





SPECIES OF CONCERN IN FOODS

- ◆ **TNase (thermostable nuclease)** and **coagulase-positive strains** are the only staphylococci that **demand further investigations** when found in foods, but the existence of both TNase and coagulase-negative enterotoxin-producing strains has been known for some time.
- ◆ **Table 23-1**
- ◆ **Staphylococcal enterotoxin (SE)**



HABITAT AND DISTRIBUTION

- The staphylococcal species are **host-adapted** (宿主適應型) with about one-half of the known species inhabiting humans solely (e.g., *S. cohnii* subsp. *cohnii*) or humans and other animals (e.g., *S. aureus*).
 - host-adapted: ability of a pathogen to colonize and cause disease in a particular host population.
- Often found on the **skin** and **openings** (開口處) of humans and other organisms.



HABITAT AND DISTRIBUTION

- The two most important sources to foods are **nasal carriers** (鼻腔帶菌者) and individual whose hands and arms are inflicted (遭受) with boils (瘡) and carbuncles (化膿).
- *S. cohnii* (科氏葡萄球菌) is found on the **skin of humans** and occasionally in **urinary tract** and **wound infections**.



HABITAT AND DISTRIBUTION

- Human skin is the habitat of both *S. epidermidis* (表皮葡萄球菌) and *S. haemolyticus* (溶血葡萄球菌), and the latter is associated with human infections.
- Grow in the presence of **10% NaCl**.
- Among the staphylococci, *S. aureus* (金黄色葡萄球菌) has been studied most as a cause of foodborne gastroenteritis.



INCIDENCE IN FOODS

- ◆ In general, **staphylococci** may be expected to exist, at least in low numbers, in any or all food products that are of **animal origin** or in those that are **handled directly by humans, unless heat-processing steps are applied** to effect their destruction.



NUTRITIONAL REQUIREMENTS FOR GROWTH

- ◆ **Amino acids** are required as nitrogen sources, and **thiamine and nicotinic acid** are required among the B vitamins.
- ◆ **Arginine** appears to be essential for enterotoxin B production.



TEMPERATURE GROWTH RANGE

- ◆ Although it is a mesophile, some strains of *S. aureus* can grow at a **temperature as low as 6.7°C**.
- ◆ In general, growth occurs over the range **7-47.8°C**, and **enterotoxins are produced between 10°C and 46°C**, with the optimum between **40°C and 45°C**.



EFFECT OF pH, WATER ACTIVITY, AND OTHER PARAMETERS

- ◆ Regarding pH, *S. aureus* can grow over the range **4.0-9.8**, but its optimum is in the range 6-7.
- ◆ With respect to a_w , the staphylococci are unique in being able to grow at values lower than for any other nonhalophilic bacteria. Growth has been demonstrated as low as 0.83 under otherwise ideal conditions, although **0.86 is the generally recognized minimum a_w** .



NaCl and pH

- ◆ Increasing NaCl concentration → raise the minimum pH of growth.
- ◆ At a pH of 7.0 and 37°C, enterotoxin B was inhibited by 6% or more NaCl (see Figure 23-1).



pH, a_w , and Temperature

- ◆ No growth occurred with a combination of pH <5.5, 12°C, and a_w of 0.90 or 0.93
- ◆ No growth occurred at pH <4.9, 12°C and a_w of 0.96.



NaNO₂, Eh, pH, and Temperature of Growth

- ◆ *S. aureus* strain S-6 grew and produced enterotoxin B in cured ham under **anaerobic conditions with a brine content up to 9.2%** but not below a pH of 5.30 and 30°C, or below a pH of 5.58 at 10°C.
- ◆ Under **aerobic conditions, enterotoxin production occurred sooner** than under anaerobic conditions. As the concentration of HNO₂ increased, enterotoxin production decreased.



STAPHYLOCOCCAL ENTEROTOXINS: TYPES AND INCIDENCE

- ◆ SEA, B, C, D, E, G, and H
- ◆ Table 23-3
- ◆ Regarding **the percentage of strains that are enterotoxigenic**, widely different percentages have been found **depending on the source of isolates**.
 - Only 10% of 236 raw milk isolates were isolates were positive. In a study of *S. aureus* from chicken livers, 40% were enterotoxigenic. In another study, 33% of 36 food isolates were enterotoxigenic.



Chemical and Physical Properties

- ◆ Table 23-4
- ◆ In their activate states, the enterotoxins are resistant to proteolytic enzymes such as trypsin (胰蛋白酶), chymotrypsin (胰凝乳蛋白酶), rennin (凝乳酶), and papain (木瓜蛋白酶), but sensitive to pepsin (胃蛋白酶), at a pH of about 2.
- ◆ **Biological activity and serological reactivity are generally associated.**



Chemical and Physical Properties

- ◆ **The enterotoxins are quite heat resistant.** The biological activity of SEB was retained after heating for **16 hours at 60°C and pH 7.3.**
- ◆ Table 23-5, $D_{110^{\circ}\text{C}} = 18 \sim 60$ (min)
- ◆ **Heat resistance: SEC>SEB>SEA**
- ◆ *S. aureus* cells are considerably more sensitive to heat than the enterotoxins (Table 23-6).



