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Rapid communications

BOTULISM AND HOT-SMOKED WHITEFISH: A FAMILY CLUSTER OF TYPE E BOTULISM IN FRANCE, SEPTEMBER 2009

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A family cluster of three cases of type E botulism were identified in south-east France in September 2009. The suspected food source of infection was a vacuum packed hot-smoked whitefish of Canadian origin purchased by the family during a visit to Finland and consumed several weeks later in France on the day prior to symptom onset. No leftover fish was available to confirm this hypothesis. Vacuum packed hot-smoked whitefish has previously been associated with cases of type E botulism in multiple countries, including Finland, Germany, the United States and Israel.

Case notification

A confirmed case of type E botulism in an individual residing in south-east France was reported to the French National Institute for Public Health Surveillance (Institut de Veille Sanitaire) by the National Reference Center (NRC) for Anaerobic Bacteria and Botulism at the Pasteur Institute in Paris on 10 September 2009. Two other members of the same family were reported as having clinical symptoms compatible with botulism. An investigation was undertaken to identify additional cases, the vehicle of transmission, and to put in place appropriate control measures.

Methods

Following notification of the cases, active case finding was carried out via contact with local health authorities, the NRC and the hospital services where cases were hospitalised. Hospital clinicians treating the patients, and thus likely to see other such cases, were reminded by telephone contact to immediately report all clinical suspicions of botulism to the local health authorities using the routine mandatory notification system for the disease.

Serum samples from the cases were analysed by the NRC. The presence of botulinum neurotoxin was confirmed by intraperitoneal administration of patient serum to mice, and the toxin type was ascertained by neutralisation with specific antibodies [1].

The food history of the cases in the three to four days before onset of symptoms was documented by the local health authorities, as were the details of purchase, transport and consumption of the suspected food product.

Based on patients' food history a fish product purchased during a family visit to Finland was suspected to have been the source of infection. A sample originating from the same batch of raw fish as the implicated product but processed one day later was collected from a local supermarket in Finland. The sample was sent to the Department of Food and Environmental Hygiene, University of Helsinki, Finland, for analysis of *Clostridium botulinum* by multiplex PCR targeted to the types A, B, E, and F neurotoxin genes [2]. Twelve 1gram samples from skin, gills or peritoneum were each inoculated into 10 ml of anaerobic tryptose-peptone-glucose-yeast extract (TPGY) medium and incubated at 30°C for three days. One ml of each culture was transferred to fresh TPGY medium and incubated overnight at 30°C. Lysed cells from 1 ml of each culture were used as template in PCR. PCR amplification products were visualised in 2% agarose gels against standard molecular weight markers.

Results

The three cases (two adults aged 52 and 46 years and one adolescent child aged 13 years) presented with classical clinical symptoms of botulism (gastrointestinal symptoms followed by descending paralysis) on 7 September 2009 and were hospitalised the following day. One of the adult cases rapidly developed quadriplegia and required intubation and mechanical ventilation for 17 days. The other two patients presented with a milder form of the disease, did not develop paralysis of limbs or respiratory muscles and were released from hospital in mid-September. The severe case remained hospitalised as of 29 September (latest information available) but had regained motor function and begun to walk.

The NRC confirmed a diagnosis of type E botulism for the severe case. Botulinum toxin type E was identified in a serum sample (8 Mouse Lethal Dose/ml) and in two from three gastric juice samples (<20 MLD/ml). Serum samples from the two milder cases were negative for botulinum toxin. A faecal sample obtained from the child was negative for botulinum toxin and *C. botulinum*. No other botulism case associated with this episode was identified.

The food investigation carried out with the family identified the consumption of vacuum packed hot-smoked whitefish (*Coregonus lavaretus*) on 6 September 2009 (the day prior to symptom onset). All three sick members of the family reported having eaten the smoked fish and a fourth non-sick family member did not consume the product. There was no leftover fish to test for the presence of toxin. The family did not report consumption of any other foods usually associated with the risk for botulism (home-canned vegetables or home-prepared meat products such as ham, sausages and pâté) in the days preceding symptom onset.

The whitefish was purchased by the family in a supermarket in a village in east Finland on 22 August 2009. The fish was smoked in Finland but was originally from Canada. It was refrigerated after purchase. The family returned to France the following day. The fish was placed in a cooling bag with ice-packs for the duration of the 14-hour journey and then refrigerated upon arrival in the family home until the day of consumption on 6 September 2009, two days before the expiry date.

The fish was not heated prior to consumption. The entire product (800-1000 g) was eaten at the meal by the three patients. The adult with a severe form of the disease reportedly consumed a greater portion of the fish than the two milder cases.

