Characteristics and sources of *Staphylococcus aureus* and staphylococcal enterotoxins

Main microbiological characteristics

Human foodborne illness is an intoxication caused by the ingestion of staphylococcal enterotoxins (SEs), heat-resistant proteins preformed in food in which SE-producing *S. aureus* (or any other staphylococcus) has been able to develop and produce its toxins.

*S. aureus* is a Gram-positive coccal bacterium, measuring from 0.5 to 1 μm in diameter. It is non-spore-forming, non-motile, facultatively anaerobic and catalase- and coagulase-positive. *S. aureus*, the type species of the genus *Staphylococcus*, also commonly called "golden staph", produces numerous toxins including SEs, which are produced by certain *S. aureus* (those carrying the genes for these toxins) and are responsible for outbreaks associated with this bacterium. To date, 21 different serotypes (SEA to SEE, SEG to SEV) have been described. Only six of them have been clearly demonstrated to have been involved in cases of intoxication: SEA (the most frequently detected serotype in intoxication) to SEE, and SEH. However, since the emetic nature of the SEG, SEI, SER, SES and SET toxins has also been demonstrated, they should be taken into account when characterising toxic episodes. Other species of coagulase-producing staphylococci can also produce SEs, specifically *S. intermedius*, but their role in intoxication cases to date remains poorly documented. Moreover, *S. aureus* is the species of coagulase-producing staphylococcus found most frequently in foods.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Growth</th>
<th>Toxins (SEs)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Optimum</td>
<td>Extremes*</td>
</tr>
<tr>
<td>Temperature (°C)</td>
<td>35-41</td>
<td>6-48</td>
</tr>
<tr>
<td>pH</td>
<td>6-7</td>
<td>4-10</td>
</tr>
<tr>
<td>aw</td>
<td>0.99</td>
<td>0.83-0.99</td>
</tr>
<tr>
<td>NaCl (%)</td>
<td>0-4</td>
<td>0-20</td>
</tr>
<tr>
<td>Atmosphere</td>
<td>aerobic</td>
<td>aero-anaerobic</td>
</tr>
</tbody>
</table>

* Extreme values given in the table have only been observed for some strains.

Hazard sources

SEs are proteins produced by coagulase-producing staphylococci, mainly *S. aureus*. These toxins, if present in food in sufficient quantities, can trigger the symptoms of intoxication (Table 2). Staphylococci are ubiquitous bacteria found on the skin, mucous membranes and nasopharyngeal area of warm-blooded animals (mammals, birds), and particularly humans. Coagulase-producing staphylococci are responsible for clinical and subclinical mastitis in cows and other ruminants.

These bacteria can also be isolated from the natural environment (soil, fresh and sea water, dust, air), the domestic environment of humans (kitchens, fridges), the hospital environment and food preparation facilities, as well as from food. As human and animal skin and mucous membranes can harbour *S. aureus*, its presence in the environment is likely due to contamination by humans or animals.
Transmission routes
Apart from cases contracted in the laboratory, all the cases of SE intoxication described to date have been foodborne. Contamination of food is mostly of human origin. It may occur either by direct or indirect contact (contaminated squamae, droplets from the respiratory tract containing the micro-organism).

Contamination of food can also be of animal origin, the most common being the contamination of milk from cows with mastitis.

S. aureus can also cause infections which are sometimes fatal (paronychia, etc.).

Recommendations for primary production
• Preventive measures in primary production should include the monitoring of animal health (e.g. mastitis), good handling practices, cleaning and disinfecting of equipment and premises, as well as strict hand washing and the wearing of a cap.

Human foodborne illness
Nature of the disease
The characteristics of the disease are presented in Table 2.

The duration of the incubation period and the severity of the symptoms depend on the amount of enterotoxins ingested and the susceptibility of each individual.

Susceptible population groups(1): no particularly susceptible population.

Dose-effect relationships(2)
Although several studies have attempted to assess the minimum dose to be ingested that triggers the first symptoms, this remains unclear. Epidemiological studies conducted following two incidents involving SEA and affecting a large number of patients reported that the ingestion of doses of between 20 and 144 ng triggered the symptoms in the exposed population.

Epidemiology
Cases of staphylococcal intoxication are monitored through the mandatory reporting of food poisoning outbreaks (FPOs). Note that many outbreaks, particularly those occurring within families, are certainly unreported or undiagnosed.

