

International Journal of Biotechnology and Food Science Vol. 2(3), pp. 53-60, April 2014

ISSN: 2384-7344 Research Paper

# Characterization and control of microbial contaminations in cold dishes

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Accepted 24th January 2014

**Abstract.** This paper investigates cold dishes microbial contamination and the quality of health changes in food production and cold storage process. Bacteria reproduce rapidly in cold dish and its hygiene has become a long time question. Detection of bacterial colony for gold hook cucumber of various major raw material and finished products was carried out. Hot boiling water was used to wash the main raw materials, statistical reduction in strains rate was observed. Flora analysis was made, and the shelf life drawn up. The total number of colonies in gold hook cucumber was 7.6 × 10<sup>4</sup> CFU/g, and cucumber occupied 83%, while dried shrimp meat occupied 17%; after blanching, the total number of colonies in the improved formulation was 2.8 × 10<sup>4</sup> CFU/g, the sterilization rate reached 63%, and the efficacy was obvious. In conclusion, cold dish kitchen production by microbial pollution is very serious, using hot boiling water to wash the main raw materials can help control the bacterial count in some degree, then prolong the shelf life and reduce the risk of food poisoning, which do a lot of benefit to the catering industry the food safety management.

**Keywords:** Gold hook cucumber cold dish, storage period, bacterial colony, food safety.

#### INTRODUCTION

Cold dish has a long historical culture; it can be traced back to the Zhou Dynasty and the Qin dynasty. In the hot summer, cold dish is an integral part of daily food, cold food is not only tasty, is easily manufactured, and can maximize the preservation of vegetable nutrition and flavor, greener and healthier. In the increasingly rapid pace of life today, with a simple cooked cold dish is an excellent choice. However, because varieties of vegetables are different, not each vegetable is suitable for cooling, in consideration of the summer high temperatures, bacteria reproduce rapidly. Thus, we should give enough attention to the cold food hygiene and storage. Vegetables materials should be washed with clean water, if not fresh to make cold dish, plus cleaning disinfection, using this gold hook cucumber will lead to gastrointestinal diseases. Cold dish are made with mostly fruits and vegetables, in the processing fresh-cut must be carry out. However, most of the production of raw materials with high moisture, nutrient-rich, in suitable conditions, is conducive to the growth of microorganisms (Sapers et al., 1990). Coupled with the cold soup in the processing and consumption of the process, it is almost impossible to implement heat sterilization. While this makes raw food dishes retain the nutrients in raw materials, at the same time, safety and health cannot be guaranteed; and the risk of food poisoning is great (Jiang et al., 2007). So, make sure to use fresh vegetables which should be cleaned, best scalded with boiling water; this can kill some unwashed residual bacteria and parasite eggs. Therefore, this study expands the research on golden hook cucumber's materials, finished product colony species and manufacturing method; in addition to analyzing the relationship between the growth and production method, and drawing some relevant guiding conclusions.

#### **MATERIALS AND METHOD**

#### Food materials

Cucumber, dried shrimp meat, garlic, cooking wine, salt,

Table 1. Basic formula of gold hook cucumber.

Materials	Cucumber	Dried shrimp meat	Cooking wine	Garlic	Salt	Monosodium glutamate	Pomade
Usage amount (g)	300	50	10	10	1.5	1.0	1.0

monosodium glutamate pomade, all of the condiments were bottled or bagged products and bought from Aachen supermarket in Yang Zhou.

#### Reagents and culture medium

The reagents used include beef extract, peptone, yeast extract, glucose, lactose, D-mannitol, agar, bile salts, Fan red; NaCl, NaOH, I2, Kl, and anhydrous magnesium chloride, anhydrous potassium sulfate, magnesium sulfate, manganese sulfate, calcium carbonate, hydrogen phosphate, dipotassium and diammonium citrate, sodium acetate, ethanol, AR grade; Gram staining solution configured according to the literature (Jiang et al., 2007).

Nutrient agar: peptone 10 g, beef extract 3 g, NaCl, 5 g, agar 18 g, water total volume 1000 ml, pH 7.2 to 7.4, at 121°C for 15 min, all of these are for the separation and the counting of the total bacterial colonies. For culture collection, agar is reduced to 8 g, the rest are the same.