An environmental investigation was carried out in the premises of the fishery production plant by the food control authority in Finland. The inspection focussed on the fish processing and storage temperatures, hygiene conditions and efficacy of in-house control of the producer. The storage temperature of the raw material, temperatures during the process and transport were found to be correct and in accordance with the in-house control plan and legislation. The raw fish was imported from Canada two months earlier and stored frozen at the premises' freezer (-18°C). The processing of the batch was started on 16 August 2009 with thawing and salting of the fish (temperature below 3°C). After hot smoking (two hours; maximum temperature 68°C) the fish was rapidly chilled (until 0.5°C), vacuum packed and stored below 3°C. The batch (about 600 kg) was transported at 0°C to the retail on 18 August 2009. The fish sample representing the same batch of raw material but processed one day later than the implicated fish product was negative for *C. botulinum* in the PCR analysis. Temperature controls carried out at the supermarket of purchase by the local food control authority showed storage temperatures for fishery products of 0.8-2.8°C.

Public health measures

European countries were informed of the event via the 'Early Warning and Response System' (EWRS) and an alert in the 'Rapid Alert System for Food and Feed' (RASFF), both issued on 11 September 2009. The information in the RASFF was subsequently transmitted to the Canadian food safety authorities. No other cases of botulism associated with this product were identified in Finland, France or other European Member States, as of 9 November 2009.

Discussion and conclusion

C. botulinum type E is an aquatic bacterium endemic in areas such as Canada and Alaska [3-5]. Type E botulism is characteristically associated with the consumption of improperly prepared foods of aquatic origin, either fresh water or marine [6]. Cases of type E botulism are very rare in France with the last episode declared in 2003 [7]. Foods associated with the occurrence of this form of botulism in France include salted herring, grey mullet, canned carp and canned sardines [8].

The negative mouse bioassay results of the serum samples of the two patients with a milder form of the disease could be explained by a lack of circulating toxin in the patients' blood. It is known that botulinum toxin cannot be detected in serum once it becomes irreversibly bound to its cell receptors and thus the detection of toxin in serum samples is believed to depend on the timeliness of sample collection and on the ingested dose of toxin, among other factors [6,9].

The epidemiological investigations support the hypothesis of the vacuum packed hot-smoked whitefish as the source of contamination of the three cases. No leftover fish was available for testing to confirm this hypothesis. An association between hot-smoked whitefish and type E botulism has been previously documented in Finland, Germany, the United States and Israel [10-13]. On two previous occasions, cases of type E botulism have been associated with whitefish imported from Canada and processed in Finland, as was the situation with the whitefish consumed by the three French cases [10,11].

Vacuum packed hot-smoked fish is a known risk food for type E botulism [14]. It is believed that the hot-smoking processes carried out on this type of fish, which typically reach temperatures of 60-80°C, are often insufficient to eliminate *C. botulinum* spores [15]. Among factors believed necessary for controlling growth and toxin production in this fish is the continuous storage of the fishery products below 3°C [10,11], information which is clearly labelled on this food product. According to the national legislation, modified atmosphere package (MAP) and vacuum packed fishery products must be stored below 3°C in production and at retail in Finland. Temperature controls carried out at the fishery production plant and the supermarket of purchase showed that storage temperatures were in accordance with the legislation. It is probable that the whitefish consumed by the three French cases was not stored below 3°C for the duration of the 14-hour return journey to France. Also, French domestic fridges are estimated to have an average temperature of 6.6°C [16] and thus well above 3°C. Assuming that the temperature of the family's fridge corresponds approximately to the estimated national average (the actual fridge temperature was not measured) the two weeks of refrigerated storage could have allowed ample time for

growth and toxin production in the anaerobic environment created by vacuum packaging.

The absence of additional cases in Finland could be explained by a limited contamination of the whitefish by *C. botulinum*. The fish sample representing the implicated batch of raw material was negative for *C. botulinum* spores. In a previous case of human infection reported in Finland, 10 fish samples from an implicated batch were also negative for *C. botulinum* [11]. This is consistent with a previous prevalence study showing that 18% of raw and 5% of processed and packaged whitefish carry type E spores [14]. The absence of further cases may also be explained by a difference in storage habits of hot-smoked whitefish between the Finnish population and foreign tourists.

This family cluster provides further evidence of the risk of type E botulism associated with consumption of vacuum-packed hot-smoked whitefish. This episode also highlights the potential public health threat of *C. botulinum* spores in incorrectly stored processed food products and underlines the importance of clear labelling of storage conditions for products purchased in the refrigerated sections of supermarkets.

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