

Appendix Two

Guidance for evaluation on on-site low-temperature disinfection of cold chain food outer packaging

A. Evaluation principle

On-site low-temperature disinfection evaluation includes process evaluation and effect evaluation. Process evaluation shall be carried out along with low-temperature disinfection for each time, and self-examination and self-evaluation are generally carried out by disinfection implementation units. Relevant regulatory authorities can conduct spot checks on disinfection process and self-examination and self-evaluation process to ensure the effectiveness of disinfection process. Regular spot checks are generally adopted for effect evaluation, and it is recommended to evaluate once every six months. Whenever changing the low-temperature disinfection method, the disinfection effect shall be evaluated, and it can be put into use only after proving that the low-temperature disinfection is effective.

B. On-site low-temperature disinfection evaluation

Disinfection implementation units shall make good records of disinfection and conduct self-evaluation in the process of disinfection for each time, which includes evaluating whether the whole disinfection operation is carried out according to the disinfection work plan, whether the low-temperature disinfection products used are legal and effective, whether the disinfection methods match the disinfection objects and environment, whether the disinfection parts are fully covered, whether the usage amount meets the requirements, whether the actuation duration of disinfection is sufficient, and whether the records of disinfection are made standardized. The contents include but are not limited to disinfection date, disinfection location, disinfection scope, disinfection object, disinfection procedure, disinfectant preparation, disinfectant concentration and dosage, actuation duration, disinfection method, disinfection equipment and personal protection etc.

The low-temperature disinfection products used shall meet the requirements of relevant national health standards and specifications, and pass the health and safety evaluation. Details of disinfectant include disinfectant name, main active ingredients and dosages, validity period, preparation method, applicable scope and applicable method etc. Details of disinfection equipment include equipment name, main sterilization factors and the intensity, scope of usage and method of usage etc.

C. Effect evaluation on on-site low-temperature disinfection

a) Evaluation objects and indicators

Effect evaluation object of low temperature disinfection is the surface of the object. According to the resistance of new coronavirus to disinfection factors, indicator microorganisms are selected, and the killing rate of indicator microorganisms is taken as the evaluation index. It is indicated that the resistance of microorganisms shall be equal to or higher than that of new coronavirus, easy to cultivate and meet the laboratory biosafety and WS/T683 requirements. *Staphylococcus aureus* (ATCC 6538) and *Escherichia coli* (8099) can be selected for chemical disinfection. Indicator microorganisms meeting the above requirements shall be selected for the physical disinfection according to the characteristics of disinfection factors.

b) Evaluation method

In accordance with GB/T38502, the bacterial tablets shall be prepared (tryptone

soybean broth is used as organic interfering substance for the on-site low-temperature disinfection evaluation), and the number of recovered bacteria in each bacterial tablet shall reach 1×10^6 CFU/tablet $\sim 5 \times 10^6$ CFU/tablet. Put the bacterial tablets of indicator microorganism into the corresponding low temperature environment for at least 30min, and ensure that the indicator microorganism reaches the same low temperature before proceeding to the next step.

Before disinfection: place the bacterial tablets on the site, with the desktop, door handles and buttons as the key objects, and each type of object shall have no less than two samples; The outer packaging of cold chain food shall be distributed on all six sides of the outer packaging; The total number of test samples shall not be less than 30.

After disinfection: After disinfecting to the actuation duration, use sterile tweezers to move the bacterial tablets into a test tube containing 5.0 mL of corresponding neutralizer, vibrate 80 times in the palm of hand or mix evenly with a blender, and neutralize for 10min. Meanwhile, a positive control group can be set up.

Laboratory cultivation: the sampling tube shall be oscillated on the mixer for 20s or vibrated forcibly for 80 times, and 1.0 ml of samples to be tested shall be inoculated in sterile plates, and two plates shall be inoculated in parallel for each sample. 15ml \sim 18ml of dissolved culture medium at $45\text{ }^{\circ}\text{C} \sim 48\text{ }^{\circ}\text{C}$ shall be added, and shake evenly while pouring. After the coagulation, it shall be cultivated for 48 hours at $36\text{ }^{\circ}\text{C}$ or plus-minus $1\text{ }^{\circ}\text{C}$ of $36\text{ }^{\circ}\text{C}$, colony counting, calculate the killing rate.

c) Result judgment

The average killing rate of indicator microorganisms on the surface of object is $\geq 99.9\%$, and the number of samples with killing rate $> 99.9\%$ accounts for more than 90%, which is judged as qualified for disinfection.

D. Attentions

- a) Combined with the characteristics of the location, clearly defined the disinfection objects, and strictly implement the disinfection work in accordance with the disinfection procedures and norms.
- b) Disinfection implementation units shall have on-site disinfection ability, and operators shall receive professional training in disinfection, master the basic knowledge of disinfection and personal protection, and be familiar with the use of disinfection equipment and the preparation of disinfectants etc.
- c) All on-site disinfection shall be recorded and kept for at least 2 years, and self-monitoring shall be carried out at the same time. Attention shall be paid to standardized operation in the process of effect evaluation, harmless treatment shall be strictly taken on samples and related test materials in accordance with biosafety requirements.
- d) When disinfecting on site, personal protection shall be done well, and regular and effective personal protective equipment shall be selected according to the site conditions and relevant standard requirements.