Role of foods
Main foods to consider
Four conditions are required for food to be contaminated by S. aureus and its enterotoxins:
• a source of enterotoxin-producing staphylococci (raw food materials, healthy or sick carrier, etc.);
• a means of transmission to the food (contaminated cutting tool, healthy or sick carrier, etc.);
• a food whose physico-chemical characteristics favour the growth of S. aureus and toxinogenesis (see the foods most “at risk”, below);
• a favourable temperature for the period required for significant bacterial multiplication and toxinogenesis.

(1) Susceptible population group: people with a higher than average probability of developing symptoms of the disease, or severe forms of the disease, after exposure to a foodborne hazard [definition used in ANSES data sheets].
(2) Relationship between the dose (the quantity of microbial cells ingested during a meal) and the effect on an individual.

Table 2. Characteristics of intoxication due to the ingestion of staphylococcal enterotoxins

<table>
<thead>
<tr>
<th>Mean incubation period</th>
<th>Target population</th>
<th>Main symptoms</th>
<th>Duration of symptoms</th>
<th>Duration of the contagious period</th>
<th>Complications</th>
<th>Carriage of SE-producing staphylococci</th>
</tr>
</thead>
<tbody>
<tr>
<td>30 min - 1 h (on average)</td>
<td>Entire population, all age groups combined</td>
<td>Nausea followed by characteristic uncontrollable vomiting (projectile vomiting)</td>
<td>18 - 24 h</td>
<td>Enterotoxins non-communicable from person to person. No contagiosity.</td>
<td>Mortality remains exceptional (mortality rate: 0.02%), affecting those most sensitive to dehydration (infants and the elderly) and people with underlying disease. Estimated hospitalisation rate: 16% in cases where the agent was confirmed.</td>
<td>Intestinal carriage: 20-30% of adults  Nasal carriage: 20-55% (37% on average) of adults  Manual carriage: 10% of hospital staff or staff of a restaurant chain</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Abdominal pain</td>
<td>Diarrhoea</td>
<td>Diarrhoea and general weakness may last a further 24 hours.</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>Dizziness</td>
<td>Chills</td>
<td>General weakness sometimes accompanied by mild fever</td>
<td></td>
<td></td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>In severe cases, headache, prostration and hypotension have been reported.</td>
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</tbody>
</table>
S. aureus can be isolated from a wide variety of foods. The foods most “at risk” are:

- foods recontaminated following heat treatment or any other process for eliminating commonplace microflora. The more the food is handled, the greater the risk is high. These include foods such as poultry meat, cooked sliced ham, mixed salads (including rice or vegetable salads), cream cakes and cooked meals handled after cooking;

- foods fermented by slow acidification allowing the growth of S. aureus during fermentation. For example, some cheeses and some fermented cured meats such as salami. The risk is increased if the fermentations used to initiate production are insufficient, or if none have been added;

- products that are dried or have reduced water content, in which the growth of S. aureus may have been facilitated at one of the manufacturing or storage steps due to a reduced aw and a favourable temperature. These foods include powdered milk, pasta, dried fish.

Dishes requiring human handling (mixed salads, cooked dishes) and dairy products were the foods most frequently associated with staphylococcal intoxications reported in Europe and France in 2009.

**Inactivation treatments in industrial environments**

<table>
<thead>
<tr>
<th>Table 3. Inactivation treatments of S. aureus in foods</th>
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</thead>
<tbody>
<tr>
<td><strong>Disinfectants</strong></td>
</tr>
<tr>
<td>S. aureus is sensitive to all disinfectants authorised in the food-processing sector, on condition that the recommended procedures for use are followed.</td>
</tr>
<tr>
<td>Irradiation</td>
</tr>
<tr>
<td>D10 = value** = D*</td>
</tr>
<tr>
<td>D10 = value** = 0.45 kGy</td>
</tr>
</tbody>
</table>

* D is the time needed to divide by 10 the initial population of a microbiological hazard.
** D10 = value is the dose of radiation required to reduce the viability of a population by 90% (1 log10 cycle) under the stated conditions.

Unlike the bacterium, SEs are stable under the heat treatment conditions usually applied to foods. Depending on the amount of enterotoxins initially present, sterilisation (of milk for example, by UHT treatment) may cause a partial denaturation of the SEs, leaving enough in the food to cause an intoxication. In addition, SEs are resistant to most treatments applied in the food industry. It must therefore be assumed that once formed in the food, the enterotoxins cannot be sufficiently inactivated to eliminate the risk of intoxication.