PSA medium: peptone 20 g, anhydrous magnesium chloride 1.4 g, anhydrous potassium persulfate 1 g, agar 13.6 g, glycerol 10 ml, distilled water 1000 ml, pH 6.9 to 7.1, at 121°C for 15 min; culture conditions of 25°C for 48 h, the culture conditions is for the separating and counting of the Pesudomonas.

MRS medium: 10 g of peptone, 10 g of beef extract, yeast extract 5 g, dipotassium hydrogen phosphate 2 g, diammonium citrate 2 g, sodium acetate 5 g, glucose 20 g, Tween 80 1 ml, magnesium sulfate 0.58 g, manganese sulfate 0.25 g, agar 15 g, calcium carbonate 20 g, distilled water 1000 ml, the pH is 6.2 to 6.4, at 121°C for 15 min; culture conditions of 30°C for 48 h for isolation and counting of the lactic acid bacteria.

VRBGA medium: yeast extract 3 g, peptone 7 g, bile 1.5 g, NaCl 5 g, lactose 10 g, neutral red 0.03 g, the crystal violet 0.002 g, agar 15 g, 1000 ml of distilled water, pH was adjusted between 7.3 and 7.5, 121°C at 15 min; culture conditions 30°C for 48 h, for isolation and counting of the Enterobacteriaceae.

MSA medium: beef extract 1 g, peptone 10 g, 10 g of D-mannitol, NaCl 75 g, agar 13 g, phenol red 0.025 g, distilled water 1000 ml, pH 7.2 to 7.6, plus 1% of the phenol red solution 2.5 ml mix, at 121°C for 15 min; culture conditions of 30°C for 48 h, to determination aureus and Micrococcus (Jiang, 2007).

#### Protocol of a basic formula of gold hook cucumber

A basic formula of gold hook cucumber according to the literature is given in Table 1.

#### Preparation of gold hook cucumber samples

Soak the dried shrimp with cooking wine for a while. Wash cucumber, then cut it into thin pieces; add garlic, salt, monosodium glutamate, then transfer them into the sauce. Preparation is done (Zhang and Ding, 1994).

Load up the plate with dried shrimp meat and cucumber, then add vinegar and just hold off stirring in the pomade until you are ready to serve.

### Gold hook cucumber contamination investigation and the source analysis

Sample the raw materials and the finished goods of respectively 25 g with sterile procedure. Make 1:10 incremental dilution, 1 ml pour plate. Make two plates for each dilution, pour nutrient agar approximately at 45°C 15 ml, and rotate the dish to mix the solution; solidify the medium and then invert the medium in the incubator at 37°C for 24 h, then get it out and count.

The number of bacteria in the main raw material is taken as the variable, the usage as weight, and measure the source of finished bacteria number by the weighted average method.

### Control of the number of bacteria in gold hook cucumber

Put the cucumber and coriander into hot water, keep for 5 min and then take statistics of the rate of bacteria reduction.

# The change in health quality of the gold hook cucumber during refrigeration:

Make the unit of 25 g cold dish samples respectively by the basis recipe and improved formulation, place the unit placed in a sterile petri dish, cool and preserve them at 4°C in the refrigerator, and get them out per 24 h, by time sequence measure the number of bacteria and graph the growth curve. Do sensory test of the product at the same time, to evaluate the quality of their health, by which the gold hook cucumber shelf life is built.

#### **RESULTS**

# Contamination investigation and the source analysis of gold hook cucumber

As shown in Table 2, the number of bacterial colonies of

<b>Table 2.</b> Date for the number of bacteria in raw materials and finished product
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Products	Weight (g)	Total number of bacterial colonies (CFU/g)
Cucumber	300	8.8 × 10 <sup>4</sup>
Dried shrimp meat	50	$7.4 \times 10^4$
Finished product	350	$8.6 \times 10^4$

Table 3. Date for the number of bacteria after blanching.

Items	The total number of bacterial colonies (CFU/g)	Total number of bacterial colonies after blanching (CFU/g)	Bacteria reduction (%)
Cucumber	8.8 × 10 <sup>4</sup>	$3.1 \times 10^4$	65
Dried shrimp meat	$7.4 \times 10^4$	1.0 × 10 <sup>4</sup>	86
Finished product	$7.6 \times 10^4$	$2.8 \times 10^4$	63

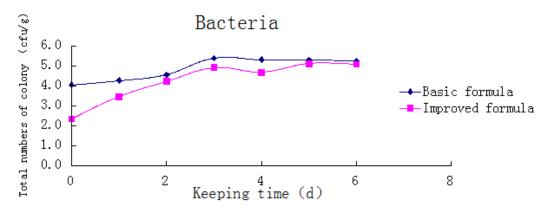


Figure 1. The changes for the number of bacteria during the refrigerated process of gold hook cucumber.

cucumber and dried shrimp meat were  $8.8 \times 10^4$  and  $7.4 \times 10^4$  CFU/g, respectively.

