# Editorial

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**Biographical notes:** Joseph J. McArdle is a Professor at the Department of Pharmacology and Physiology, New Jersey Medical School, UMDNJ, NJ. He received his PhD from the State University of New York at Buffalo. He has several decades of experience in neuromuscular research and has authored more than a 100 papers.

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The anaerobic bacterium *Clostridium botulinum* produces 7 neurotoxins, A through G. Each botulinum neurotoxin (BoNT) consists of a heavy (Hc) and a light (Lc) chain joined by a single disulfide bond. The Hc enables holotoxin binding to, endocytosis into, and trafficking within motor nerve terminals. The pH of the endosome compartment favours reduction of the disulfide bond and allows release of the Lc. This metalloendoprotease cleaves SNARE proteins essential to the exocytosis of the neurotransmitter acetylcholine. The magnitude and duration of the resulting muscle paralysis depends on the substrate selectivity and intraterminal lifetime of the Lc metalloendoprotease.

Ease of production and extreme potency makes BoNTs potential bioweapons. Therefore, the Centers for Disease Control classifies BoNTs as Class A select agents. Conversely, clinical experience demonstrates that BoNTs are effective in the treatment of neurological diseases, pain, autonomic and cardiovascular disorders, as well as for cosmetic purposes. This growth of clinical applications increases the probability of toxic side effects to the BoNTs. Therefore, biodefense as well as clinical considerations indicate the need for therapies to prevent and reverse poisoning with BoNTs.

Early immunisation with antitoxin is currently the best therapy for BoNT poisoning. However, antitoxin administration can be delayed following exposure to a bioweapon containing BoNT. Furthermore, BoNT-administered to treat pathology can produce transient neuromuscular symptoms so that antitoxin is withheld to allow development of the desired therapeutic effect. Thus, both bioweapon and therapeutic use of BoNT may allow distribution of neurotoxin to compartments inaccessible to antitoxin. Prophylactic and therapeutic measures to complement antitoxin would be beneficial.

This issue of *The Botulinum Journal* presents ongoing work having practical implications for biodefense as well as the clinical use of BoNTs. Hale et al. introduce a mouse model for monitoring the movement and compartmentalisation of BoNTs *in vivo*. This model can be valuable for the development of protocols to reduce distribution and facilitate clearance of BoNTs. Lebeda and Adler analyse clinical and laboratory data relating to the time characteristics of BoNT action. These data, when modelled mathematically in terms of the hydrodynamic properties of BoNT/A, suggest that BoNTs may be slowly distributed from a protected storage reservoir within the body. Couesnon et al. present an electronmicroscopic comparison of BoNT entry into neuronal and intestinal cell lines. This comparison indicates that BoNT enters neuronal cells *via* clathrin coated pit-dependent endocytosis. In contrast, free molecules of BoNT appear to utilise a non-specific uptake process to transcytose the gut epithelium. Petro and Schengrund examined BoNT action on a neuronal cell line. Extraction of membrane cholesterol with methyl-Beta-cylodextrin to disrupt lipid rafts induced axonogenesis, elevated membrane ganglioside levels, and increased exo- and endocytosis. These events are proposed to increase uptake of BoNT. Potian et al. examined the effect of clathrin coated pits on BoNT uptake into and action on a cholinergic neuronal cell line as well as native motor nerve endings. Their work suggests that pharmacologic and physical treatments which disrupt clathrin coated pit formation act *in vivo* to protect against BoNT-induced muscle paralysis. Finally, Potian et al. present data showing that peptides that inhibit BoNT/A cleavage of SNAP-25 *in vitro* are not effective prophylactics or therapeutics of BoNT-induced muscle paralysis *in vivo*. This work addresses a disconnect between *in vivo* and *in vitro* studies and emphasises the need to study the action of metalloendoprotease inhibitors on primary cell targets. While the study of BoNT interactions with native cells is essential to the development of effective antidotes, discovery of homologous toxin binding sites, endocytitic pathways, intracellular trafficking, and substrate cleavage in non-neuronal cells may reveal additional therapeutic uses for the BoNTs.