

# Wound botulism after traumatic open fracture in Italy

## Botulismo da ferita a seguito di frattura traumatica esposta in Italia

Francesco Giuseppe De Rosa<sup>1</sup>, Maria Laura Stella<sup>1</sup>, Sara Astegiano<sup>2</sup>, Silvia Corcione<sup>1</sup>, Ilaria Motta<sup>1</sup>, Lucia Decastelli<sup>2</sup>, Giovanni Di Perri<sup>1</sup>

<sup>1</sup>Department of Medical Sciences, University of Turin; Infectious Diseases, Amedeo di Savoia Hospital, Turin, Italy;

<sup>2</sup>Veterinary Medical Research Institute for Piemonte, Liguria and the Valle D'Aosta, Italy

On 20<sup>th</sup> October 2012, a 41 year old woman fell down from a height of two meters while free climbing and reported open fractures at the right leg. At the arrival in the Emergency Department, by helicopter transfer, there was a exposed subastragalic fracture (grade III according to Tschernie classification), with soft tissues trauma and soil contamination (Table 1). The wound was cleaned with repeated washings with saline solution and then disinfected by means of povidone iodine and hydrogen peroxide. The patient underwent prompt surgical reduction and external fixation of the fracture. The patient was empirically treated with amoxicillin clavulanate which was changed after six days to intravenous ciprofloxacin and ertapenem after the isolation of *Enterobacter asburiae* and *Morganella morganii* from wound swabs. Nine days after the first surgery, a new debridement was performed and foul-smelling discharge from the wound was reported.

On November 6<sup>th</sup>, the patient complained about blurred vision, which persisted notwithstanding a negative ophthalmologic consult. On November 13<sup>th</sup> the patient was discharged from the Trauma Center but three days later she was admitted to a Neurology ward with diplopia, ptosis, and mild dysphagia. On examination, bilateral ptosis and complete ophthalmoplegia were noticed; she also had slurred speech and difficulty in swallow-

**Table 1** - The Oestern and Tschernie classification for open fractures uses wound size, level of contamination, and fracture pattern to grade open fracture.

Grade I	<ul style="list-style-type: none"> <li>• Open fractures with a small puncture wound without skin contusion</li> <li>• Negligible bacterial contamination</li> <li>• Low-energy fracture pattern</li> </ul>
Grade II	<ul style="list-style-type: none"> <li>• Open injuries with small skin and soft tissue contusions</li> <li>• Moderate contamination</li> <li>• Variable fracture patterns</li> </ul>
Grade III	<ul style="list-style-type: none"> <li>• Open fractures with heavy contamination</li> <li>• Extensive soft tissue damage</li> <li>• Often, associated arterial or neural injuries</li> </ul>
Grade IV	<ul style="list-style-type: none"> <li>• Open fractures with complete or incomplete amputations</li> </ul>

ing solid food. Deep tendon reflexes were normal. A brain CT and a lumbar puncture were normal. A wound botulism was suspected and multiple samples from blood, wound and rectum were taken.

The specimens were sent to the Regional Veterinary Medical Research Institute, where the samples were tested for *Clostridium botulinum* by microbiological method, by Real-time PCR assay for genes codifying toxins and by the mouse bioassay, which is the gold standard laboratory test for confirmation of botulism. Briefly, six pairs of mice were injected intraperitoneally with patient serum mixed with antitoxin ABE (pair 1), antitoxin CD (pair 2), or without antitoxin (pair 3). The mice were observed for up to 72 hours for signs of botulism and/or death. If no pairs had signs of diseases or died, the test is negative for

### Corresponding author

Francesco Giuseppe De Rosa

E-mail: francescogiuseppe.derosa@unito.it

*C. botulinum* toxin; otherwise if one pair of mice is protected with antitoxins the test is considered positive.

In this case *C. botulinum* was absent in rectal and wound swabs and in serum; the real-time PCR assay was negative and the serum sample tested on the mouse bioassay resulted positive to toxin ABE, consistent with the diagnosis of wound botulism.

No antitoxin was administered to our patient, since the signs and symptoms were mild and there was a history of allergy. Interestingly, there was no further worsening of symptoms and the patient didn't need intensive support. Antimicrobial therapy was continued for 24 days with meropenem and the patient was discharged at home.

## ■ DISCUSSION

*C. botulinum* is a spore-forming, strictly anaerobic organism that is naturally present in soil, dust, watery sediments, as well as human and animal intestines. There are three major forms of botulism: food-borne, caused by the ingestion of the preformed toxin in food (toxins type involved are A, B, E and rarely F); intestinal, typically occurring in infants after the local production of toxins by intestinal spores; and wound botulism [1]. The species can be divided into four groups: group I, proteolytic in culture and producing toxin A, B, and F; group II, non proteolytic and producing toxin types B, E, and F; group III, that produces toxin types C and D, and group IV that produces type G. Each strain of *C. botulinum* typically produces one single neurotoxin which acts at the neuromuscular junction by blocking the release of acetylcholine, resulting in a symmetrical descending flaccid palsy with slow recovery, since the sprout of new receptors from the nerve endplate is needed. *C. botulinum* producing toxins A, B and C is found in soil, while type E is frequent in aquatic sediment. To our knowledge, only type A and B have been reported in wound botulism. In our case the mouse bioassay model resulted positive for toxins A, B, or E, as only mice treated with polyvalent antitoxin ABE were alive after 72h. The available serum was not furthermore tested since toxins may be transiently present or at low levels [2].