Dried shrimp meat belongs to the field of aquatic product processing products. These nourishing foods with rich nutrition are easily polluted and suitable for bacterial growth.

The bacterial number which has different types of raw material is the variable, usage amount of weight. The total bacterial count of the gold hook cucumber by the basic formula is  $8.6\times10^4$  CFU/g; cucumber occupies 88%, while dried shrimp meat occupies 12%

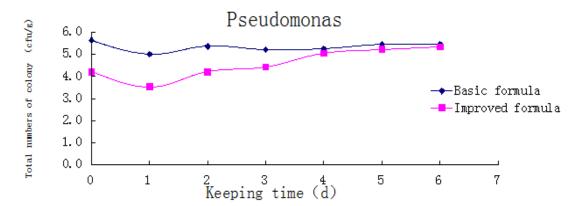
#### Control of the number of bacteria

According to the data in Table 3, the bacteria reduction after blanching was recorded, and the most significant was shrimp meat which gave 86%. Meanwhile, cucumber alone recorded 65% reduction in bacteria, and the finished product showed 23% reduction. It reflects that blanching is a very effective means of reducing bacteria, which played an important role in controlling the bacteria number of gold hook cucumber.

Changes in the quality of health during the refrigerated process of gold hook cucumber

The changes for the number of bacteria during the refrigerated process of gold hook Cucumber: From Figure 1, during storage, the number of based and improved formula bacteria in 1 to 3 days rise slowly; in the 4 to 6 day tend to be stable and with a weak decline. Low temperature inhibited the growth of bacteria, and in initial stage the number of improved formulation bacteria was significantly less than the number of based formulation bacteria, illustrating that blanching effect for reducing the number of bacteria is obvious.

The changes for the number of Pseudomonas during the refrigerated process of gold hook cucumber: Figure 2 shows that the basic formula of Pseudomonas were in a downward trend in the first day, then the basic formula keep a high trend. Low temperature environment on inhibiting the growth and reproduction of bacteria Pseudomonas cells is obviously. After the first day of decline, improved formula in 2 to 6 days showed a steady upward trend. In the whole growth process, Pseudomonas



**Figure 2.** The changes for the number of Pseudomonas during the refrigerated process of gold hook cucumber.

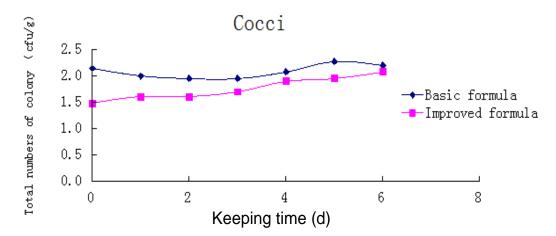


Figure 3. The changes for the number of cocci during the refrigerated process of gold hook cucumber.

with a higher order of magnitude, became the dominant spoilage bacteria.

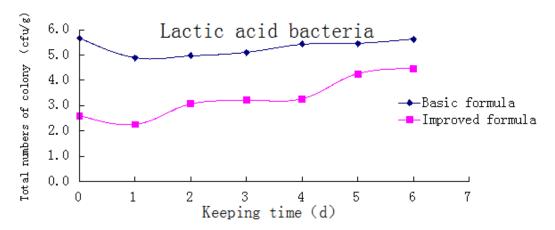
The changes for the number of cocci during the refrigerated process of Gold Hook Cucumber: Figure 3 shows that the basic formula in 1 to 3 days has a stable growth then along with a slight decline, and the improved formula cocci number showed an upward trend; however, the total number was fewer than other flora minimals, so the cocci did not play a dominant role in the corrupting process.

