfectant. The samples, after neutralization should be examined in the microbiological laboratory on the same day.

Disinfection is considered as satisfactory if there is no growth of test microbes (E. coli) in all samples examined. To check the quality of final disinfection, it is necessary)to examine samples for the presence of salmonellae.