**Monitoring in food**

Regulation (EC) no. 2073/2005, as amended, on microbiological criteria for foodstuffs, sets:

- a process hygiene criterion for coagulase-producing staphylococci (CPS) in milk and milk products and in shelled and shucked products of cooked crustaceans and molluscan shellfish;

- a safety criterion for staphylococcal enterotoxins in milk and milk products in the event of failure to respect the hygiene criterion.

CPS are counted according to the French NF EN ISO 6888 standard - parts 1, 2 and 3. The first two parts are conventional techniques for counting colonies on selective agar medium after direct inoculation of decimal dilutions of the food and incubation at 37°C. The third part describes a screening and counting method using the most probable number technique after an enrichment step. Staphylococcal enterotoxins types SEA to SEE are detected according to the method of the European Union Reference Laboratory for CPS including S. aureus, using qualitative immunochromatographic tools. To date, commercially available kits are only able to detect toxins SEA to SEE.

**Recommendations to operators**

- Equipment and premises must be scrupulously cleaned and disinfected, given the strong adherence of staphylococci to surfaces.

- To account for the fact that a great many operators are healthy carriers, the cleaning and disinfecting of hands and the wearing of a cap which covers the hair entirely are good hygiene practices (GHPs) that must be followed.

- In addition, food handlers with skin lesions should be excluded from the handling of unpackaged food, unless the lesions are properly covered (by wearing gloves). Similarly, if any nasopharyngeal symptoms are observed, masks must be worn. Given the high proportion of healthy carriers, and the fact that carriage is not constant in most individuals, screening for S. aureus during medical visits is needless. Contamination is prevented through rigorous application of the GHPs mentioned above.

- As these measures are not sufficient to achieve a zero rate of infection, the staphylococci must be destroyed by appropriate treatment (thermal or other), before they can multiply, or their multiplication must be prevented by maintaining food below 6°C. Respecting the cold chain is therefore vital with regard to staphylococci.

- Any technological food processes applied in a hazardous temperature zone (from +10°C to +45°C) must be of short duration or based on parameters other than temperature to stop the growth of the bacteria, such as a pH below 5.

As staphylococci are heat sensitive whereas their enterotoxins are heat stable, decontamination of a product heavily contaminated with S. aureus cannot be guaranteed by a heat treatment. This will destroy the bacteria but not their enterotoxins, if present. Once formed in the food, the enterotoxins cannot be effectively eliminated.

**Domestic hygiene**

**Recommendations for consumers**

- Good hygiene practices should be followed when handling and preparing food (hand washing, cleaning equipment, etc.).

- Before preparing meals, disinfect and protect any wounds or pus-filled pimples with a waterproof dressing, or use gloves.

- It is essential to respect the cold chain and to refrigerate cooked food as soon as possible, if not consumed immediately.

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(3) S. aureus is the species of coagulase-producing staphylococcus found most frequently in foods.
(4) Microbiology of food and animal feeding stuffs - Horizontal method for the enumeration of coagulase-positive staphylococci (Staphylococcus aureus and other species).
References and links

**General references**

- Rizzi V. (2011). Update on EFSA activities and *S. aureus* reporting in animals and food. Workshop of the NRLs for Coagulase Positive Staphylococci - Maisons-Alfort, France.
- SCVPH (2003). Opinion of the scientific committee on veterinary measures relating to public health on staphylococcal enterotoxins in milk products, particularly cheeses. SCVPH plenary meeting, March 27-28.

**Useful links**

- ANSES: [www.anses.fr](http://www.anses.fr)
- EFSA: [www.efsa.europa.eu](http://www.efsa.europa.eu)
- European Union Reference Laboratory ([http://www.ansespro.fr/eurl-staphylococci/](http://www.ansespro.fr/eurl-staphylococci/)) and National Reference Laboratory (NRL) for coagulase-positive staphylococci including *Staphylococcus aureus* and staphylococcal enterotoxins: ANSES Maisons-Alfort Laboratory for Food Safety.
- National Centre of Reference (NCR) for staphylococci: Hospital Group of East, Centre of Microbiology and Pathology, Institute of Microbiology, Claude Bernard University, Lyon-I (Lyon).