The changes for the number of lactic acid during the refrigerated process of gold hook cucumber: Figure 4 shows that the basic formula and improved formula of lactic acid bacteria were in a downward trend in the first day, The number of basic formula colonies growing a regular flat in the 2 to 6 days The number of improved formula lactic acid bacteria growth slowed in 1 to 4 days, then with a rapid growth momentum and got the maximum on the sixth day. And, the number of improved

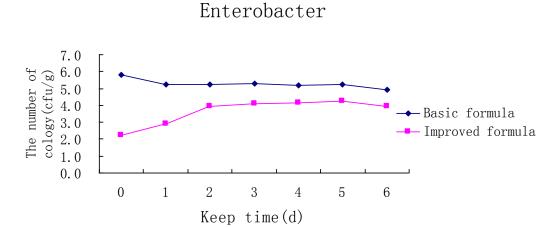
formulation bacteria was significantly less than the number of based formulation bacteria, illustrating blanching effect for reducing the number of bacteria is obvious.

The changes for the number of enterobacter during the refrigerated process of gold hook cucumber: Figure 5 shows that the basic formula of enterobacter were in a downward trend, it can be inferred that enterobacter growth is at a disadvantage in the competition process. Improved formula colonies grow rapidly in 1 to 2 days, the relative balance of 3 to 6 days. Found by the two formulations compared, the number of enterobacter in the storage period is low, In addition to temperature, but also on antagonistic microflora may be relevant.

The changes for the number of microzyme during the refrigerated process of gold hook cucumber: Figure 6 shows that, although the number of microzyme was in a certain proportion during the whole storage period, the



**Figure 4.** The changes for the number of lactic acid during the refrigerated process of gold hook cucumber.



**Figure 5.** The changes for the number of enterobacter during the refrigerated process of gold hook cucumber.

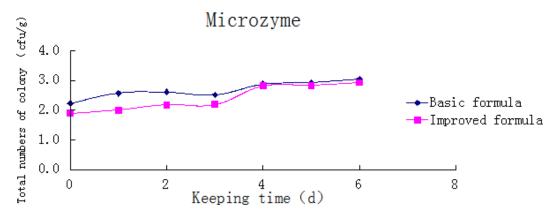


Figure 6. The changes for the number of microzyme during the refrigerated process of gold hook cucumber.

absolute content and relative content were gradually declining. It may be possibly relative to the suitable

growth temperature of microzyme. The most suitable temperature range is 20 to 30°C; it will be not conducive

Table 4. Phenotypic characteristics of typical bacteria.

Identified bacteria	Colony characteristics	Staining results
Enterobacter	In VRBGA medium purple color, 2 to 3 mm in diameter, smooth surface moist, neat edge	Gram-negative bacilli, no spores, no capsule, surrounded by flagella
Pseudomonas	The PSA was translucent colonies on the medium, Smooth and moist	Gram-negative, no spores, small cell
Lactic acid bacteria	The colonies on MRS medium diameter 0.5 to 1.5 mm, low convex, marginal integrity	Rod-shaped cells, do not form endospore, gram-positive
Microzyme	In high salt Czapek medium was milky white, Colony 3 mm cream, moist, sticky, marginal integrity, colonies of about 3 mm	Single individual cell, oval, no flagellum, big cell
Cocci	MSA medium yellow, colony of about 1mm, Smooth and moist, neat edge	Gram-positive, no flagellum, no spores, quadruple

Table 5. Pseudomonas identification test.

Test	P (basic formula)
Determination of catalase	+
Oxidase test	+
Fermentation of glucose	+ (gas production)
Utilization of carbon sources	+
Determination of amino acid decarboxylase	-
Fructan formation test	-
Nitrate reduction test	-
Hydrolysis of starch	-
Liquefaction of gelatin	+
Identification results	Pseudomonadaceae

Note: "+" expressed positive, "-" expressed negative.

to the growth when the temperature is lower than 4°C.

### Gold hook cucumber bacterial genus identification test

# Identification of lactic acid bacteria in the preservation process of gold hook cucumber

The identification of lactic acid bacteria is given in Tables 4 to 8.

#### DISCUSSION

#### Sources of bacterial hazards of gold hook cucumber

Source of cold dish bacteria can be broadly divided into two categories. The first is the original bacteria, which are the ones present in the raw material itself, for example, aquatic bacteria widely distributed around the world in the water environment, thus the fish and shellfish living in these waters will be hold different bacteria (Lin, 2005). The other is non-original bacteria itself, which is related to the unsanitary operation conditions, the most common route of infection is through self-contaminated soil and the spread of food processors, and human and animal feces and domestic waste water pollution, kitchener's hands, condiment, water, air, insects and so on (Jiang, 2008).