Production

- ◆ **Enterotoxin production** tends to be favored by the **optimum growth conditions** of pH, temperature, Eh, and so on.
- ◆ The **optimum temperature** for enterotoxin production is in the **40 - 45°C** range.
- ◆ **Staphylococcal enterotoxins** have been reported to **appear in cultures as early as 4 - 6 hours**.



Mode of Action

- ◆ **Staphylococcal enterotoxins**, along with the **toxic shock syndrome toxin (TSST)** are **bacterial superantigens**.
- ◆ Superantigens are proteins that **activate many different T cell clones**.
 - result in excessive activation of the immune system.
- ◆ **SEs stimulate helper T cells to produce cytokines** such as the interleukins (IL), gamma-interferon, and tumor necrosis factor.
- ◆ Among the cytokines, an **overabundance of IL-2 is produced** and it appears to be responsible for many or most of the symptoms of staphylococcal gastroenteritis.

腹部絞痛



SYNDROME

- ◆ The symptoms of staphylococcal food poisoning usually **develop within 4 hours** of the ingestion of contaminated food.
- ◆ The **symptoms**—nausea, vomiting, abdominal cramps (腹部絞痛)(which are usually quite severe), diarrhea, sweating, headache, prostration (疲勞,虛脫), and sometimes a fall in body temperature — **generally last from 24 to 48 hours** and the **mortality rate is very low or nil**. The usual treatment for healthy persons consists of **bed rest and maintenance of fluid balance**.
- ◆ Upon cessation of symptoms, the **victim possesses no demonstrable immunity** to recurring attacks, although animals become resistant to enterotoxin after repeated oral doses.

THE GASTROENTERITIS SYNDROME



- ◆ Proof of staphylococcal food poisoning is established by **recovering enterotoxigenic staphylococci** from **leftover food** and from the **stool cultures of victims**.
- ◆ Because the symptoms are referable to the **ingestion of preformed enterotoxin**, it is conceivable that stool culture might be negative for the organisms, although this is rare.
- ◆ The minimum quantity of enterotoxin needed to cause illness in humans is about **20 ng** (毫微克).
- ◆ Regarding the pathogenesis of enterotoxin in humans, **most of the symptoms caused by IL-2, including vomiting and diarrhea**.



INCIDENCE AND VEHICLE FOODS

- ◆ Vehicle foods for staphylococcal enteritis usually are **products made by hand and improperly refrigerated after being prepared.**
- ◆ The small outbreaks that occur in homes are not reported to public health officials.
- ◆ A large percentage of the reported cases of all types are those that result from banquets, generally involving large numbers of persons.



ECOLOGY OF *S. AUREUS* GROWTH

- ◆ **Staphylococci do not compete well with the normal flora** of most foods, → especially true for those that contain large numbers **of lactic acid bacteria** → in both **fresh and frozen foods.**
- ◆ Bacteria known to be antagonistic to *S. aureus* growth include *Acinetobacter* (不動桿菌屬), *Aeromonas* (產氣單胞菌屬), *Bacillus* (芽孢桿菌屬), *Pseudomonas* (假單胞菌屬), *S. epidermidis* (表皮葡萄球菌), the Enterobacteriaceae, the Lactobacillaceae, enterococci (腸球菌), and others.



PREVENTION OF STAPHYLOCOCCAL AND OTHER FOOD-POISONING SYNDROMES

- ◆ When susceptible foods are produced with **low numbers of staphylococci**, they will **remain free of enterotoxins** and other food-poisoning hazards **if kept either *below 40°F (4.4°C) or above 140°F (60°C)*** until consumed.



PREVENTION OF STAPHYLOCOCCAL AND OTHER FOOD-POISONING SYNDROMES

- ◆ The 5 factor most frequently involved in foodborne-disease outbreaks for the years 1961 through 1972
 - 1. inadequate refrigeration**
 - 2. preparing foods far in advance of planned service**
 - 3. infected persons' practicing poor personal hygiene**
 - 4. inadequate cooking or heat processing**
 - 5. holding food in warming devices at bacterial growth temperatures**



PREVENTION OF STAPHYLOCOCCAL AND OTHER FOOD-POISONING SYNDROMES

- ◆ **Inadequate refrigeration** alone comprised **25.5%** of the contributing factors. The five listed contributed to **68%** of outbreaks.
- ◆ **Table 23-9**
- ◆ Susceptible foods should not be held within the staphylococcal growth range **for more than 3 – 4 hours.**