Two severe familial cases of botulism were reported to the health authorities in Brittany, north-west France, on 11 August 2008. An investigation was undertaken to identify additional cases, the vehicle of transmission, and to put in place adapted control measures.

Methods
Following notification of the cases, health authorities issued a communication to French hospitals, anti-poison and toxin centres and general practitioners to alert health professionals. No specific case definition was used for the investigation; the health professionals were requested 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 and samples recovered from the food investigation were analysed by the National reference laboratory (NRL) for anaerobic bacteria and botulism at the Pasteur Institute, Paris. The presence of botulinum toxin was confirmed by intraperitoneal administration of patient serum to mice, and the toxin type was ascertained by the specific neutralisation technique.

The food history of the cases in the three to four days before onset of symptoms was documented.

Results
The two cases, a mother (in her 60s) and daughter (in her 20s), presented with gastrointestinal symptoms accompanied by dysphagia, blurred vision and facial paralysis on 9 August 2008. Both patients were hospitalised the day of symptom onset with a rapid evolution towards generalised and complete paralysis. The two women required intubation and mechanical ventilation. They remain in this condition in intensive care as of 3 September, with minor early signs of improvement. A trivalent antitoxin (toxin types A, B, E) was administered to the patients on 13 August. This antitoxin was imported from a commercial laboratory in Germany as botulism antitoxins are not commercially available in France. An authorisation for temporary usage of the product was issued by the French Health Products Safety Agency (Afssaps).

The diagnosis of botulism (toxin type A) was confirmed for both cases by the NRL, by detection of botulinum toxin in blood samples of the patients. No other botulism cases associated with this episode were identified.

The investigation of the food history for both women revealed that they had consumed an industrially produced pre-cooked Mexican-style “Tex-Mex” dish, chicken enchiladas, the day before onset of symptoms. These chicken enchiladas are sold as a pre-prepared kit consisting of several sachets containing a cheddar cheese sauce, a pre-cooked chicken and vegetable mix and two wheat tortillas. The product is consumed after reheating in a microwave oven. Microbiologic testing of the remaining chicken and vegetable mix revealed the presence of Clostridium botulinum and a high level botulinum toxin type A contamination (2.8x10^5 mouse lethal doses/g). The remaining cheese sauce was negative for botulinum toxin.

The epidemiological investigation of the two cases suggested that the contaminated enchiladas had been mistakenly stored at room temperature for two weeks between purchase and consumption, contrary to the producer’s recommendation of refrigerated storage. They were consumed one day after the use-by date. However, the recommended storage conditions on the packaging are not easily visible to the consumer.

Risk analysis
The chicken enchiladas had been produced in France. The incriminated batch of enchiladas had a ‘use-by’ date of 7 August 2008. This batch was distributed only in France. Other batches of the enchiladas as well as pre-cooked chicken fajitas are also distributed in Belgium, Switzerland and Spain.

Stored production samples from the contaminated batch of enchiladas as well as other batches of enchiladas and fajitas and other products produced by the company around the same time were analysed and tested negative for botulinum toxin and C. botulinum.

A risk-analysis carried out on 14 August at the production plant concluded that the plant conforms to hygiene and safety regulations. An investigation of the fabrication protocols showed that the fabrication process includes a pasteurisation step of heating the product to 85°C for two hours.

Public health measures
The company issued a recall of the implicated batch of enchiladas on August 12. As a precautionary measure, this recall was then widened to include all enchiladas and fajitas produced by the firm. The population was informed of this outbreak through national
inter-ministerial press releases and posters placed in supermarket chains. European countries were informed via the ‘Early Warning and Response System’ and an alert in the ‘Rapid Alert System for Food and Feed’.

In light of the potential role of incorrect product conservation in facilitating the multiplication of \textit{C. botulinum} and toxin production in the contaminated enchiladas, a generalised reminder about respecting the storage conditions of such products was communicated by the French authorities. The producer of the enchiladas agreed to change the packaging of this and similar products to make the recommended storage conditions more visible for the consumer.

**Discussion and conclusion**

The two cases represent the clinically most severe cases of botulism reported in France in recent years. Botulism has been mandatorily notifiable in France since 1986, and 96 cases were reported between 2003 and 2006 [1]. Only two cases of botulism due to toxin type A, associated with the more severe form of the disease, were notified during this period, compared to 51 cases of toxin type B (53%) and four of toxin type E (4%) [1,2]. One-third of the cases notified during this period were not confirmed [1].

The epidemiological and environmental investigations support the hypothesis that the two cases ingested the toxin following incorrect storage of the chicken enchiladas which contained a strain of \textit{C. botulinum} after production. Prolonged storage at room temperature could explain the unusually high level of toxin in the chicken and vegetable mix.

Intoxications with \textit{C. botulinum} producing toxin type A are often associated with vegetable-based products that at some point contained soil with \textit{C. botulinum} spores [2,3].

The thermo-resistance of \textit{C. botulinum} spores varies by strain and according to factors such as the lipid and protein content of the food matrix [2]. Exposure to a temperature of 110-120°C for between 0.4 to 6 minutes is necessary to inactivate 90% of a population of \textit{C. botulinum} A spores [2]. It is thus probable that the pasteurisation step during the enchiladas’ fabrication process does not prevent the survival of spores present in primary ingredients or potentially introduced during the fabrication process. Thus, correct refrigerated storage of such processed food products is essential to avoid germination of the spores and toxin production.

Certain ingredients used in the production of “Tex-Mex” food products, including industrially produced cheddar-cheese sauce and home-canned jalapeno peppers, have previously been implicated in outbreaks of botulism in the United States [4,5].

This family cluster highlights the potential public health threat of \textit{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. In addition, the episode, widely reported in the national media, has served to remind the general population in France that compliance with food storage recommendations is a prerequisite for food safety.