The materials of cold dish contain some original bacteria; some raw materials during processing without passing through hot boiling water treatment are bound to retain various types of bacteria, thus increasing the chances of food poisoning (Torok et al., 1997).

#### Control of bacterial hazards of cold dish

Approximately 35% of fruits and vegetables corruption are caused by bacteria, and therefore the prevention and

Table 6. Enterobacter identification test.

Test		E (basic formula)	
Fermentation of glucose		+ (gas production)	
Lactose fermentation		+	
Mannitol fermentation		+	
Sucrose fermentation		+	
Maltose fermentation		+	
Dynamic test		+	
Hydrogen sulphide test		-	
Indole test		-	
V-P test		+	
Utilization of carbon sou	rces	+	
Determination of catalas	se	+	
Oxidase test		-	
Citrate test		+	
Triple sugar iron test	Cant	+	
Tripic sugai iron test	Bottom	+	
Gas production		+	
Hydrogen sulfide		<u>-</u>	
Identification results		Enterobacter	

Note: "+" expressed positive, "-" expressed negative.

Table 7. Experimental identification of lactic acid bacteria.

Project	E (basic formula)		
Gram stain	+		
Catalase test	+		
Fermentation of glucose	+ (gas production)		
Identification results	Listeria spp. (Ling and Dong, 1999)		

Note: "+" expressed positive, "-" expressed negative.

Table 8. Cocci identification test.

Experimental project		C-1 (basic formula)
Gram stain		+
Cells arranged		Irregular clumps
Clusosa matabaliam in tuna	Acid production	+
Glucose metabolism in type	Gas production	-
Power experiment		-
Determination of catalase		+
Identification results		Staphylococcus (Dong and Miaoying, 2001)

Note: "+" expressed positive, "-" expressed negative.

control of bacteria particularly is important. The main control measures include: strengthening health education, changing bad hygiene practices; focusing more attention on summer and autumn. Cutting off the route of

transmission, and strengthening the food sanitary inspection, their production, processing, storage and preparation processes, such as scientific management prevents pollution, control reproduction and kill the pathogen

(Bai, 2010).

Erwinia and Pseudomonas play an important role in the spoilage of fruits and vegetables. Cucumber is also possible to be infected before harvest. In these bacteria caused corruption, *Erwinia carotovora* is most important. It can occur on the plants before harvest, often irrupted through damaged tissue, and major tissue can be damaged in a few days. Therefore, the good raw material purchasing is very important. To choose good color vegetables, appearance of no damage, fresh and smell good, artificial cooling method can be used for storage. Make sure that the vegetables are kept at low temperature. Storage cellar, ventilation, plastic film packaging can also be used (Liu, 2006). The experiment should be selected dried shrimp meat with clean appearance, pale yellow color and luster, shrimp tail bend downward like a hook, succulent dense hard and with no odor (Liu, 1991).

Special attention is required on the contaminated fish and shellfish which microbial species is very different from the classes of livestock and poultry; with growth in different waters, microbial species will be different. Since the reason for survival in the waters is low temperature, most pollutions are caused by low temperature bacteria. These bacteria can multiply quickly at 5°C low temperature conditions, but their breeding is relatively slow above 30°C. Therefore, using the low temperature the storage will also have reproduction microorganisms; cold dish should be blanched before processing (Li, 2009).

The experimental results show that it is necessary to control raw materials bacteria of cold dish, and is also very effective. Control of bacterial hazards of cold dish. cannot do well without regular hygiene work, for example, human health, hand washing disinfection, with special container, cryopreservation of food and raw materials; especially, the personnel to develop sterile operation consciousness, when cutting and seasoning to avoid hands direct contacting with food (Jiang, 2008). Besides, production, food raw materials processing, transportation, storage process, to strictly abide by the rules, do a good job cleaning disinfecting preparations, to prevent the production and operation environment, personnel, equipment, containers, and materials such as cross-contamination and secondary pollution (Jin et al., 2006).

#### Establishment of cold dish food safety standard

Due to the particularity of cold dish raw materials, if not in accordance with the standards of hygiene and safety operation, easily lead to the occurrence of foodborne illness.

Building dish food safety standards will have a positive impact on rational management of catering industry, enhancing the supervision and law enforcement, safeguarding the legitimate rights and interests of broad consumer. Efforts should be made to establish new concept of food safety supervision and management, based on risk assessment, set up a suitable to our own national conditions and international standards of safety standard system (Han, 2007).

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