Reports of wound infections by *C. botulinum* are rare: the first cases were described in 1951 in USA [3]. In the 90s, in California, USA, an epidemic of wound botulism was registered among black tar heroin users; a few years later a small epidemic was reported in some European countries [4,5]. A number of cases reported in literature occurred in young men and were related to trauma, often with bone fractures associated with deep wounds and avascular areas [6, 7].

After the contamination, the incubation time appears to be longer in wound botulism than in the food-borne infection, attributed to the multiplication of clostridium within the wound: signs and symptoms appear when enough toxin is produced. The inoculum size does not influence the type or onset of clinical manifestations [8]. Symptoms are consistent with descending symmetrical flaccid paralysis: early manifestations are due to the cranial nerves' palsies and include blurred vision, diplopia, ptosis, dysphagia, dysphonia and dysarthria, later accompanied by autonomic signs such as dry mouth or urinary dysfunction. Deep tendon reflexes may be decreased and fever or sensory defects are only present with concomitant soft tissues infection [6].

To our knowledge, this is the first report of wound botulism in Italy. There are two main considerations arising from our report: the first is that *C. botulinum* resisted to the initial wound disinfection and the second related to the possible pathogenesis of such an infection.

Regarding the first consideration it is known that the spores are more resistant to disinfection than bacteria: only strong disinfectants such as sodium hypochlorite, hydrogen peroxide, ozone and hydrogen peroxide vapor are effective against spores [9-11]. In our case hydrogen peroxide, effective on *C. botulinum* due to its lack of catalase enzymes, was correctly used on the wound but probably the disinfection of contaminated tissues was incomplete, possibly due to the fact that its sporicidal activity at room temperatures is weak [12].

Regarding the second consideration, our patient recalled that the landing of the helicopter was hampered by windy conditions while the wound was uncovered and exposed to high quantity of dust polluted during the attempts of landing. Since the spores of *C. botulinum* are transported by air quickly, contaminating surfaces and even

foods, we may therefore hypothesize that the wound could be contaminated after the trauma, perhaps during the repeated attempts of landing of the helicopter [1].  
If this pathogenesis is correct, we should remem-

ber ourselves that prevention is always better than treatment.

*Keywords:* wound botulism, *Clostridium botulinum*, trauma, clostridium toxin ABE, external fixation.

## SUMMARY

Seventeen days after a traumatic open fracture, a *Clostridium botulinum* wound infection was diagnosed, with self-limiting symptoms. This is the first report

of wound botulism in Italy and the authors discuss the possible role of aerosolized contamination of the wound prior to hospital admission.

## RIASSUNTO

Riportiamo il primo caso di infezione da *Clostridium botulinum* in Italia in una paziente con frattura esposta traumatica.

Si è indagato sulla possibilità della contaminazione della ferita per aerosol, durante il recupero in elicottero della paziente, prima del ricovero in ospedale.

## REFERENCES

- [1] Mandell G.L., Bennet J.E., Dolin R. Principles and practice of infectious diseases (7<sup>th</sup> edition) 2010, vol 2, Chapter 245, 3097-3101. Churching Livingstone Elsevier.
- [2] Woodruff B.A., Griffin P.M., McCroskey L.M., et al. Clinical and laboratory comparison of botulism from toxin types A, B, and E in the United States, 1975-1988. *J. Inf. Dis.* 166, 1281-1286, 1992.
- [3] Davis J.B., Mattman L.H., Wiley M. *Clostridium botulinum* in a fatal wound infection. *J. Am. Med. Ass.* 146, 646-648, 1951.
- [4] Brett M., Hallas G., Mpmugo O. Wound botulism in the UK and Ireland. *J. Med. Microbiol.* 53, 555-561, 2004.
- [5] Palmateer N., Hope V., Roy K., et al. Infection with spore-forming bacteria in persons who inject drugs, 2000-2009. *Emerg. Infect. Dis.* 19, 29-33, 2013.
- [6] Reller M., Douche R., Maslanka S., Torres D.S., Manock S.R., Sobel J. Wound botulism acquired in Amazonian rain forest of Ecuador. *Am. J. Trop. Med. Hyg.* 74, 628-631, 2006.
- [7] Taylor S., Wolfe C., Dixon T., Ruch D.S., Cox G.M. Wound botulism complicating internal fixation of a complex radial fracture. *J. Clin. Microbiol.* 48, 650-653, 2010.
- [8] Dezfulian M., Bartlett J. Kinetics of growth and toxigenicity of *Clostridium botulinum* in experimental wound botulism. *Infect. Immun.* 452-454, 1985.
- [9] Oie S., Obayashi A., Yamasaki H., et al. Disinfection methods for spores of *Bacillus atrophaeus*, *B. anthracis*, *Clostridium tetani*, *C. botulinum* and *C. difficile*. *Biol. Pharm. Bull.* 34, 1325-1329, 2011.
- [10] Johnston M.D., Lawson S., Otter J.A. Evaluation of hydrogen peroxide vapour as a method for the decontamination of surfaces contaminated with *Clostridium botulinum* spores. *J. Microbiol. Methods* 60, 403-411, 2005.
- [11] Zoutman D., Shannon M., Mandel A. Effectiveness of a novel ozone-based system for the rapid high-level disinfection of health care spaces and surfaces. *Am. J. Infect. Control.* 39, 873-879, 2011.
- [12] Shin S.Y., Calvisi E.G., Beaman T.C., Pankratz H.S., Gerhardt P., Marquis R.E. Microscopic and Thermal Characterization of Hydrogen Peroxide Killing and Lysis of Spores and Protection by Transition Metal Ions, Chelators, and Antioxidant. *Appl. Environ. Microbiol.* 60, 3192-3197, 1994.